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Cover Photo: From top to clock wise direction.
Inflorescence bearing multiple flowers in a cluster - *Rhododendron cowanianum* Davidian (PC: Pratikshya Chalise)
Vanda cristata Wall. ex Lindl. (PC: Sangram Karki)
Seedlings developed in half strength MS medium of *Dendrobium crepidatum* Lindl. & Paxton (PC: Prithivi Raj Gurung)
Pycnoporus cinnabarinus (Jacq.: Fr.) Karst. (PC: Rajendra Acharya)
Preparative HPLC (PC: Devi Prasad Bhandari)
Flower head of *Mimosa diplotricha* C. Wright (PC: Lila Nath Sharma)

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Editorial

It is our pleasure to bring out the current issue of Journal of Plant Resources, Volume 18, Number 1, a continuation of research publication by the Department of Plant Resources. Twenty seven peer reviewed articles based on original research have been incorporated in this issue. The articles have been categorized as Taxonomy, Ecology, Ethno-botany, Biotechnology, Microbiology, and Phytochemistry.

This issue intends to cover the research activities of the department as well as other research organizations. We encourage the young researchers to pursue quality research and contribute to build scientific knowledge on plant resources. We like to establish a link between the inference of scientific research and societies through dissemination of knowledge and information. We believe that the research findings will be helpful to the scientific community as well as general public to update the information on recent activities & development of plant science in Nepal.

We would like to thank all peer reviewers whose critical comments and suggestions helped to improve the quality of the journal. We acknowledge the contribution of the contributors for their interest in publishing their valued work in this journal and looking forward to further cooperation and collaboration with this department.

We apologize in advance for any mistakes in this issue and at the same time promise to improve the future issues based on your valued input.

J. Pl. Res. Vol. 18, No. 1, pp 1-5, 2020

Mimosa diplotricha (Fabaceae): A New Report of Invasive Weed from Eastern Tarai of Nepal

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Abstract

Mimosa diplotricha C. Wright (Fabaceae), an invasive weed is recorded for the first time in Nepal from South-eastern lowland of Tarai. A key to the Nepalese species of *Mimosa*, taxonomic description of *M. diplotricha*, notes on its habit, habitat, and distribution is provided. A variety *M. diplotricha* var. *inermis* (Adelb.) Veldkamlis also reported from the same area.

Introduction

Mimosa L. belongs to the sub family Caesalpinioideae in the family Fabaceae (The Legume Phylogeny Working Group [LPWG], 2017). The genus is one of the largest genera in the Fabaceae with more than 500 species mainly native to the New World (Barneby, 1991; Simon et al., 2011; Gehlot et al., 2013). The genus is much diverse in Americas (from United States to Argentina) with 496 species reported, less diverse in Madagascar with 34 species, and only few species are reported from east Africa and Asia (Simon et al., 2011). The genus is represented by only three species in China (Wu & Nielsen, 2010), eight species in India (Gamble, 1920; Debnath et al., 2017), and two in Nepal (Rajbhandari & Rai, 2019). The report of *M. diplotricha* from Eastern Tarai, hence, brings the total number of species of *Mimosa* in Nepal to three.

The genus *Mimosa* grows in wide range of habitats from lowland tropical regions to subtropical forests in different types of soil conditions. It thrives well in soils with low level of nutrients and organic matter with low pH (Gehlot et al., 2013). Because of its ability to grow in different soil conditions, few species have become the pantropical weeds, and *M. diplotricha* is one of them. Other well-known invasive weeds are *M. pigra* L. and *M. pudica* L., *M. pigra* is among 100 of theworld's worst invasive alien species (Lowe et al., 2000). *M. pigra* has not been reported from Nepal yet but *M. pudica* is among the 26 most problematic invasive weeds of Nepal (Tiwari et al. 2005; Shrestha, 2019).

The genus is usually characterized by the armed stem, pinnate and often sensitive leaves, globose inflorescence arising from the axils of leaves, and compressedflat pods divided transversely into one-seeded segments.

The specimens of *Mimosa* were collected and photographed during the field work in Eastern Tarai from August to November 2019. The specimens were identified as *M. diplotricha* and reported here for the first time of its presence in Nepal. A key to distinguish it from other species of *Mimosa* in Nepal is given below. Moreover, a variety *M. diplotricha* var. *inermis* (Adelb.) Veldkamlis also recorded.

Materials and Methods

This study is based on the field studies of populations of *M. diplotricha* in eastern Nepal in Jhapa and Morang districts (Figure 1), and herbarium studies of specimens (including the types)deposited at National Herbarium and Plant Laboratories (KATH), Tribhuvan University Central Herbarium (TUCH), Royal Botanic Garden Edinburgh (E), and Royal Botanic Gardens Kew (K) (online images). The specimens were checked against the relevant floras and checklists (Gamble, 1920; Ohashi, 1979; Gierson & Long, 1987; Press et al., 2000; Nielsen & Wu, 2010; Rajbhandari & Rai, 2019) to confirm its identification.The photographs were studied and

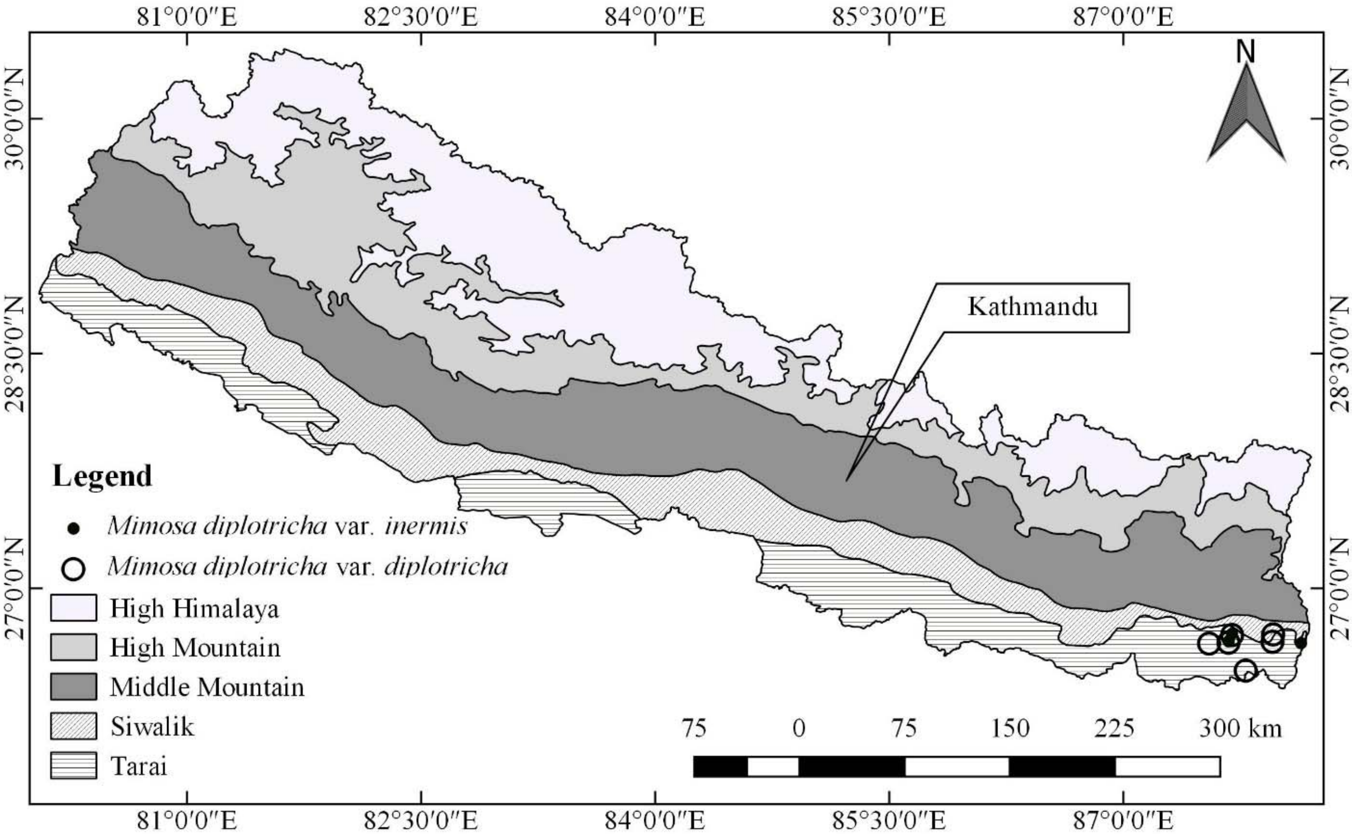

Figure 1: Physiographic map of Nepal showing collection locations

the identification was further confirmed by the expert of invasion ecology (pers.comm. K.V. Sankaran, Kerala Forest Research Institute, India; 4 December 2019). All the specimens collected during the field work were deposited at TUCH and KATH herbarium.

Taxonomic treatment

Key to the species of *Mimosa* of Nepal

1a. Leaves pinnae 2-4 (1 -2 pairs) .... *M. pudica*
1b. Leaves pinnae 12-20 (6-10 pairs) 2
2a. Pods 7-11 cm long .... *M. himalayana*
2b. Pods 1.5-3 cm long .... *M. diplotricha*

Mimosa diplotricha C. Wright, Anales Acad. Ci. Med. Habana 5: 405. 1868. Nepali name: Aarakande (आराकाँडे), Thulolajjawatijhar (ठूलो लज्जावती झार)

English name: creeping sensitive plant, giant false sensitive plant; giant sensitive plant (Centre for Agriculture and Bioscience International [CABI], 2019)

Annual or perennial gregarious subshrub. Stem scrambling or prostrate, ascending stems up to 5 mlong, profusely branched, 4-angled, hirsute, with or without prickles. Leaves bipinnate, 10–18 cm long, stipulate, stipules linear upto 4 mm, hirsute. Petiole 3-7 cm, with or without prickles; pinnae 6-8 pairs, 3–5 cm. Leaflets 20–28 pairs per pinnae, linear-oblong, 4–7 × 2–3 mm, base obtuse, margin entire, apex mucronate, white villous both sides. Inflorescence, axillary, solitary or in pairs, heads globoseup to 1.5 cm in diameter; peduncles 0.5–1.0 cm, hirsute. Flowers sessile, bisexual. Calyx small, ca. 3 mm. Corolla narrowly infundibuliform, ca. 2 mm, 4-lobed, outside slightly pubescent. Stamens 8, filamentsup to8mm long, unequal, hairy, anthersca. 2 mm. Pods in clusters up to 30, slightly curved, 1.5–3 × 0.4–0.6 cm, 2-5 seeded, compressed, hirsute, suture with bristles up to 3 mm. Seeds yellow-brown, upto 3 mm.

Flowering: October-November

Fruiting-November-January

Habitat: Grows in flood plains, riverbanks, roadsides, abandoned fields and forest fringes

Distribution: It is native of neotropics and Caribbean, invasive in wet tropics and subtropics including south and south EastAsia, Africa and Pacific islands (Sankaran & Suresh 2013).

Key to the varieties

1a. Stems with downward facing prickles .... *M. diplotricha* var. *diplotricha*

1b. Stems without prickles .... *M. diplotricha* var. *inermis*

Mimosa diplotricha var.***diplotricha*** (Figure 2).

Synonym: *Mimosa invisa* Mart.

Specimens examined:

Eastern Nepal. Jhapa District: Arjundhara-3, Kaidale, 26.714018° N, 87.960224°E, 159 m, 7 Nov 2019, M.R. Bist & L.N. Sharma J03 (TUCH, KATH); Arjundhara-4, Bering khola, 26.667144°N, 87.955622° E, 122 m,7 Nov 2019, M.R. Bist & L.N. Sharma J04 (TUCH, KATH); Gauriganj-1, Punjibari, 26.483205° N, 87.784154°E, 66 m, 8 Nov 2019, M.R. Bist & L.N. Sharma J05 (TUCH, KATH); Damak-9, 26.661229°N, 87.673891° E, 116 m, 8 Nov 2019, M.R. Bist & L.N. Sharma J06 (TUCH, KATH); Damak-2, Beldangi, 26.710273° N, 87.696780° E, 146 m, 8 Nov 2019, M.R. Bist & L.N. Sharma J09 (TUCH, KATH). Morang District: Pathari-10, Bhutanese refugee camp, 26.655846° N, 87.549441° E, 126m,8 Nov 2019, M.R. Bist & L.N. Sharma J08 (TUCH, KATH).

Mimosa diplotricha var. ***inermis*** (Adelb.) Veldkamp, Fl. Males. Bull. 9(4): 416 (1987) (Figure 3).

Figure 2: *Mimosa diplotrcha* var. *diplotricha*. a. Dense mat of the species in open habitat, b. Stem showing inflorescence, c. Flower head close up, d. Pods in clusture, e. Single pod

Figure 3: *Mimosa diplotricha* var. *inermis*. a. Dense mat of the species in open habitat, b. Stem showing fruits, c. Stem showing inflorescence, d. Pods in a clusture

Specimens examined:

Eastern Nepal. Jhapa District: Mechi-7, new bus park Kakadvitta, 26.649609°N, 88.138704° E, 109 m, 6 Nov 2019, M.R. Bist & L.N. Sharma J02 (TUCH, KATH); Damak-9, 26.661229° N, 87.673891° E, 116 m, 8 Nov 2019, M.R. Bist & L.N. Sharma J07 (TUCH, KATH); Damak-2, Beldangi, 26.710273° N, 87.696780° E, 146 m, 8 Nov 2019, M.R. Bist & L.N. Sharma J10 (TUCH, KATH).

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References

Barneby, R.C. (1991). Sensitivae Censitae:a description of the genus Mimosa Linnaeus (Mimosaceae) in the New World. *Memoirs of the New York Botanical Garden, 65,* 1–835.

Centre for Agriculture and Bioscience International. (2019). *Mimosa diplotricha* Sauvalle. In: *Invasive Species Compendium*. Wallingford, UK: CAB International. www.cabi.org/isc.

Debnath, A., Paul C. & Debnath B. (2017). Two invasive alien giant sensitive weeds-new additions to the flora of Tripura, India. *Bioscience Discovery*, *8*(4), 720–725.

Gamble, J.S. (1920). The Indian Species of *Mimosa*. *Bulletin of Miscellaneous Information (Royal Botanic Gardens, Kew),1*, 1–6.

Gehlot, H.S., Tak, N., Kaushik, M., Mitra, S., Chen, W.M., Poweleit, N., Panwar, D., Poonar, N., Parihar, R., Tak, A., Sankhla, I.S., Ojha, A., Rao, S.R., Simon, M.F., Junior, Fabio Bueno dos Reis, Perigolo, N., Tripathi, A.K., Sprent, J.I., Peter J.W.Y., James, E.K. & Gyaneshwar, P. (2013) An invasive *Mimosa* in India does not adopt the symbionts of its native relatives. *Annals of Botany, 112* (1), 179–196. doi.org/10.1093/aob/mct112.

Gierson, A.J.C., & Long, D.G. (1987). Leguminosae. In: Gierson, A.J.C., & Long, D.G. (Eds.) *Flora of Bhutan* (pp. 607–738), Vol.1, part 3, UK: Royal Botanic Garden, Edinburgh.

Lowe, S., Browne, M., Boudjelas, S., & De Poorter, M. (2000). *100 of the World's Worst Invasive Alien Species: A Selection from the Global Invasive Species Database* (Volume 12). Auckland, New Zealand: World Conservation Union (IUCN).

The Legume Phylogeny Working Group. (2017). A new subfamily classification of the Leguminosae based on a taxonomically comprehensive phylogeny. *Taxon, 66* (1), 44–77.

Ohashi, H. (1979). Leguminosae. In: Hara, H. & Williams, L.H.J. (Eds.), *An Enumeration of the Flowering Plants of Nepal* (pp. 103–133), Vol. 2. London: British Museum (Natural History).

Press, J.R., Shrestha, K.K. & Sutton, D. A. (2000). *Annotated Checklist of the Flowering Plants of Nepal*. London: Natural History Museum.

Rajbhandari, K.R. & Rai, S.K. (2019). *A Handbook of Flowering Plants of Nepal,* Vol.2. Kathmandu: Department of Plant Resources.

Sankaran, K.V. & Suresh, T.A.(2013). *Invasive Alien Plants in the Forests of Asia and the Pacific*. Bangkok: Food and Agriculture Organization of the United Nations, Regional Office for Asia and the Pacific.

Shrestha, B.B. (2019). Management of invasive alien plant species in Nepal: current practices and future prospects. In: Garkoti, S.C., van Bloem, S., Fule, P.Z., & Semwal, R.L. (Eds.), *Tropical Ecosystems: Structure, Functions and Global Change*. (pp. 45–68). Singapore: Springer Nature.

Simon, M.F., Grether, R., de Queiroz, L.P., Sarkinen, T.E., Dutra, V.F. & Hughes, C.E. (2011). The evolutionary history of Mimosa (Leguminosae): toward a phylogeny of the sensitive plants. *American Journal of Botany*, *98*(7), 1201–1221

Tiwari, S., Siwakoti, M. Adhikari, B. & Subedi, K. (2005). *An Inventory and Assessment of Invasive Alien Plant Species of Nepal*, Nepal: IUCN - The World Conservation Union.

Wu, D. & Nielsen, I.C. (2010). Mimosa. In: Wu, Z.Y., Raven, P.H. & Hong, D.Y. (Eds.), *Flora of China* (pp. 53–54), Vol.10. Beijing: Science Press. & St Louis: Missouri Botanical Garden Press.

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Some Freshwater Green Algae of Raja-Rani Wetland, Letang, Morang: New for Nepal

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Abstract

Freshwater green alga of Raja-Rani wetland has been studied. A total 36 algal samples were collected from 12 sites by squeezing the submerged aquatic plants. Present paper describes 35 green algae under 18 genera from Raja-Rani wetland as new record for Nepal.Genus *Euastrum* consists 5 species; genera *Cosmarium*, *Staurodesmus*, and *Staurastrum* consist 4 species each; genera *Scenedesmus*, *Closterium*, *Pleurotaeniuum* and *Xanthidium* consist 2 species each; and rest genera consist only single taxa each. Water parameters of the wetland of winter, summer and rainy seasons were also recorded.

Keywords: Chlorophyceae, *Cosmarium*, New report, *Staurodesmus*, *Triploceras,Xanthidium*

Introduction

Algae are the simplest photosynthetic thalloid plants, usually inhabited in water and moist environment throughout the world. Green algae are the largest and most diverse group of algae, with about 8000 species known (Guiry, 2012). They have wide range of habitats as they grow in freshwater, marine, subaerial, terrestrial, epiphytic, endophytic, parasitic, symbiotic, epizoic and endozoic conditions. In freshwater environment, green algae preferably grow in stagnant water like ponds, pools, puddles, lakes etc. Their size also ranges from microscopic unicellular, colonial to extensively large filamentous forms. Their characteristic fresh green colouration is due to the presence of chlorophyll a and b which are not masked by other accessory pigments. Green algae are ecologically important as they are the major producers in the freshwater ecosystem.

Numerous taxonomic studies on algae have been carried out in the world. Contributions made by Turner (1892), West & West (1902), Bruehl & Biswas (1926), Geitler (1932), Fritsch & Rich (1937), Scott & Prescott (1960), Komarek (1983), Kouwets (1987), Gandhi (1999), Stastny (2010), Matthews (2016) etc. for the algal flora of different parts of the world are acknowledgeabe. In the context of Nepal, even though our country has diverse climatic condition and rich aquatic habitats for algae, extensive exploration is lacking in the history.Suxena & Venkateswarlu (1968), Hickel (1973), Joshi (1979), Subba Raju & Suxena (1979), Shrestha & Manandhar (1983), Hirano (1984), Ishida (1986), Watanabe & Komarek (1988), Haga & Legahri (1993), Watanabe (1995), Baral (1996, 1999), Das & Verma (1996), Prasad (1996), Komarek & Watanabe (1998), Simkhada et al. (2006), Rai & Misra (2010), Jüttner et al. (2011), Jha & Kargupta (2012),Krstiæ et al. (2013), Necchi et al. (2016), Shrestha & Rai (2017), Rajopadhyaya et al. (2017), Rajopadhyaya & Rai (2016-18), Rai & Khadka (2017), Rai & Rai (2018), Rai & Poudel (2019),etc. have collected many algae from different places at different times in Nepal. Most of the exploratory works on algae were carried out in and around Kathmandu Valley and in the Eastern and Central mountain regions of Nepal. At present, total algae of Nepal is 998 including 29 endemic species (Rai & Ghimire, 2020).

Especially on chlorophycean algae of Nepal, contributions have been made by the following authors: chlorophyceae (Prasad et al. 1985, Sahay et al. 1992, Rai 2009), desmids (Förster 1965, Watanabe 1982- *Closterium*; Bando et al. 1989; Habib & Chaturvedi 1995, 1997; Ichimura 1997-

Closterium ehrenberghii; Rai & Misra 2008; Rai et al. 2008; Rai 2014), and chlorococcales (Nakano & Watanabe 1988; Chaturvedi& Habib 1996; Nozaki 1998, 1990- colonial volvovales; Nozaki & Kuroiwa 1990, 1991- *Volvulina compacta* and *Gonium multicoccum* respectively; Rai & Misra 2012- *Pediastrum*; Rai 2013- *Scenedesmus*), filamentous green algae (Muller 1965- *Spirogyra nepalensis*; Dwivedi 1985- *Chaetophora*; Haga 1988; Akiyama 1989- *Trentepohlia*; Kargupta & Jha 1997- *Stigeoclonium*; Rai & Misra 2007- *Spirogyra* and *Sirogonium*; Rai 2012- *Oedogonium*). Chlorophycean algae of Raja-Rani wetland has been published earlier by Godar & Rai (2018). From the same collection, only the new reports for Nepal are being described in this paper.

Materials and Methods

Study area

Raja-Rani (Dhimal Pokhari) wetland lies in 26°44.9'22"N and 87°28.9'10"E, at an altitude 470 m asl in Letang Muncipality 1, Morang district, Nepal (Figure 1 and 2). It is located north-west from Letang Bazaar after crossing the Chisang River, at Dhimal Danda. It is one of the important wetland of Nepal situated on the Chure hill range. There are three ponds namely -Raja, Rani and Chhori (Rajkumari), located inside the southern part of the Sal forest. All these ponds are stretched along north-south direction. The largest one is Raja Pokhari (King's pond) that lies in the west with water depth about 7 m in rainy, 3 m in winter and 2 m in summer season. East one is Rani Pokhari (Queen's pond) which has siltation problem, so its depth is decreasing. Though, its depth is 7 m in rainy, 3 m in winter and 2 m in summer season.The degraded pond of the north-east corner is called Chhori Pokhari (Rajkumari pond) which has minimum water depth as 2 m in rainy, one half meter in winter and it is completely dried in the summer season.

The wetland basin extends about 133 ha where as three ponds cover only about 20 ha of land surrounding by mixed forest (Figure 2). The Raja-Rani Community Forest covers about 1700 ha. The wetland system has no permanent source of water as this is being fed by ground source "Jaruwa", seasonal forest fed streams and direct precipitation. This wetland has been managed by Raja-Rani Community Forest Users Group. The local people formed a Raja-Rani Pokhari Conservation Committee particularly for managing this wetland. The wetland area is rich in biodiversity (Ministry of Forests and Soil Conservation [MoFSC] 2012).

The aquatic vegetation in the pondas floating species were *Eichhornia cracipes*, *Pistia stratiotes*, *Spirodella polyrhiza, Azolla imbricata, Nymphaea nouchali, Nymphoides hydrophylla, N. indica, Nelumbo nucifera*; as submergedspecies were *Ceratophyllum demersum*, *Ottelia alismoides*, *Hydrilla verticillata* etc. Other wetland species were Pirre Jhar (*Polygonum hydropiper*), Narkat (*Phragmites kharka*), *Cyperus esculentus*, *C. pilosus*, *Floscopa scandens*, *Hygrophila polysperma, H. quadrivalvis, Panicum psilopodium*, *Persicaria hydropiper*, *Rotala rotundifolia*, *Saciolepis indica* etc. The surrounding terrestrial vegetation is mixed type forest dominated by Sal (*Shorea robusta*), Rajbrikshya (*Cassia fistulosa*), Jamun (*Syzygium cuminii*), Odane (*Sterculia villosa*), Chatiwan (*Alstonia scholaris*) etc.

The climate of the Churia region ranges from subtropical to warm temperature characterized by hot and sub-humid summers, intense monsoon rain, and cold dry winters. The average annual minimum temperature ranges from 12°-19°C, with the average annual maximum temperature ranges from 22°-30°C in this region. The total annual rainfall varies from minimum of 1,138 mm to the maximum of 2,671 mm throughout the Churia (Department of Forest Research and Survey [DFRS] 2014).

Algae collection and identification

Algae were collected three timesat an interval of 3 months i.e., in Falgun (winter season, sample no. W1 to W12), Jestha (summer season, sample no. S13 to S24) and Bhadra (rainy season, sample no. R25 to R36) months of 2072/73BS (Table 1).Total 36 samples were collected from 12 sites i.e., 9 from Raja pondand 3from Rani pond, more or less at an

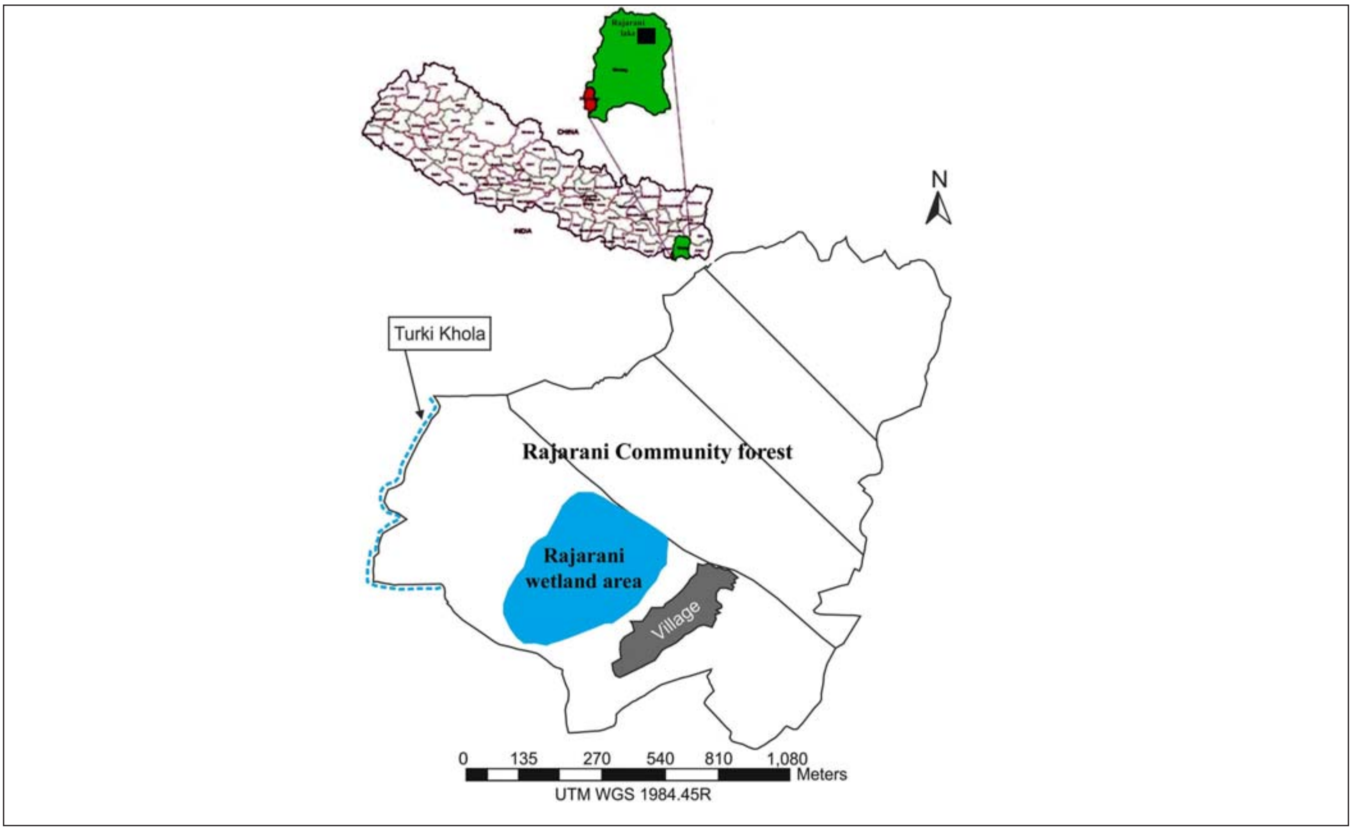

Figure1: Map of Raja-Rani (Dhimal Pokhari) wetland, Bhogateni, Letang Municipality, Morang, Nepal.

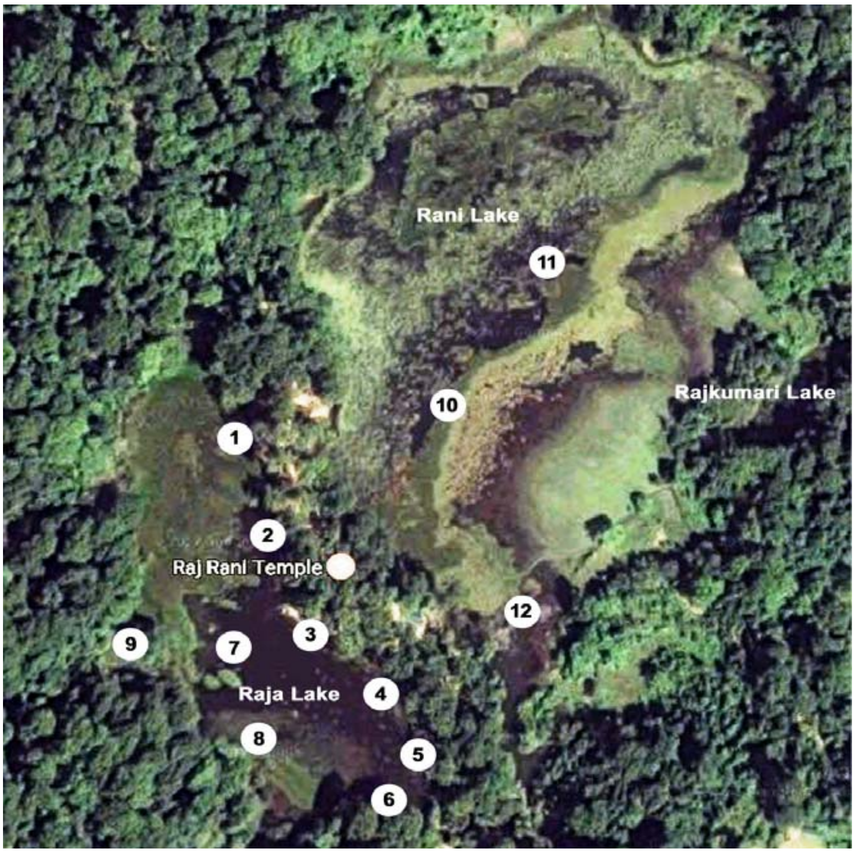

Figure 2: Raj, Rani and Rajkumari ponds showing algal sampling sites (1 to 12).

equal distance from the peripheral littoral zone of the pond (Fig. 2). Collection was done by squeezing submerged parts of aquatic macrophytes (*Eichhornia crassipes* (Mart.) Solms, *Ceratophyllum demersum* L., *Ottelia alismoides* (L.) Pers., *Hydrilla verticillata* (L.f.) Royle, *Pistia stratiotes* L., etc.), between 10 am to 2 pm in the day. The samples were preserved in 4% formaldehyde solution in airtight polythene bottles thentagged and labeled properly indicating sample no., sampling site, date of collection etc. using permanent marker. Field note was also maintained accordingly.

Field observations were also made to record the conditions of wetland and algal habitat in every site. The exact time of sample collection and other surrounding aquatic and land vegetations of the wetland were also noted in the field diary. The history of Raja-Rani wetland was recorded by interviewing the staff of pond management team and the villagers.The geographical position *viz*., latitude, longitude and altitude of each site were measured with the help of Garmin e-trex GPS meter. The water temperature and pH of each site were measured with the help of digital thermometer and portable Hanna pH meter, respectively. For the analysis of other water parameters in the laboratory, water samples were taken in airtight black bottles. Finally, photographs of collection sites were also taken with the help of Canon Digital camera.

Samples were brought to the Phycology Research Lab, Department of Botany, Post Graduate Campus, Biratnagar. All the samples were screened for green algae under the light microscope and only the selected samples were further investigated for taxonomic purpose. For the confirmation of green and blue-green algae, iodine solution and 1% aqueous methylene blue stains were used. Number of algae and their dominancy were observed and calculated. Algal dimension (length, breadth etc.) was measured with the help of calibrated ocular micrometer. Photomicrography was done for each taxa under 40X and 100X objectives using Olympus Ch20i microscope attached with camera. Free-hand camera lucida drawings werealso made forsome large sized algae during identification. Algae were identified following Prescott (1951), Scott & Prescott (1961), Philipose (1967), Nurul Islam (1970), Croasdale & Flint (1986), Kouwets (1987), Hirano (1992), Prasad & Misra (1992), Croasdale et al. (1994) etc.by cross-checking with illustrations and description given. Classification and current accepted name of algae were confirmed by visiting different online sites *viz*., www.algaebase.org; http://www.digicodes.info/index.html; http://

Table1: Description of algal sampling in Raja-Rani wetland

Sample no.	Date of collection	Sampling sites	Latitude (N)	Longitude (E)	Altitude (m)
W1	2072/11/15	1	26° 44' 954"	87° 28' 866"	409
W2	,,	2	26° 44' 941"	87° 28' 867"	454
W9	,,	9	26° 44' 942"	87° 28' 902"	463
W10	,,	10	26° 44' 930"	87° 28' 922"	457
W11	,,	11	26° 44' 938"	87° 28' 934"	461
W12	,,	12	26° 44' 970"	87° 28' 882"	456
S13	2073/02/15	1	26° 44' 954"	87° 28' 866"	409
S14	,,	2	26° 44' 941"	87° 28' 867"	454
S15	,,	3	26° 44' 936"	87° 28' 876"	465
S18	,,	8	26° 44' 937"	87° 28' 878"	466
S19	,,	7	26° 44' 873"	87° 28' 902"	460
S20	,,	8	26° 44' 937"	87° 28' 878"	466
S21	,,	9	26° 44' 942"	87° 28' 902"	463
S22	,,	10	26° 44' 930"	87° 28' 922"	457
S23	,,	11	26° 44' 938"	87° 28' 934"	461
S24	,,	12	26° 44' 970"	87° 28' 882"	456
R34	2073/05/15	10	26° 44' 930"	87° 28' 922"	457
R35	,,	11	26° 44' 938"	87° 28' 934"	461

Note: W = Winter, S = Summer, R = Rainy seasons

Table2: Water analysis of Raja-Rani wetland in different seasons

Sampling seasons	Water parameters								
	Temp (ºC)	pH	DO (mg/l)	Conductivi-ty (µS/cm)	Turbidity (NTU)	Total Dissolved Solids (mg/l)	Total Kjeldahl Nitrogen (mg/l)	Phosphorus (mg/l)	Potassium (mg/l)
Winter	20	6.92	-	55.0	8.34	27.5	2.8	0.06	0.34
Summer	24	6.50	-	112.3	51.00	561.15	2.7	0.24	4.27
Rainy	23	7.15	4.22	47.2	73.00	23.60	4.2	0.24	-

Table 3: Algae new to Nepal, reported from Raja-Rani wetland, Letang, Morang

1	*Pediastrum angulosum* var. *laevigatum*	19	*Euastrum subhypochondrum*
2	*Tetraedron regulare*	20	*Micrasterias rotata* var. *curvata*
3	*Coelastrum astroideum*	21	*Cosmarium auriculatum*
4	*Scenedesmus graevenitzii* var. *alternans*	22	*Cosmarium rectangulare*
5	*Scenedesmus tropicus*	23	*Cosmarium regnesi*
6	*Selenastrum westii*	24	*Cosmarium taxichondrum* var. *undulatum*
7	*Coleochaete pulvinata*	25	*Xanthidium* cf *aculeatum*
8	*Oedogonium reinschii*	26	*Xanthidium burkillii* var. *alternans*
9	*Gonatozygon aculeatum*	27	*Staurodesmus brevispina*
10	*Closterium archerianum*	28	*Staurodesmus cuspidatus* var. *curvatus*
11	*Closterium setaceum* var. *elongatum*	29	*Staurodesmus dickiei* var. *rhomboideus*
12	*Pleurotaenium coroniferum*	30	*Staurodesmus mamillatus* var. *mamillatus*
13	*Pleurotaenium maculatum*	31	*Staurastrum contectum*
14	*Triploceras gracile*	32	*Staurastrum freemanii* var. *nudiceps*
15	*Euastrum ansatum* var. *rhomboidale*	33	*Staurastrum longispinum*
16	*Euastrum denticulatum* var. *quadrifarium*	34	*Staurastrum unicorne* var. *ecorne* f. *retusum*
17	*Euastrum pulchellum*	35	*Teilingia excavata*
18	*Euastrum sinuosum* var. *germanicum*		

desmids.science4all.nletc. All the collected samples have been deposited in the Phycology Research Lab, Department of Botany, P.G. Campus Biratnagar.

Results and Discussion

Water analysis

In summer, water temperature, conductivity and total dissolved solids were found distinctly high but water pH and Total Kjeldahl nitrogen were low in comparison to other seasons. Similarly, in rainy season, water pH, turbidity and total Kjeldahl nitrogen were high but conductivity and total dissolved solids were low. In winter, water temperature, turbidity, and phosphorus were distinctly low (Table 2).

Taxonomic description

A total of 35 green algae under 18 genera are reported as new for Nepal from Raja-Rani wetland (Table 3). These are confirmed as new to Nepal after detail investigation and cross checking with relevant literatures published till the date.

The taxonomy of these algae are described one by one below. Each taxa is appended with author/s name and location of figure (in parenthesis) followed by source of identification, morphological characters, dimension and sample number. Classification of algae in the present work is based on Guiry & Guiry (2019).

Family: Hydrodictyaceae
Genus: ***Pediastrum*** Meyen (1829)

1. ***Pediastrum angulosum*** var. ***laevigatum*** Raciborski (Fig. 3: 3)
   Philipose 1967, P. 118, Fig. 39.
   Colonies 8-16-32-64 celled, compact without perforations, usually single layered and round, elliptical or kidney-shaped, sometimes large and two-layered with small irregular perforations; interior cells transversely elongated, irregularly hexagonal; marginal cells also transversely

elongated, wider above, truncate at the base; outer side deeply emarginated and with slightly converging lobes having obtuse or rounded ends; colonies up to 150 µm in diameter; cells up to 35 µm in diameter.
Sample no. W11

Genus: ***Tetraedron*** Kützing (1845)

2. ***Tetraedron regulare*** Kütz. [New name: ***Tetraëdriella regularis*** (Kützing) Fott] (Fig. 3: 4-6)
Philipose 1967, p.146, fig. 60e
Cells tetragonal, pyramidal, with concave or straight sides; angles with a blunt, stout, slightly curved and acuminate spines; cell wall smooth; cells 16-34 µm long; spines 3.5-4.5 µm long.
Sample no. S13

Family: Scenedesmaceae
Genus: ***Coelastrum*** NÓgeli (1849)

3. ***Coelastrum astroideum*** De Notaris (Fig. 3:7)
Olenina et al. 2006, P. 125.
Coenobium spherical, of 8, 16 cells; cells ovoid, with thickening in the apical part, connected to adjacent ones by the basal part, but without connecting processes between the cells; chloroplast single, parietal, laminate, with one pyrenoid; cells 6-10 µm in diameter.
Sample no. S21

Genus: ***Scenedesmus*** Meyen (1829)

4. ***Scenedesmus graevenitzii*** C. Bernard var. ***alternans*** (Fig. 3:8)
Philipose1967, P. 254, Fig. 164 g.
Colonies flat, usually 8 celled, sometimes 4 celled with cells arranged in a distinctly alternating series; adjacent cells adnate to each other along a short portion of their length only; cells ellipsoid to ovoid-ellipsoid with rounded ends, about twice longer than broad; cells 13-16 µm long.
Sample no.S18

5. ***Scenedesmus tropicus*** W.B. Crow [Current accepted name: *Desmodesmus tropicus*(W.B. Crow) E. Hegewald] (Fig. 3: 9-10)
Philipose 1967, P. 279, Fig. 185.
Colony four-celled and subquadrate; cells more or less biconvex in the middle, attenuated towards the ends and with inflated poles; adjacent cells connected to each other by two narrow processes leaving a linear intercellular perforation; poles of terminal cells provided with a long recurved spine; chloroplast parietal and with a single pyrenoid; colony 28-38 µm long, 26-33 µm broad; cells 26-33 µm long, 7-10 µm broad.
Sample no. W9

Family: Selenastraceae
Genus: ***Selenastrum*** Reinsch (1867)

6. ***Selenastrum westii*** G.M. Smith [Current accepted name: *Ankistrodesmus gracilis* (Reinsch) Korshikov] (Fig. 3:11-12)
Smith 1920, P. 133, Pl. 31, Figs. 8-10; Philipose 1967, P. 221. Fig. 129; Prescott 1951, P. 257, Pl. 57, Fig. 10.
Colonies of 2-4-8 irregularly arranged cells with their convex sides in contact, rarely free; cells lunate to arcuate but not sickle shaped, and with acuminate apices; chloroplast without a pyrenoid; cells 15-39 µm long, 1.5-3 µm broad.
Sample no.R35

Family: Coleochaetaceae
Genus: ***Coleochaete*** Brébisson (1844)

7. ***Coleochaete pulvinata*** A. Braun (Fig. 3: 16)
Prescott 1951, P. 129, Pl. 18, Figs. 7, 8.
Thallus with irregularly branched filaments radiating from common center; cells oblong or pyriform, larger at anterior end; oogonia and antheridia not found; cells 25-40 µm in diameter.
Sample no.S13

Family: Oedogoniaceae
Genus: ***Oedogonium*** Link ex Hirn (1900)

8. ***Oedogonium reinschii*** Roy ex Hirn (Fig. 4: 1)
Prescott 1951, P. 193, Pl. 34, Figs. 1-3; Tiffany & Britton 1952, P. 100, Pl. 18, Fig. 149.
Vegetative cells subellipsoid to hexagonal or fusiform, especially the latter shape in the lower portions of the filament; 7.5-8 µm in diameter, 8.5-19.2 µm long. Oogonia 1-2; pyriform-

globose; operculate; division median; 19-20 µm in diameter, 15-21 µm long. Oospores depressed globose; not filling the oogonia longitudinally; wall smooth; 15-18 µm in diameter, 14-15 µm long.
Sample no. S21

Family: Gonatozygaceae
Genus: ***Gonatozygon*** De Bary (1858)

9. ***Gonatozygon aculeatum*** Hast. (Fig. 4:2-3)
Scott & Prescott 1961, P. 8, Pl. 1, Fig. 7; Croasdale & Flint 1986, P. 40, Pl. 2, Figs. 1, 2.
Cells cylindrical, slightly dilated, club shaped and truncate; cell wall bearing dense tapered spines; chloroplast band shaped, rarely stellate with few ridges; cells 125-270 µm long, 10-16 µm broad.
Sample no. S23

Family: Closteriaceae
Genus: ***Closterium*** Nitzsch ex Ralfs (1848)

10. ***Closterium archerianum*** Cl. ex Lund. (Fig. 4: 4-5)
Croasdale & Flint 1986, P. 53, Pl. 12, Figs. 12-14.
Cells strongly and evenly curved, tapered to the narrow apices which are rounded or somewhat obliquely truncate, outer margin with 106-150 degree arc; wall often brownish, clearly striate; zygospore globose; cells 163-270 µm long, 16-30 µm broad; apices 3.5-4 µm broad; striae 5-8 in l0 µm.
Sample no.S14

11. ***Closterium setaceum*** Ehr. var. ***elongatum*** West et G.S. West (Fig. 4:6-8)
West & West 1905, P. 499, Pl. 6, Fig. 2; Krieger 1937, Pl. 33, Fig. 11.
Cells with short, fusiform portion tapering abruptly into long slender cylindrical processes (horns) which are longer than the swollen median portion and nearly straight except at their incurved ends; median portion only with chloroplast and striated wall, rarely apparently smooth; cells 300- 800 µm long, 6-16 µm broad
Sample no.S23

Family: Desmidiaceae
Genus: ***Pleurotaenium*** NÓgeli (1849)

12. ***Pleurotaenium coroniferum*** (Borge) Krieg. (Fig. 4:9-10)
Nurul Islam 1970, P. 912, Pl. 5, Fig. 16; Hirano 1992, P. 15, P l. 8, Fig. 6.
Cells of medium size, 11 times longer than breadth, slightly attenuated toward the end, lateral margin undulated; apex truncate and with a series of conical granules; cells 233 µm long, 21 µm broad, apex 19.5 µm wide.
Sample no.S19

13. ***Pleurotaenium maculatum*** (Turner) Carter (Fig. 4:11-12)
Hirano 1992, P. 16; Opute 2000, P. 140, Fig. 5: 8, 9
Cells large and robust, very long, about 16 times longer than broad; semicells of even diameter, with one distinct basal swelling above the isthmus and terminating in an expanded apex ornamented by a ring of 13-16 prominent conical tubercules; cells 686-756 µm long, 40-55 µm broad; isthmus 45 µm wide.
Sample no. S19

Genus: ***Triploceras*** J.W. Bailey (1851)

14. ***Triploceras gracile*** Bail. (Fig. 4:13-15)
Nurul Islam 1970, P. 915, Pl. 2, Fig. 3; Pl. 5, Fig. 9; Nurul Islam & Yusuf Haroon 1980, P. 556, Pl. 4, Figs. 52-53; Croasdale & Flint 1986, P. 77, Pl. 15, Figs. 1-3, 8.
Cells elongated, cylindrical; semicells slightly tapering towards the apex which has 2 short, slightly spreading lobes or processes, each of which bears 2 spines with 9-15 whorls of low elevation each bearing a stout, broad based spine, the lower spine extending horizontally, the spine above upwardly directed; cells 206-668 µm long, 21-53 µm broad, apex 24-40 µm wide.
Sample no. W12

Genus: ***Euastrum*** Ehrenberg ex Ralfs (1848)

15. ***Euastrum ansatum*** Ehr. ex Ralfs var. ***rhomboidale*** Ducell (Fig. 4: 16)
http://www.digicodes.info/Euastrum_ansatum_var_rhomboidale.html#2009015007; Stastny

2009, P. 144. Pl. 9, Fig. 1.
Cell twice as long as broad, deeply constricted, sinus narrowly linear; semi-cells pyramidate with rounded basal angles, lower part of the side convex, upper part slightly concave, apex rotundo-truncate, deep incision; cells 85-90 µm long, 39-42 µm broad; isthmus 14 µm wide.
Sample no.R35

16. ***Euastrum denticulatum*** var. ***quadrifarium*** Krieg. (Fig. 4: 17)
Scott & Prescott 1961, P. 25, Pl. 13, Fig. 11; Croasdale & Flint 1986, P. 89, Pl. 22, Fig. 15.
Cells small, face of semi-cells showing a central protuberance, bearing a circle of four curved granules which form a square, two pores above them; cells 23-28 µm long, 15-24 µm broad; isthmus 5-6 µm wide.
Sample no.W12

17. ***Euastrum pulchellum*** Brébisson (Fig. 5:1-2)
West & West 1905, P. 46, Pl. 38, Figs. 14, 15.
Cells small, deeply constricted, sinus narrowly linear with slightly dilated extremity; semicells 3 lobed with shallow sinus between the lobes; polar lobe widely rectangular oblong, apex truncate with deep narrow median incision, angles with short diverging spines; lateral lobes rounded, with 6-8 acute granules; cells 37-40 µm long, 28-33 µm broad; isthmus 7.5µm wide.
Sample no. S23

18. ***Euastrum sinuosum*** var. ***germanicum*** (Racib.) Krieg. (Fig. 5:3)
Croasdale & Flint 1986, P. 98, Pl. 18, Fig. 10.
Cells longer than broad, deeply constricted; sinus narrow and linear; semi-cells 5 lobed, with 3-5 central mucilage pores, usually in a triangular pattern; polar lobe typically extended and well set-off from the rest of the semi-cell, broadly truncate with a shallow median notch; cells 57-74 µm long, 3l-47 µm broad; isthmus 10-19 µm wide; apex 18-23 µm wide.
Sample no.W12

19. ***Euastrum subhypochondrum*** Fritsch et Rich. (Fig. 5: 4)
http://www.digicodes.info/Euastrum_subhypochondrum.html#2006030031; Scott & Prescott 1961, P. 41, Pl. 10, Fig. 8
Cells as long as broad; semi-cells 3-lobed, polar lobe narrow with parallel sides, truncate or only very slightly retuse, not notched, shorter than basal lobes; lateral lobes slightly tapered upwardly; margins of lobes spinose; face showing a large central protuberance bearing a circle of granules surrounding one or two large granules, and having scattered spines within the lobes; cells 54-60 µm long, 52-58 µm broad; isthmus 12 µm wide.
Sample no. S15

Genus: ***Micrasterias*** C. Agardh ex Ralfs (1848)

20. ***Micrasterias rotata*** (Grev.) Ralfs var. ***curvata*** Hirano (Fig. 5:5-6)
Hirano 1992, P. 61, Pl. 27, Fig. 3.
Cells large, longer than broad, sub-elliptic outline, deeply constricted; sinus narrowly linear; semi-cells 7 lobed, deep and slightly open incision between the lobes, quite open incision between the lobules; polar lobe long and grow out of other lobes, with subparallel sides, slightly swollen at the middle, apex not expanded, with 4 apical spines; lateral lobes divided by 3 incisions into 4 lobules, apex of each lobule curve towards polar lobe; cells 396 µm long, 127-167 µm broad, isthmus 39.6 µm wide.
Sample no. R35

Genus: ***Cosmarium*** Corda ex Ralfs (1848)

21. ***Cosmarium auriculatum*** Reinsch (Fig. 5:8-9)
Scott & Prescott 1961, P. 54, Pl. 26, Fig. 4; Nurul Islam 1970, P. 923, Pl. 15, Figs. 13-15; Nurul Islam & Yusuf Haroon 1980, P. 574, Pl. 15, Figs. 208, 209; Bharati & Hegde 1982, P. 736, Pl. 3, Fig. 3; Prasad & Misra 1992, P. 153, Pl. 22, Fig. 14.
Cells more or less circular, as long as broad, constriction not deep; sinus opens outwards with rounded apex; semicell sub-elliptical to sub-semicircular, sides 5 undulate with sharp and pointed ridges; cell wall punctate; cells 45-48 µm long, 42-55 µm broad; isthmus 20-22 µm wide.
Sample no.S19

22. ***Cosmarium rectangulare*** Grun. (Fig. 5:12)
Croasdale & Flint 1988, P. 97, Pl. 38, Fig. 4.
Cells slightly longer than broad; sinus deep and closed; semicells subhexagonal-reniform, sides from rectangular-rounded basal angles extending parallel in lower half, then tapering gradually to the flattened apex; semicells in side view obovate-circular, in end view elliptic, often swollen in the middle; cells 26-51 µm long, 21-44 µm broad; isthmus 7-15 µm wide.
Sample no.W10

23. ***Cosmarium regnesi*** Reinsch (Fig.5: 13)
Scott & Prescott 1961, P. 68, Pl. 32, Fig. 24; Bharati & Hedge 1982, P. 750, Pl. 11, Fig. 9; Croasdale & Flint 1988, P. 99, Pl. 37, Fig. 14 (as var. *regnesi*).
Cells small in size; deeply constricted with widely open sinus, rounded in the interior; semicells transversely oblong-rectangular, with 6 (rarely 8) evenly spaced marginal teeth (2 lateral and 2 or 4 apical), sides and apex between the teeth widely excavate; wall smooth; cells with process 15 µm and without process 10 µm long, with process 15 µm broad; isthmus 4.5 µm wide.
Sample no.W11

24. ***Cosmarium taxichondrum*** Lund. var. ***undulatum*** Scott et Prescott (Fig. 5:14-16; Fig. 6: 1-3)
Scott & Prescott 1958, Pl. 14, Fig. 5; Nurul Islam & Irfanullah 1999, P. 95, Pl. 2, Figs. 22- 24.
Cells as long as broad, broadly elliptical at top view; sinus open; wall tricrenulate just above the sinus; large granules with pores on face of each semicell and median supra-isthmian granule not distinct; cells 26-28 µm long, 26-27 µm broad; isthmus 8 µm wide.
Sample no. W11

Genus: ***Xanthidium*** Ehrenberg ex Ralfs (1848)

25. ***Xanthidium*** cf ***aculeatum*** Ehrenberg ex Ralfs (Fig. 6:4-6)
Croasdale & Flint 1988, P. 119, Pl. 60, Figs. 8-9.
Cells as long as broad or slightly longer; sinus barely open from a narrow rounded interior; semicells elliptic reniform or elliptic-subsemicellular, apex sub-truncate and often slightly elevated; each side of the semicells bearing about 20 or more stout spines, irregularly arranged in a broad band near and on the margin; centre of the semicell with a prominent truncate protuberance bearing peripheral granules or lobes; cells 64-76 µm long, 60-75 µm broad; isthmus 19-22 µm wide; spines 4-7 µm long.
Sample no. W10

26. ***Xanthidium burkillii*** West & West var. ***alternans*** Skuja (Fig. 6: 7-8)
Skuja 1949, P. 149, Pl. 33, Fig. 16; Scott & Prescott 1961, P. 80, Pl. 40, Fig. 2; Hirano 1992, P. 63, Pl. 36, Fig. 1.
Cells slightly broader than long; sinus widely open to nearly closed; semicells subelliptic, the paired apical spines nearly as long as the others; central area slightly swollen and smooth; cell 80-84 µm long, 87-94 µm broad with spine; 45-49 µm long, 45-48 µm broad without spine; isthmus 23 µm wide; spines 12-22 µm long.
Sample no.R35

Genus: ***Staurodesmus*** Teiling (1948)

27. ***Staurodesmus brevispina*** (Breb.) Croasd. [Current accepted name: *Staurastrumbrevispina* Bréb.] (Fig. 6:9)
Croasdale et al. 1994, P. 40, Pl. 73, Figs. 12-18.
Cells as long as broad; semicells broadly oval, not tapered, corners rounded with a very short spine or papilla; sinus linear and narrow, at least in inner part; isthmus moderate: apex convex, sometimes subtruncate or slightly retuse in middle; cells in end view triangular with concave sides and rounded angles terminating in spine or papilla; cells 27-52 µm long, 27-57 µm broad; isthmus 8-17 µm wide.
Sample no. S22

28. ***Staurodesmus cuspidatus*** var. ***curvatus*** (W. West) Teil. (Fig. 6:11)
Croasdale et al. 1994, P. 44, Pl. 66, Figs. 19-22.
Cells as long as broad; semicells more or less triangular or cup shaped, extending into the spines and moderately convex sides; isthmus narrow; apex concave or straight; spines long,

divergent, vertically pointed; cells 19-40 µm long, 20-36 µm broad without spines; isthmus 7 µm wide.
Sample no. W9

29. ***Staurodesmus dickiei*** var. ***rhomboideus*** West et West ex Lill. (Fig.6: 12)
Croasdale et al. 1994, P. 46, Pl. 76, Figs. 8-10;
Cells as long as broad; semicells rhomboidal, apex straight or angularly convex; spines somewhat longer and stouter than the typical and strongly convergent; cells 31-43 µm long, 31-46 µm broad; isthmus 9-11 µm wide.
Sample no. W9

30. ***Staurodesmus mamillatus*** (Nordst.) Teil. var. ***mamillatus*** (Fig. 6: 13-15)
Croasdale et al. 1994, P. 52, Pl. 67, Figs. 1-6.
Cells small in size; semicells oval–triangular, angles not merging into spines; isthmus fairly narrow and elongated, forming a one third of the length of the semicells or longer; apex mostly straight or convex; spines moderate in length, divergent to convergent, fairly narrow arising rather abruptly from the rounded end of the semicells; cells 16-34 µm long, 14-28 µm broad: isthmus 5-7 µm wide.
Sample no.S14

Genus: ***Staurastrum*** Meyen ex Ralfs (1848)

31. ***Staurastrum contectum*** Turn. [Current accepted name: *Staurastrum quadrangulare* var. *contectum* (Turner) Grönblad] (Fig. 6:16)
Scott & Prescott 1961, P. 88, Pl. 54, Fig. 6; Hirano 1992, P 70, Pl. 39, Fig. 2
Cells as long as broad or slightly longer; sinus widely open; semicells obtriangular, lateral margin slightlyconvex; apex concave, apical angles with a pair of sharp spines and with a slender process just below the spines; cells 27 µm long, 44 µm broad (with spine); isthmus 12 µm wide.
Sample no.S23

32. ***Staurastrum freemanii*** West et G.S. West var. ***nudiceps*** Scott et Prescott (Fig. 7: 3-4)
Scott & Prescott 1961, P. 92, Pl. 43, F.3; Hirano 1992, P. 73, Pl. 38, Figs. 1,8.
Cells subfusiform or subtriangular; sinus acute, opening widely; angles produced to form moderately long, stout, tapering processes which are divergent and end in 3 (or rarely 2) stout spines; apex slightly convex, rarely flat; cells 30 µm long, 30 µm broad without spines; 54-60 µm long, 63-72 µm broad with spines; isthmus 10 µm wide.
Sample no.S24

33. ***Staurastrum longispinum*** (Bailey) W. Archer (Fig. 7: 5-8)
West & West 1923, P. 33, Pl. 134, Fig. 1.
Cells very large, deeply constricted; sinus acute, opening widely; semicells subelliptical or subtriangular, dorsal margin slightly convex, ventral margin more strongly so, angles very slightly produced and provided with 2 stout spines of varying length, projecting obliquely outwards and lying in the same vertical plane, two spines either parallel or converging; cell wall thick, punctate; cells 90-120 µm long, 73-100 µm broad (without spines); isthmus 36-41 µm wide; spines 9.5-32.5 µm long.
Sample no. W2

34. ***Staurastrum unicorne*** Turn. var. ***ecorne*** Turn. ex West et G.S. West f. *retusum* Scott et Prescott (Fig. 7:12-15)
Scott & Prescott 1961, P. 116, Pl. 53, Figs. 3, 4; Croasdale. et al. 1994, P. 62, Pl. 68, Figs. 1-5 (as *S. unicornis* var. *unicornis*).
Cells small, as long as broad or little broader; sinus rounded or sharp; semicells inversely triangular, the angles extended and capitate or knob-like, with or without a short spine; isthmus not or slightly elongated; apex convex in middle; cells 20-27 µm long, 22-31 µm broad; isthmus 6-7.5 µm wide.
Sample no.R34

Genus: ***Teilingia*** Bourrelly (1964)

35. ***Teilingia excavata*** (Ralfs ex Ralfs) Bourrelly (Fig. 7:16)
Croasdale et al. 1994, P. 168, Pl. 130, Figs. 1-4.
Filament short or long, straight, with or without mucilaginous sheath; cells small; sinus large,

rounded and moderately deep (excavated); semicells broadly elliptic; apex flattened and in face view, 2 of its 4 granules (attachment organelles) visible near the outer margin; in side view cell oblong-elliptic; cell wall smooth or with small granules; cells 7.5-17 µm long, 7-14 µm broad; isthmus 5-7.5 µm wide.
Sample no. S22

Other green algae viz., *Nephrocytium hydrophilum* (Turner) Wille (Fig. 3: 1), *Botryococcus braunii* Kützing (Fig. 3: 2), *Kirchneriella contorta* (Schmidle) Bohlin [New Name: *Raphidocelis danubiana* (Hindák) Marvan, Komárek & Comas] (Fig. 3: 13), *Quadrigula chodatii* (Tanner-Fullman) G.M. Smith (Fig. 3: 14-15), *Actinotaenium* cf. *wollei* (West et West) Teiling (Fig. 5: 7), *Cosmarium dorsitruncatum* (Nordstedt) G.S. West (Fig. 5: 10-11), *Staurodesmus cuspidatus* (Bréb.) Teil (Fig. 6: 10), *Staurastrum disputatum* West et G.S. West var. *sinense* (Lutk.) West et G.S. West [Current accepted name: *Staurastrum sinense* Lutkemüller] (Fig. 7: 1-2) and *Staurastrum setigerum* Cleve (Fig. 7: 9-11) were also reported from this wetland which were not described in previous paper by Godar & Rai (2018). But, these species have been reported earlier from Bagh-Jhoda Wetland (Rajopadhyaya & Rai, 2016-18), Hasina Wetland (Rai & Rai, 2018) and Jagadishpur Tal (Rai & Paudel, 2019).

In this study, interesting but rare species were *Nephrocytium hydrophilum, Quadrigula chodatii, Triploceras gracile, Euastrum subhypochondrum, Cosmarium regnesi, Staurastrum freemanii* var. *nudiceps* and *S. disputatum* var. *sinense. Pleurotaenium coroniferum, P. maculatum, Staurastrum unicorne var. ecorne f. retusum, Tetraedron regulare* and *Kircheneriella contorta* were also interesting algae not found commonly in other localities in the country.

Two species viz., *Actinotaenium* cf. *wollei* and *Xanthidium* cf. *aculeatum* were closely similar to their type species. The general appearance of the materials from Raja-Rani agrees well with the description given by Kouwets (1997) for *A. wollei* and Croasdale & Flint (1988) for *X. aculeatum*. In our specimens, *A.* cf. *wollei* was slightly larger in dimention with more broad at the centre and tapering towards the apices, quite rectangular outline rather elliptical outline of semicells. *Xanthidium aculeatum* observed here has quite broad and long spines than the type. These two specimens may belong to new species or variety. For this, a detailed study of the shape of chloroplast in living material and zygospore is necessary to clear up the taxonomy.

The small algae *Cosmarium regnesi* described here is more or less similar to its variety *productum* but due to rare species further confirmation could not be done. *Cosmarium taxichondrum* var. *undulatum* were observed in varieties of forms in different age of growth. *Micrasterias rotata* var. *curvata* has no distinct constriction at apical lobe and the semicell is quite elongated otherwise it is closely related with *M. torreyi* var. *sachlanii* described by Scott & Pescott (1961). In this study *Tetraedron regulare* and *T. minimus* were also present simultaneously.

Conclusion

Present paper described a total of 35 green algae first time from Nepal including 9 other previously reported algae. Thus, the total green algae reported from Raja-Rani wetland including the previous report of 72 algae by Godar & Rai (2018) is 116. This study showed that Raja-Rani wetland is rich in algal diversity and harbours many interesting green algae. The wetland may consist of many other groups of interesting algae. Thus, further extensive explorations are essential to explore many more other groups of algae.

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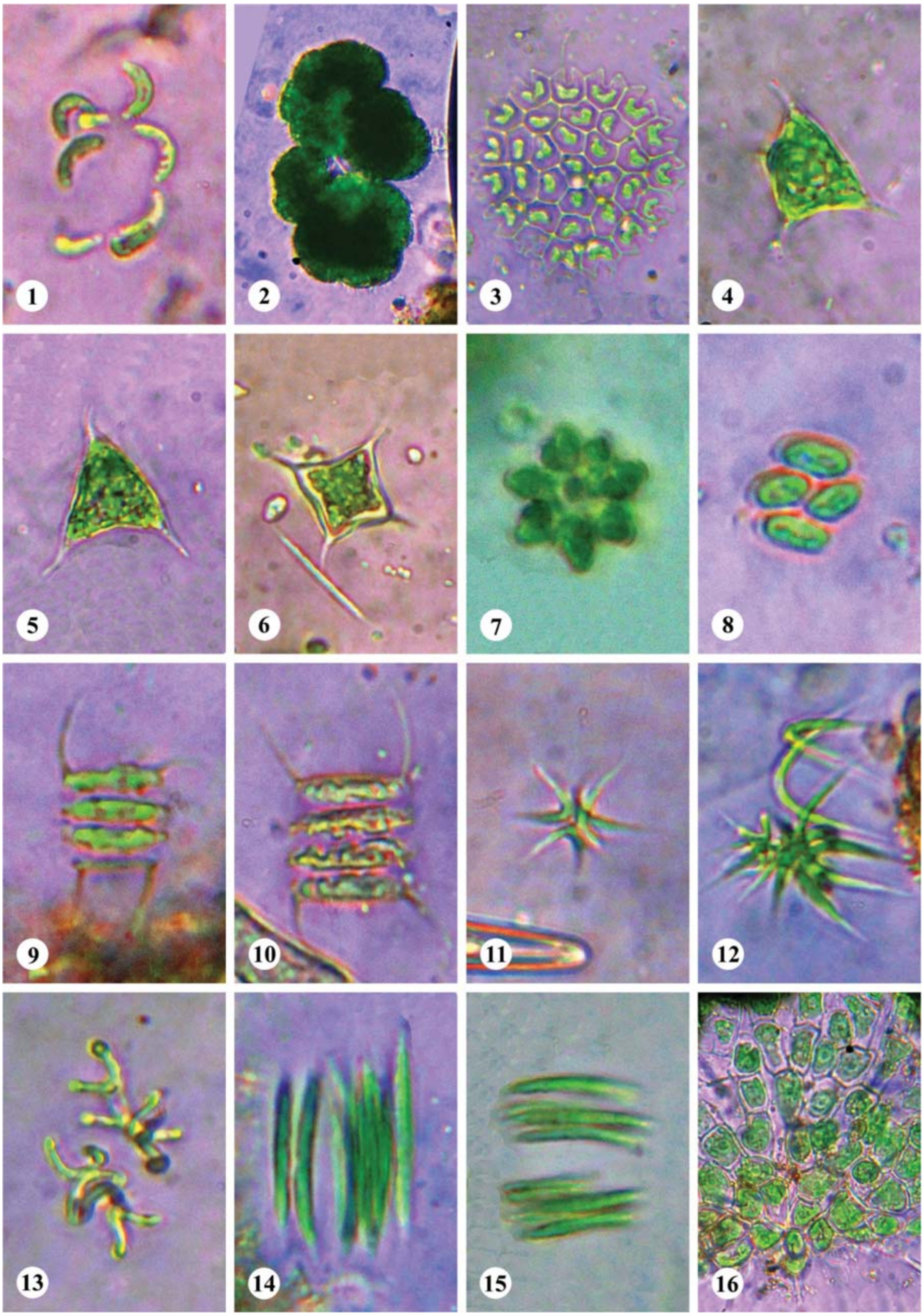

Figure 3: 1. *Nephrocytium hydrophilum* 2. *Botryococcus braunii* 3. *Pediastrum angulosum* var. *laevigatum* 4-6. *Tetraedron regulare* 7. *Coelastrum astroideum* 8. *Scenedesmus graevenitzii* var. *alternans* 9-10. *S. tropicus* 11-12. *Selenastrum westii* 13. *Kirchneriella contorta* 14-15. *Quadrigula chodatii* 16. *Coleochaete pulvinata*

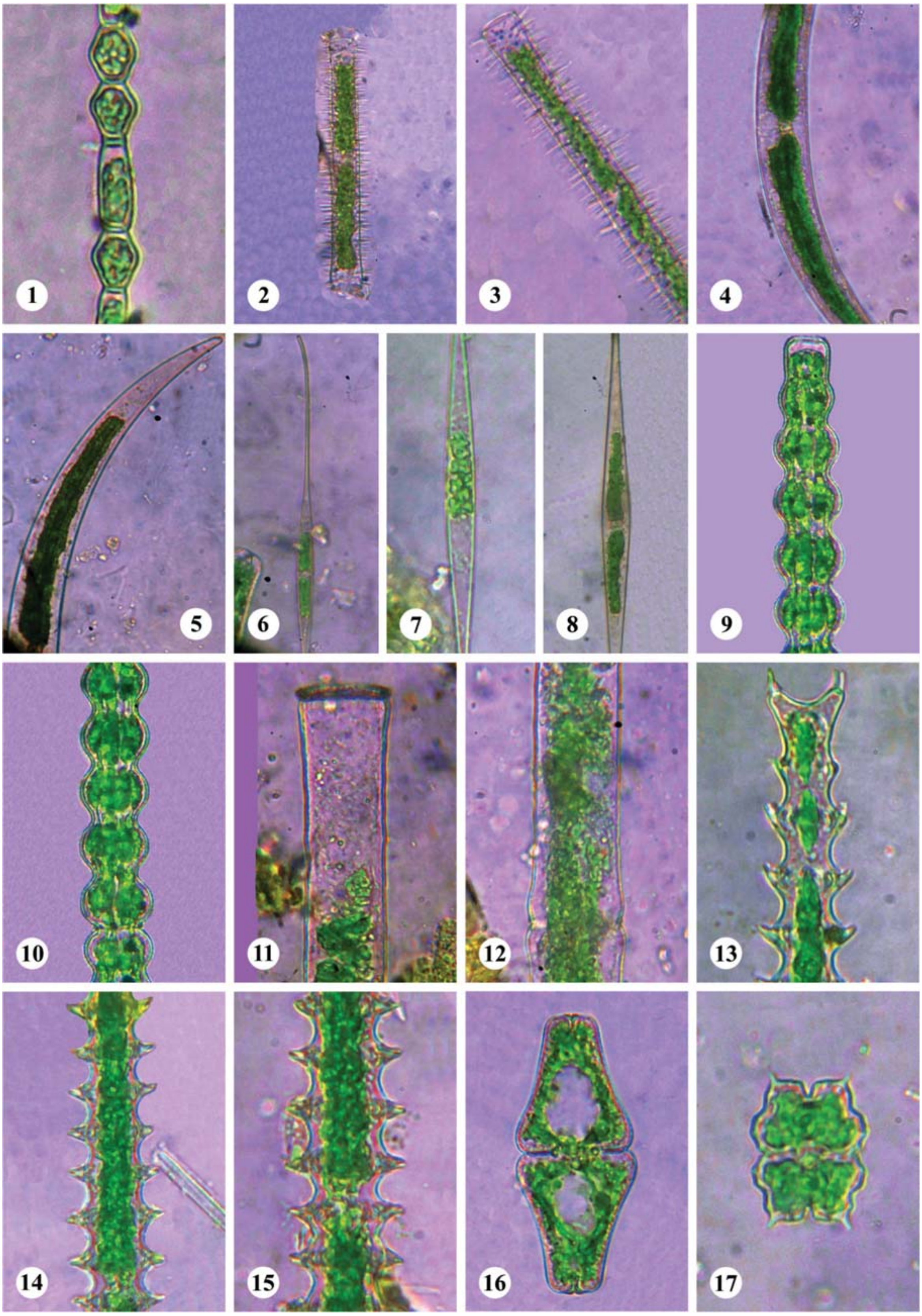

Figure 4: 1. *Oedogonium reinschii* 2-3.*Gonatozygon aculeatum* 4-5. *Closterium archerianum* 6-8. *Closterium setaceum* var. *elongatum* 9-10. *Pleurotaenium coroniferum* 11-12. *P. maculatum* 13-15. *Triploceras gracile* 16. *Euastrum ansatum* var. *rhomboidale* 17. *E. denticulatum* var. *quadrifarium*

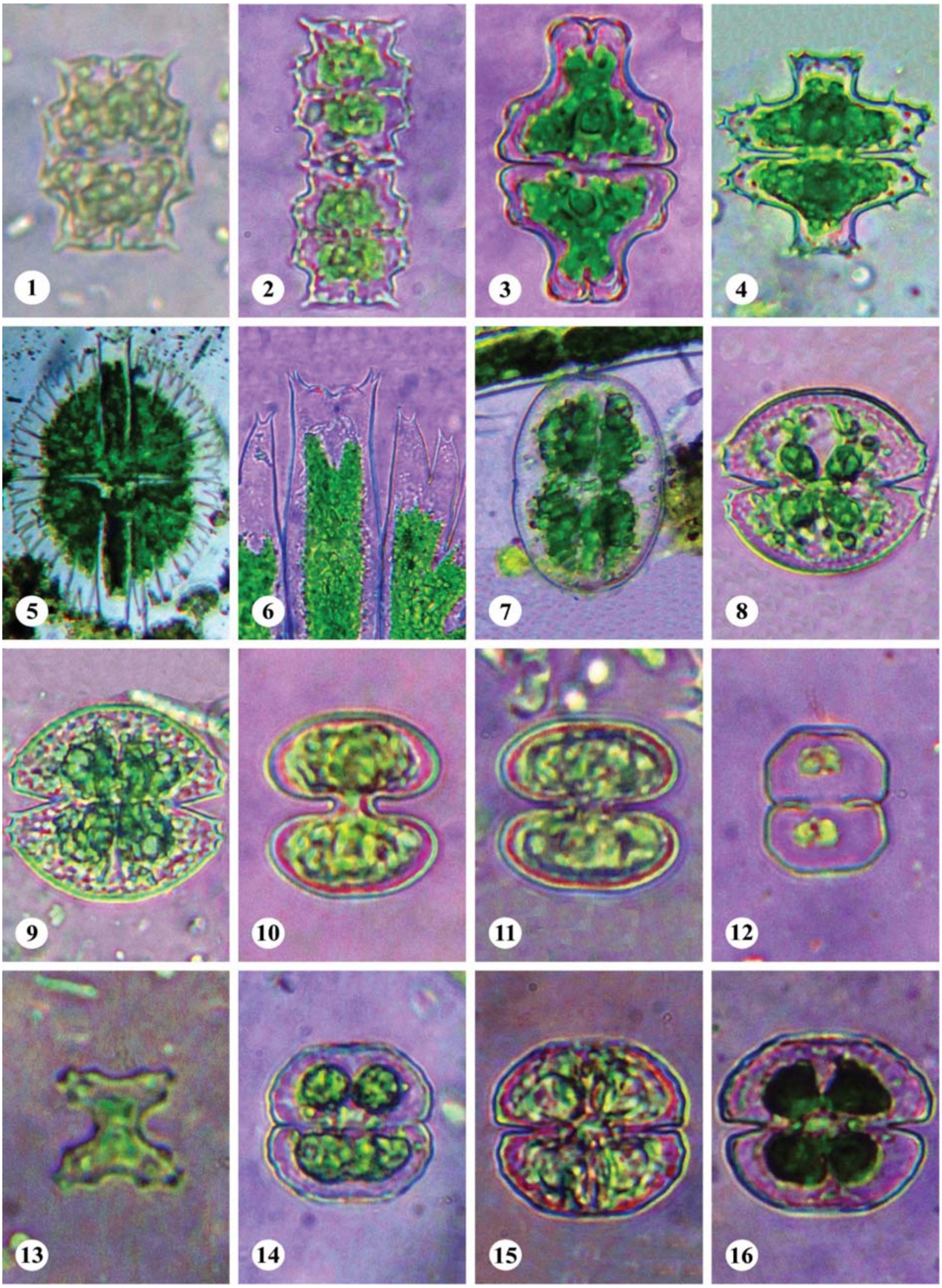

Figure 5: 1-2. *Euastrum pulchellum* 3. *E. sinuosum* var. *germanicum* 4. *E. subhypochondrum* 5-6. *Micrasterias rotata* var. *curvata* 7. *Actinotaenium* cf. *wollei* 8-9. *Cosmarium auriculatum* 10-11. *C. dorsitruncatum* 12. *C. rectangulare*13. *C. regnesi* 14-16. *C. taxichondrum* var. *undulatum*

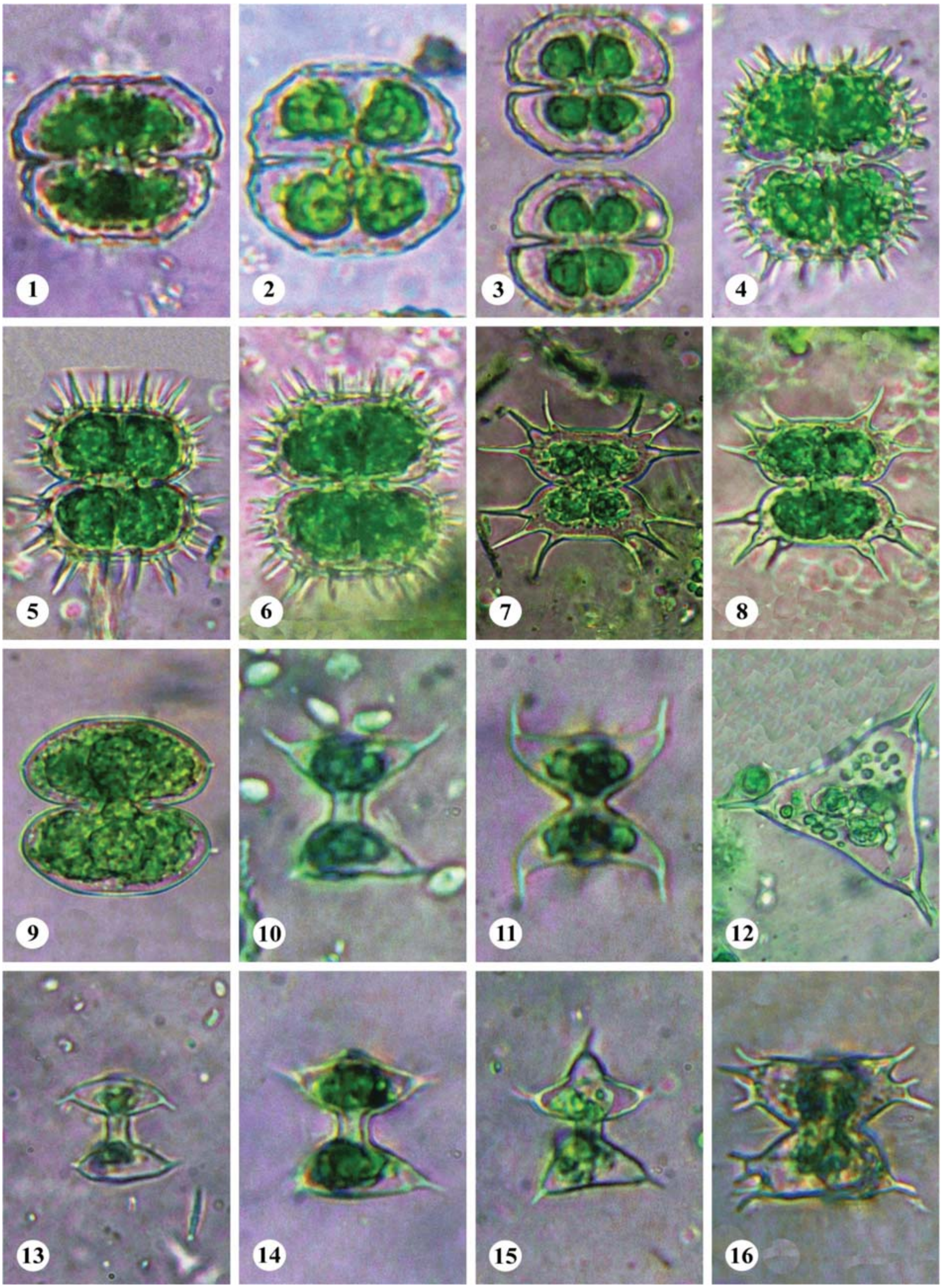

Figure 6: 1-3. *Cosmariumtaxichondrum* var. *undulatum* 4-6. *Xanthidium* cf *aculeatum* 7-8. *X. burkillii* var. *alternans* 9. *Staurodesmus brevispina* 10. *S. cuspidatus* 11. *S. cuspidatus* var. *curvatus* 12. *S. dickiei* var. *rhomboideus* 13-15. *S. mamillatus* var. *mamillatus* 16. *Staurastrum contectum*

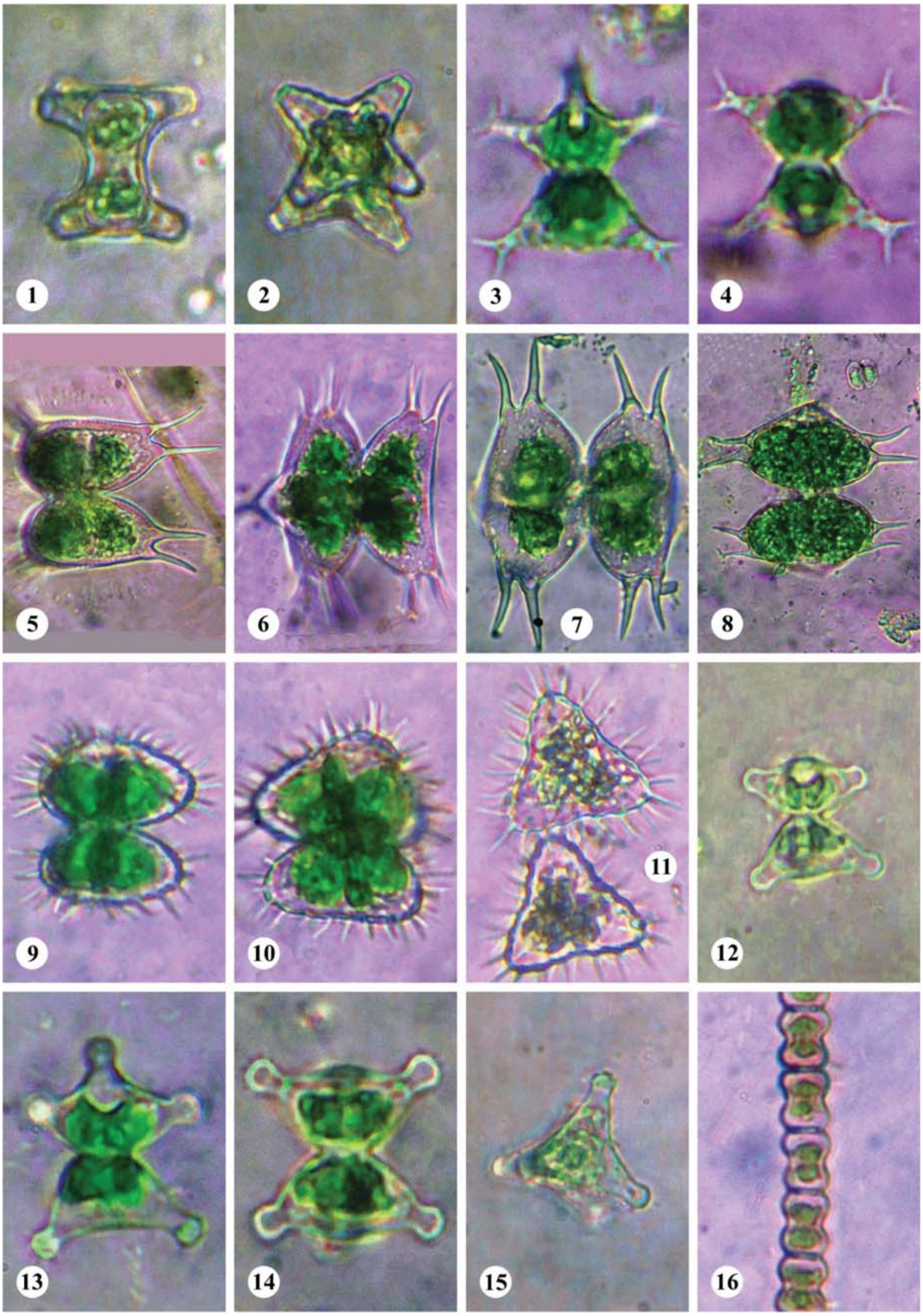

Figure 7: 1-2.*Staurastrum disputatum* var. *sinense* 3-4. *S. freemanii* var. *nudiceps* 5-8. *S. longispinum* 9-11. *S. setigerum* 12-15. *S. unicorne* var. *ecorne* f. *retusum* 16. *Teilingia excavata*

References

Akiyama, M. (1989). Some *Trentepohlia* species from Kathmandu, Nepal Himalaya. *Bull. Natn. Sci. Mus., Ser. B (Bot.), Tokyo, 15*(3), 81-90.

Bando, T., Nakano, T., & Watanabe, M. (1989). The desmid flora of Kathmandu, Nepal. *Bull. Natn. Sci. Mus., Ser. B (Bot.)*, *Tokyo, 15*(1), 1-25.

Baral, S.R. (1996). Diversity of blue-green algae (Cyanobacteria) in Nepal. *In*:P.K. Jha *et. al*. (Eds.), *Environment and biodiversity in the context of South Asia* (pp. 176-181). Regional conference on Environment and Biodiversity. 7-9 Mar., 1994, Ecological Society, Kathmandu, Nepal. Proceedings.

Baral, S.R. (1999). Algae of Nepal. *In*: T.C. Majupuria, & R. Kumar(Eds.), *Nepal Nature's Paradise* (pp. 655-681). Gwalior, India.

Bharati, S.G., & Hegde, G.R. (1982). Desmids from Karnataka State and Goa, Part III. Genus *Cosmarium* Corda. *Nova Hedwigia*, *36*, 733-757.

Bruehl, P., & Biswas, K. (1926). Algae of Loktak lake. *Mem. Asiatic. Soc. Bengal*, *8*(3), 257-315.

Chaturvedi, U.K., & Habib, I. (1996). A systematic account of Chlorococcales from Nepal. *Phykos*, *35*(1-2), 129-137.

Croasdale, H., & Flint, E.A. (1988). *Flora of New Zealand*, *Fresh water algae, chlorophyta, desmids with ecological comment on their habitats* (pp. 147p). Vol. II. Botany Division, D.S.I.R., Christchurch, New Zealand.

Croasdale, H., & Flint, F.A. (1986). *Flora of New Zealand*, *Fresh water algae, chlorophyta, desmids with ecological comments on their habitat* (pp. 133p). Vol. I.,Wellington, New Zealand:V.R. Ward, Government Printer.

Croasdale, H., Flint, E.A., & Racine, M.M. (1994). *Flora of New Zealand*, *Fresh water algae, chlorophyta, desmids with ecological comments on their habitat* (pp. 218p). Vol. III.,Lincoln, Canterbury, New Zealand.

Das, S.N., & Verma, B.N. (1996). Algal flroa of Chitwan and Nawalparasi districts of Nepal. *Phykos*, *35*(1-2), 119-127.

Dwevedi, N.P. (1985). Some new records of Chaetophorales from Nepal. *Mendel*, *1-2*(2), 120-123.

Förster, K. (1965). Beitrag zur kenntais der Desmidiaceen-flora von Nepal. Erg. Forsch-unturn Nepal Himalaya. *Khumbu Himal*,*1,2*, 25-58.

Department of Forest Research and Survey. (2014). *Churia forests of Nepal* (2011-2013). Forest Resource Assessment Nepal Project, Kathmandu, Nepal: Author.

Fritsch, F.E., & Richs, F. (1937). Contribution to our knowledge of the fresh water algae of the Africa XII- Algae from Bel fast pan Transuaal-Trans. *Roy. Soc. S. Africa*., *25*(2), 153-288.

Gandhi, H.P. (1999). *Fresh water diatom of Central Gujrat*. Deharadun, India: B. Singh & M.P. Singh (pp. 324p).

Geitler, L. (1932). *Cyanophyceae in Rabenhorst's Kryptogamen flora*, *Leipzig 14*. 1196p.

Godar, K., & Rai, S.K. (2018). Freshwater green algae from Raja-Rani wetland, Bhogateni-Letang, Morang, Nepal. *Journal of Plant Resources*, *16*(1), 1-17.

Guiry, M.D. (2012). "How many species of algae are there?", *Journal of Phycology*, *48*(5), 1057-63. doi: 10.1111/j.1529-8817.2012.01222.x. PMID 27011267.

Guiry, M.D., & Guiry, G.M. (2019). *AlgaeBase*. World-wide electronic publication, National University of Ireland, Galway. http://www.algaebase.org; searched on December 2019.

Habib, I., & Chaturvedi, U.K. (1995). On some desmids from Nepal. *J. Ind. bot. Soc.*,*74*(1-4), 277-282.

Habib, I., & Chaturvedi, U.K. (1997). Contribution to the knowledge of desmids from Nepal. *Phykos*,*36*(1-2), 27-36.

Haga, M. (1988). A brief note on filamentous Chlorophyceae from Kathmandu. *In*: M. Watanabe, & S.B. Malla (Eds.), *Cryptogams of* *the Himalayas*, *The Kathmandu Valley,* (pp. 47-55) Vol. 1, Tsukuba, Japan: National Science Museum.

Haga, M., & Legahri, M.K. (1993). *Hydrurus foetidus* (Villars) Trevisan from mountain streams in Pakistan and Nepal. *In*: T. Nakaike & S. Malik (Eds.), *Cryptogamic flora of Pakistan*, (pp. 1-11) Vol. 2, Tokyo, Japan: National Science Museum.

Hickel, B. (1973). Limnological investigations in lakes of Pokhara valley, Nepal. *Int. Rev. ges Hydrobiol.*, *58*(5), 659-672.

Hirano, M. (1984). Fresh water algae from East Nepal. Study reported of *Baika Junior College*, *32,* 197-215.

Hirano, M. (1992). Desmids from Thailand and Malaysia. *Contributions from the Biological Lab*, Kyoto University, *28*(1), 1-98.

Ichimura, T. (1997). Natural populations of the *Closterium ehrenbergii* (Desmidiales, Chlorophyta) species complex in Nepal. *Phycol. Res.*, *45*(1), 47-54.

Ishida, Y. (1986). Limnological study in Phewa, Begnas and Rupa lakes. *In*: Y. Ishida (Ed.), *Studies on distribution, adaptation and evolution of microorganisms in Nepal Himalayas, second report,* Japan: Ministry of Education, Science and Culture.

Jha, S., & Kargupta, A.N. (2012). Record of two genera: *Spirulina* and *Arthrospira* (Nostocales, cyanophyceae) along river Koshi basin of Nepal and Bihar. *Phytomorphology*, *62*(1-2), 25-31.

Joshi, A.R. (1979). Contribution to our knowledge on myxophyceae of Nepal. *J. Nat. Hist. Mus.* (Nepal), *3*(2), 35-41.

Jüttner, I., Chimonides, J., & Cox, E.J. (2011). Morphology, ecology and biogeography of diatom species related to *Achnanthidium pyrenaicum* (Hustedt) Kobayasi (Bacillariophyceae) in streams of the Indian and Nepalese Himalaya. *Algological Studies*,*136*/*137*, 45-76. doi: 10.1127/1864-1318/2011/0136-0045.

Kargupta, A.N., & Jha, S. (1997). The genus *Stigeoclonium* from the North Bihar and bordering areas of Nepal. *Phykos*, *36*(1-2), 37-42.

Komarek, J. (1983). Contribution to the Chlorococcal algae of Cuba. *Nova Hedwigia*, *37*(1), 65-180.

Komarek, J., & Watanabe, M. (1998). Contribution to the attached Cyanoprokaryotes from submerged biotopes in Sagarmatha National Park, Eastern Nepal. *Bull. Natn. Sei. Mas.*, Ser. B, Tokyo, *24*(4), 117-135.

Kouwets, F.A.C. (1987). Desmids from the Auvergne (France). *Hydrobiol.*,*146*, 193-263.

Kouwets, F.A.C. (1997). Contributions to the knowledge of the French Desmid flora I. New and noteworthy taxa from the Central and Eastern Pyrenees. *Arch. Protistenkd*., *148*, 33-51.

Krieger, W. (1937). *Die Desmidiaceen Europas mit Berucksichtigung der aussereuropaischen Arten Pt. 13 (1) of Dr. Rabenhorst's Kryptogamen-Flora von Deutschland, Osterreich und der Schweiz. Conjugatae. Die Desmidiaceen* (pp. 1-712). Leipzig: Ed. Akad. Verlags gesellschaft.

Krstiæ, S.S., Pavlov, A., Levkov, Z., & Jüttner, I. (2013). New *Eunotia* taxa in core samples from Lake Panch Pokhari in the Nepalese Himalaya. *Diatom Research*, *28*(2), 203-217. doi: 10.1080/0269249X.2013.782343.

Matthews, R.A. (2016). *Freshwater algae in Northwest Washington*, Vol. 2, Chlorophyta and Rhodophyta. A collection of open access books and monographs. I. http://cedar.wwu.edu/cedarbooks/1

Ministry of Forest and Soil Conservation. (2012). *Nepal Biodiversity Strategy*. Kathmandu, Nepal: Author.

Müller, J. (1965). *Spirogyra nepalensis*nov. spec. *Khumbu Himal*, *1*(4), 299-300.

Nakano, T., & Watanabe, M. (1988). Some species of Chlorococcales from Nepal. *In*: M. Watanabe & S.B. Malla (Eds.), *Cryptogams of the Himalayas* (pp 57-65) *Vol. 1, The Kathmandu valley*, Tsukuba, Japan: National Science Museum.

Necchi, O. Jr., West, J.A., Rai, S.K., Ganesan, E.K., Rossignolo, N.L., & de Goër, S.L. (2016). Phylogeny and morphology of the freshwater red alga *Nemalionopsis shawii* (Rhodophyta, Thoreales) from Nepal. *Phycological Research*, Japan, *64*, 11-18. doi: 10.1111/pre.12116.

Nozaki, H. (1988). Colonial Volvocales Chlorophyta from Kathmandu, Nepal. *In*: M. Watanabe & S.B. Malla (Eds.), *Cryptogams of Himalayas* (pp. 39-46) *Vol. 1.,*Tsukuba, Japan: National Science Museum.

Nozaki, H. (1990). Colonial Volvocales (Chlorophyta) from Central and East Nepal. *In:* M. Watanabe & S.B. Malla (Eds.), *Cryptogams of the Himalayas* (pp. 41-47), *Vol. 2, Central and eastern Nepal*, Tsukuba, Japan:National Science Museum.

Nozaki, H., & Kuroiwa, T. (1990). *Volvulina compacta*, a new species (Volvocaceae, Chlorophyta) from Nepal. *Phycologia*, *29*(4), 410-417.

Nozaki, H., & Kuroiwa, T. (1991). Morphology and sexual reproduction of *Gonium multicoccum* (Volvocales, Chlorophyta) from Nepal. *Phycologia*, *30*(5), 381-393.

Nurul Islam, A.K.M. (1970). Contributions to the knowledge of desmids of East Pakistan, Part I. *Nova Hedwigia*, *20*, 903-983.

Nurul Islam, A.K.M., & Irfanullah, H.M. (1999). New records of desmids for Bangladesh-III. 24 taxa. *Bangladesh J. Plant Taxon.*, *6*(2), 91-104.

Nurul Islam, A.K.M., & Yusuf Haroon, A.K. (1980). Desmids of Bangladesh. *Int. Revue ges. Hydrobiol.*, *65*(4), 551-604.

Olenina, I., Hajdu, S., Edler, L., Andersson, A., Wasmund, N., Busch, S., Göbel, J., Gromisz, S., Huseby, S., Huttunen, M., Jaanus, A., Kokkonen, P., Ledaine, I., & Niemkiewicz, E. (2006). *Biovolumes and size-classes of phytoplankton in the Baltic Sea* HELCOM Balt. Sea Environ. Proc. No. 106, 144pp. (PDF), PEG_BVOL.zip

Opute, F.I. (2000). Contribution to the knowledge of algae of Nigeria. I. Desmids from the Warri/Forcados Estuaries. Part II. The elongate baculiform desmids. *J. Limnol.*, *59*(2), 131-155.

Philipose, M.T. (1967). *Chlorococcales*. I.C.A.R. monograph on algae, New Delhi. 365p.

Prasad, B.N., & Misra, P.K. (1992). *Fresh water algal flora of Andaman and Nicobar islands* (pp. 284). Vol.II, Dehradun, India: B. Singh & M.P. Singh Publ.

Prasad, P.K., Sinha, R.S.P., & Verma, B.N. (1985). Some rare green algae from Nepal. *Nat. Acad. 'Sci. Letters'*, India, *8*(2), 33-34.

Prasad, V. (1996). *Blue green algae of Birganj* (*Nepal*). Short term project. Nepal: Research Division, Tribhuvan University.

Prescott, G.W. (1951). *Algae of the western great lakes area* (pp. 977). WM.C. Brown Publishers, Dubque, Iowa.

Rai, D.R., & Rai, S.K. (2018). Freshwater algae (excluding diatoms and red algae) from Hasina wetland, Sundar Haraicha, Morang, Nepal. *Himalayan Journal of Science and Technology*, *2*, 1-12.

Rai, S.K. (2009). Some chlorophycean algae from Maipokhari lake, Ilam, east Nepal. *J. Nat. Hist. Mus*, Nepal, *24*, 1-8.

Rai, S.K. (2012). Five new species of *Oedogonium* Link (Chlorophyta), a freshwater filamentous algae from Nepal. *Nepalese Journal of Biosciences*, *2*, 17-23.

Rai, S.K. (2013). Occurrence of genus *Scenedesmus* Meyen (Chlorophyceae) from East Nepal. *Nepalese J. of Biosciences*, *3*, 26-37.

Rai, S.K. (2014). Additional desmids (Chlorophyceae) to eastern flora of Nepal. *Nepalese J. of Biosciences*, *4*, 40-46.

Rai, S.K., & Ghimire, N. (2020). Algal explorations in Nepal. *In*: M. Siwakoti, P.K. Jha, S. Rajbhandary, S.K. Rai (Eds.), *Plant Diversity of Nepal* (pp. 16-40), Kathmandu:Botanical Society of Nepal,

Rai, S.K., & Khadka, S. (2017). Diatoms of Bagmati River, Kathmandu, Nepal. *Nepalese Journal of*

Biosciences, *7*(1), 1-25.

Rai, S.K., & Misra, P.K. (2007). *Spirogyra* Link and *Sirogonium* Kutzing species: New to algal flora of Nepal. *Ecoprint*, *14*, 89-96.

Rai, S.K., & Misra, P.K. (2008). On some desmids from Koshi Tappu Wildlife Reserve, Nepal. *Ecoprint*, *15*, 47-58.

Rai, S.K., & Misra, P.K. (2010). Freshwater cyanophyceae from east Nepal. *Bangladesh J. of Plant Taxonomy*, *17*(2), 121-139.

Rai, S.K., & Misra, P.K. (2012). Taxonomy and diversity of genus *Pediastrum* Meyen (Chlorophyceae, Algae) in East Nepal. *Our Nature*, *10*, 167-175.

Rai, S.K., & Paudel, S. (2019). Algal flora of Jagadishpur Tal, Kapilvastu, Nepal. *J. Pl. Res.*, *17*(1), 6-20.

Rai, S.K., Rai, R.K., & Poudel, N. (2008). Desmids from Bees-Hazaar lake, Chitwan, Nepal. *Our Nature*, *6*, 58-66.

Rajopadhyaya, R., & Rai, S.K. (2016-18). Some freshwater algae from Bagh-Jhoda wetland, Morang: New for Nepal. *J. Nat. Hist. Mus.*, *30*, 15-27.

Rajopadhyaya, R., Joshi, S., Shrestha, S., & Rai, S.K. (2017). Some new and interesting cyanobacteria from Baghjhoda pond, Eastern Nepal. *Himalayan Journal of Science and Technology*, *1*(1), 1-8.

Sahay, A.P., Das, P.K., & Verma, B.N. (1992). Studies on the algal flora of Nepal-I: Chlorophyceae. *Geophytology*, *20*(2), 155-158.

Scott, A.M., & Prescott, G.W. (1958). Some freshwater algae from Arnhem land in the northern territory of Australia. *Rec. American-Australian Sci. Expn*. Arnhem land, *3*, 9-136.

Scott, A.M., & Prescott, G.W. (1960). Notes on Indonesian freshwater algae IV. *Reinwardtia*, *5*(3), 323-340.

Scott, A.M., & Prescott, G.W. (1961). Indonesian desmids. *Hydrobiologia*, *17*(1-2), 1-132. WMC. Publishers.

Shrestha, B., & Manandhar, J.D. (1983). Contribution to the algal flora of Kathmandu valley. *J. Inst. Sci. and Technol*, *6*, 1-6.

Shrestha, G. & Rai, S.K. (2017). Algal flora of Rajarani Lake, Dhankuta and their seasonal distribution. *Our Nature,15*(1-2), 44-54. doi: http://dx.doi.org/10.3126/on.v15i1-2.18793.

Simkhada, B., Jüttner, I. & Chimonides, P.J. (2006). Diatoms in lowland ponds of Koshi Tappu, eastern Nepal - Relationships with chemical and habitat characteristics. *Internat. Rev. Hydrobiol., 91*(6), 574-593.

Skuja, H. (1949). Zur Süsswasseralgenflora Burmas. *Nova Acta Regiae Societatis Scientiarum Upsaliensis, Series 4*, *14*(5), 1-188.

Smith, G.M. (1920). Phytoplankton of the Inland lakes of Wisconsin. *Bull. Wis. grol. Nat. Hist. Surv., 57,* 1-243.

Stastny, J. (2009). The desmids of the Swamp Nature Reserve (North Bohemia, Czech Republic) and a small neighbouring bog: species composition and ecological conditionof both sites. *Fottea*, *9*(1), 135-148.

Stastny, J. (2010). Desmids (Conjugatophyceae, Viridiplantae) from the Czech Republic; new and rare taxa, distribution, ecology. *Fottea, 10*(1), 1-74.

Subba Raju, N. & Suxena, M.R. (1979). Algae and Testacea of the Cho Oyu (Himalayas) Expedition-II; Cyanophyta, Chlorophyta, Euglenophyta, Chrysophyta and Testacea. *Hydrobiologia*, *67*(2), 141-160.

Suxena, M.R. & Venkateswarlu, V. (1968). Algae of the Cho Oyu (E. Himalaya) Expedition-I. Bacillariophyceae. *Hydrobiologia*, *32*, 1-26.

Tiffany, L.H. & M.E. Britton. (1952). *The algae of Illinois*. New York:Hafner publishing Co., 407p.

Turner, W.B. (1892). The fresh water algae of East India. *Kongl. Sv. Vet.* Akademiens Handlingar, *25*(5), 1-187.

Watanabe, M. & Komarek, J. (1988). Blue-green algae from Kathmandu. *In*: M. Watanabe & S.B.

Malla (Eds.), *Cryptogames of the Himalaya, The Kathmandu valley* (pp. 1-20), *Vol. 1,* Tsukuba, Japan: National Science Museum.

Watanabe, M. (1982). Observations on the genus *Closterium* from Nepal. *In*: Y. Otani (Compiled), *Reports on the Cryptogamic Study in Nepal*(pp. 47-59), Tokyo, Japan: National Science Museum.

Watanabe, M. (1995). Algae from lake Rara and its vicinities, Nepal Himalayas. *In*: M. Watanabe & H. Hagiwara (Eds.), *Cryptogams of the Himalayas, Nepal and Pakistan* (pp 1-20), *Vol. 3,* Tsukuba, Japan: National Science Museum.

West, W. & West, G.S. (1902). A contribution to the fresh water algae of Ceylone. *Trans. Linn. Soc. Bot.*, 2^{nd} Ser., *6*(3), 123-215.

West, W. & West, G.S. (1904-1912). *Desmidiaceae: A monograph of the British Desmidiaceae*, *1*, 1-224, 1904; *2*, 1-204, 1905; *3*, 1-272, 1908; *4*, 1-191, 1912. London: Ray Society.

West, W. & West, G.S. (1923). *A monograph of the British Desmidiaceae* (pp. 300). Vol. 5. London: Ray Society.

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Algal Flora of Gajedi Lake, Rupandehi District, Central Nepal

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Abstract

The study was carried out for the documentation of algal flora of Gajedi Lake of Kanchan Rural Municipality of Rupandehi district. Algal samples were collected by using plankton net for planktonic forms and squeezing submerged aquatic macrophytes for epiphytic forms from 10 sampling sites from periphery littoral zone of the lake during May, 2019. Altogether 33 taxa belonging to 10 orders, 14 families, 5 classes and 18 genera were reported in this study. Chlorophyta with 15 taxa was found to be the dominant phylumof algae which was followed by Charophyta (10),Cyanophyta (5), Euglenophyta (2) and Glaucophyta (1). *Scenedesmus* with eight taxa was found to be the dominant genera which was followed by *Coelastrum*, *Cosmarium*, *Euastrum* (3 taxa each), *Staurastrum, Oscillatoria* (2 taxa each)and so on.

Keywords: *Coelastrum, Cosmarium*, Cyanophyceae, *Scenedesmus.*

Introduction

Algae are the photosynthetic organisms and can convert the anaerobic atmosphere of the earth into an aerobic atmosphere by the process of oxygenic photo-phosphorylation. The plant body in algae is always a thallus and is not differentiated into root, stem and leaves. They have a varied size from minute unicellular plants (less than 1µ in diameter in some planktons) to very large highly differentiated multi cellular forms. Phytoplanktons that include algae as their exclusive constituent are directly related to fish populations of the oceans and thereby control the availability of 'sea food'. They are considered very important and their beneficial roles include, an excellent source of single-cell protein, hydrocarbons, biogas, polysaccharides such as agar-agar, antibiotics, colouring pigments as well as some important medicines. They also provide some harmful effects to the human such as, produce water blooms, toxins as well as some diseases.

Several studies have been carried out on algal flora of different Lakes of Nepal (Hirano, 1955; Hirano, 1963; Watanabe, 1995; Rai and Rai, 2012; Rajopadhya et al., 2017; Shrestha & Rai, 2017; Rai & Poudel, 2019) but the work has been done on documentation of algal flora in Nepal is not sufficient. So that documentation of algal flora in unexplored habitats is more important. The algal flora of Gajedi lake has not been studied till date. Hence, a database on number of algal presences is essential in order to document and understand the country's algal wealth. Thus, the database can be used as a baseline to search for more algal species reported to occur as well as act as first comprehensive documentation on the algae in this area.

Materials and Methods

Study area

Gajedi lake (27°39'51" N and 83°16'34" E, Elevation 133 m asl) is located in the Danapur (previously Gajedi) VDC, of Rupandehi district of province-5, southern Nepal located close to the pilgrimage site Lumbini. The village name is a part of Kanchan Rural Municipality (Figure 1). Butwal is notable for being the closest city to the Gajedi Lake and is geographically significant city in Nepal as it is located at the intersection of Nepal's two major highways, the Mahendra Highway and the Siddhartha Highway. This lake is a tourist attraction site as it is located in the jungle and is popular for internal as well as external tourism especially for picnics, boating activities and jungle safaris.

The lake covers an area of about 19 hectares (District Forest Office [DFO], 2073) surrounded by *Shorea robusta* forest and the common constituting tree species of this forest were *Adina cordifolia, Dalbargia sissoo*, *Dalbergia latifolia, Terminalia alata*, *Terminalia bellirica*, *Terminalia chebula*, *Pterocarpus marsupium*, *Schleichera oleosa* etc. to Eastern and North-Western sides, and by private land and settlement area to South-Western facing side (Dhakal et al., 2019). The aquatic macrophytes found in the lake include *Eichhornia crassipes*, *Hydrilla verticillata*, *Nelumbo nucifera*, *Pistia stratiotes*, etc.

The climate of the study site is subtropical and shares monsoon type of climate with dry winter and rainy summer. The average annual precipitation of the nearest meteorological station to the study sites i.e, Butwal showed mean annual precipitation 2600 mm and the mean maximum/minimum temperature recorded were 42.5°C/7.5°C (Thapa & Poudel, 2018).

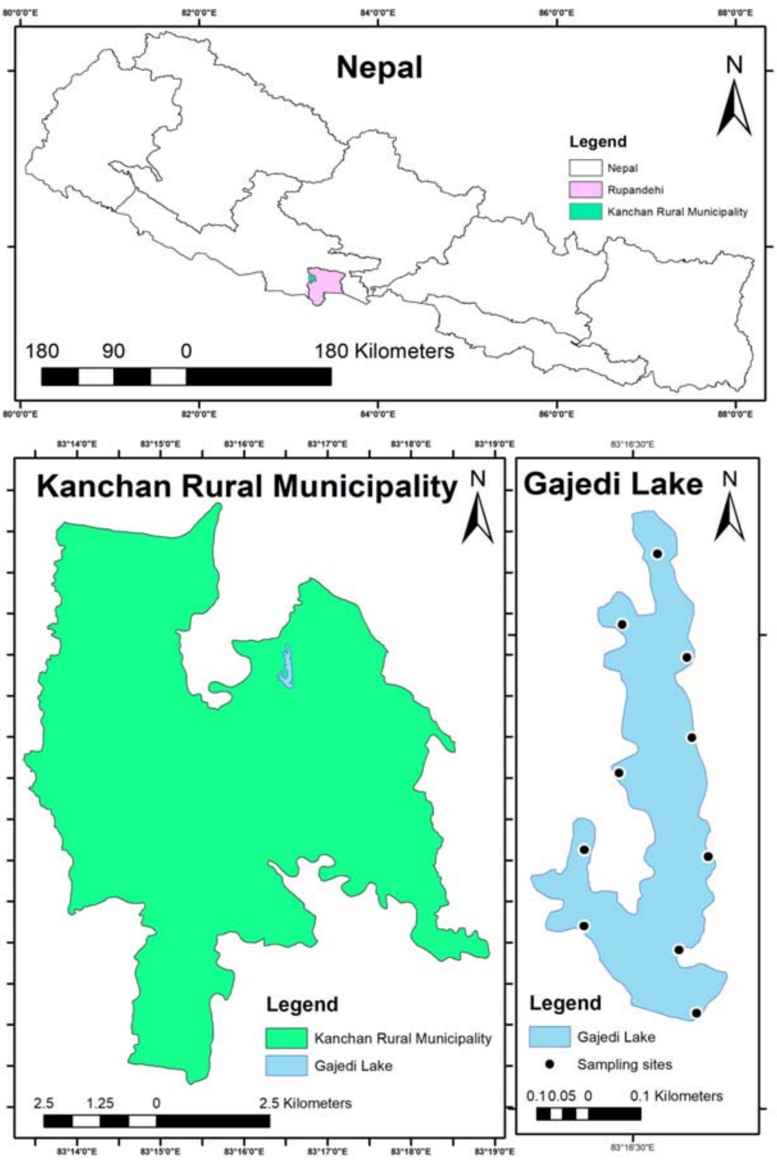

Figure 1: Location map of Gajedi Lake, showing algal sampling sites

Collection and identification of algal samples

A total of 30 algal samples were collected from peripheral littoral zone of the lake during May2019 but the data of only 10 samples were described in this paper. Geographical position (Latitude, Longitude, Elevation) and physico-chemical parameters like temperature, pH, conductivity (by Hanna Multi-range EC meter, HI8733) were measured on the spot (Table 1). Algal samples were collected by using plankton net for planktonic forms and squeezing submerged aquatic macrophytes for epiphytic forms. Each sample was assigned with collection number starting from G1, G2, G3,.....G10 and preserved in 4% formaldehyde solution in airtight glass bottles and brought to the laboratory of National Herbarium and Plant Laboratories, Godawari. All the collected materials have been deposited in the Cryptogams Section (Algae, Fungi and Lichen), National Herbarium and Plant Laboratories, Godawari, Lalitpur for further examination. Samples were screened then; microphotography was done for each specimen under 40X objective using Huma Scope Premium LED microscope fitted with the digital camera. Taxa (excluding of diatoms), were identified consulting various articles, literatures and monographs (West & West, 1904; Prescott, 1951; Prescott & Scott 1952; Deshikachary, 1959; Philipose, 1967; Hirano, 1969; Croasdale &Flint, 1988; Rai & Misra, 2008 and 2010; Rai, 2013; Halder, 2016; Shrestha & Rai, 2017; Godar & Rai, 2018; Mhaske & Talwankar, 2018). Nomenclature, as well as classification follows Guiry & Guiry (2019).

Table 1: Physico-chemical parameters of water at different collection sites of Gajedi Lake

S.N.	Collection No.	pH	Temperature (°C)	Conductivity (µs/cm)
1.	G1	6.5	32	306
2.	G2	6.5	29	337
3.	G3	6.5	29	334
4.	G4	6.5	28	316
5.	G5	6.5	28	316
6.	G6	7	34	303
7.	G7	6.5	28	316
8.	G8	6.5	28	316
9.	G9	7	34	303
10.	G10	7	34	303
Mean		**6.65**	**30.4**	**315**

Results and Discussion

In the present study, a total of 33 algal taxa belonging to 18 genera, 14 families, 10 orders and 5 classes have been reported from Gajedi Lake, Rupandehi, Nepal (Table 3). It included six taxa identified up to generic level only, (viz., *Oedogonium* sp., *Oscillatoria* sp., *Phacus* sp., *Phormidium* sp., *Scenedesmus* sp. and *Staurastrum* sp.) and three taxa closely related to corresponding species (viz., *Ankistrodesmus* cf. *falcatus*, *Cosmarium* cf. *seelyanum* and *Trachelomonas* cf. *oblonga* Lemmermann Wolle (Corda) Ralfs. Among the classes, Chlorophyceae have the maximum number of algal species followed by Conjugatophyceae and Cyanophyceae. However, Euglenophyceae and Glaucophyceae were represented by single taxa each. Similarly, among the genera, *Scenedesmus* had the highest species number (i.e, eight taxa) followed by *Coelastrum*, *Cosmarium*, *Euastrum* (three taxa each), *Staurastrum*, *Oscillatoria* (two taxa each) and so on. The detailed list of the algal flora of Gajedi lake is presented in Table 2.

This preliminary investigation of 10 samples showed that the lake is rich in algae. The mean temperature at the collection sites are very congenial being within the range of 28°C to 34 °C, the pH was in near to neutral (6.5) or neutral (7) with mean conductivity value 315 (ìs/cm) which possibly are the regulating factors for the high number of algal species. Further, the water body of the lake is nutritionally rich due to loading of anthropogenic wastes and it may be the reason behind the excessive growth of algal taxa.

Taxonomic description

Each taxon is appended with author/s name, figure number (in parenthesis), reference used for identification followed by brief morphological characters.

1. ***Chroococcus minutus*** **(**Kützing) Nägeli (Fig. 2: 1-2)
Desikachary 1959, P. 103, Pl. 24, Figures 4, 15; Rai and Mishra 2010, P. 123, Pl. 1, Fig. 5.

Cells spherical or oblong, solitary or in groups of 2-4, light blue-green; sheaths not lamellated, colourless; colonies 15-20 µm long, 10-13 µm broad; cells diameter 6-15 µm with sheath, 4-10 µm without sheath

2. ***Oscillatoria princeps*** Vaucher ex Gomont (Fig. 2: 3)
Desikachary 1959, P. 210, Pl. 37, Figures 1, 10, 11, 13, 14; Rai and Mishra 2010, P. 2, Pl. 1, Fig. 10-11.

Trichomes usually straight, not constricted at the cross walls, mostly forming a thallus, blue-green, or more or less brownish; end-cells flatly rounded, slightly capitate; Trichomes (15-) 20-50 (-80) µm broad; cells (2-) 2.5-6.5 (-8.7) µm long.

3. ***Oscillatoria*** **sp. (**Fig. 2: 4)
Wehr and Sheath 2003, P.155, Fig. 16.

Trichomes straight or slightly irregularly undulate, motile by gliding or oscillating; sheaths absent in vegetative state; endsscrew-like coiled.

4. ***Phormidium*** **sp.** (Fig. 2: 5)
Wehr and Sheath 2003, P.141, Fig. 12A.

Filaments arranged in tufts, not in fascicles, forming flat, slimy mats; filaments vary in curvature, without pseudo-branches, usually entangled, slightly too strongly waved or loosely and irregularly screw-like coiled; sheaths facultative.

5. ***Spirulina princeps*** West and G.S. West (Fig. 2: 6)

Desikachary 1959, P. 107, Pl. 36, Figure 7; Rai and Mishra 2010, P.130, Pl. 2, Fig. 4.

Trichomesshort, regularly spirally coiled, blue-green; spirals 11-12 µm broad, 9.5-11 µm distant; trichomes 4.5-5 µm broad.

6. ***Sphaerocystis schroeteri*** Chodat (Fig. 2: 7)
Prescott 1951, P. 83, Pl. 3, Figures 6, 7; Godar and Rai 2018, P.4, Fig. 3.

Colonies more or less spherical, consists of both undivided and recently divided cells; cells arranged in small spherical clusters within the colonial envelope; colonies up to 500 µm in diameter; cells 6-20 µm in diameter.

7. ***Oedogonium* sp.** (Fig. 2: 8-9)
Shrestha and Rai 2017, P. 47, Pl. 1, Fig. 15.

Filaments solitary, unbranched; vegetative cells cylindrical, capitate, with numerous pyrenoids; basal cell with holdfast; terminal cell obtuse.

8. ***Pediastrum tetras*** (Ehrenberg) Ralfs (Fig. 2: 10-11)
Philipose 1967, P. 128, Fig. 45.

Colonies oval, composed of 4-8-16 (-32) cells, without intercellular spaces; marginal cells blobbed divide by a linear incision up to the middle of the cell, each lobe truncates or further divided into two lobes; colonies 20-33 µm in diameter; cells 8.5 µm long, 9.5 µm broad.

9. ***Coelastrum astroideum*** De Notaris (Fig. 2: 12)
Halder 2016, P. 50, Pl. 1, Fig. 2.

Coenobium spherical, composed of 8-16 cells; sheaths gelatinous, delicate; cells oval, smooth walled, closely interconnected by gelatinous process; coenobium 38-46 µm in diameter; cells 10-15 µm in diameter.

10. ***Coelastrum cambricum*** W. Archer (Fig. 3: 13-14)
Philipose 1967, P. 230, Fig. 138a.

Colonies spherical, composed of 32 cells; cells spherical, thickened at the poles, with circular or triangular intercellular spaces; colonies up to 70 µm in diameter; cells 6-12 µm in diameter.

11. ***Coelastrum proboscideum*** Bohlin (Fig. 3: 15)
Philipose 1967, P. 229, Fig. 137.

Colonies pyramidal, composed of 4-8-16 cells; cells conical, truncate and six-sided, thickened at the poles, with large and polygonal intercellular spaces; colonies 17-30 µm in diameter; cells 8-11 µm in diameter.

12. ***Scenedesmus abundans*** (O.Kirchner) Chodat (Fig. 3: 16)
Rai 2013, P. 28, Fig. 23-25.

Colonies composed of 2-4 cells; cells ovoid; cells arranged in a linear series, external cells with one or more median lateral spines in addition to spines from four corners of the colony, internal cells with 1-2 spines from their poles, or rarely without spines; cells 6-15 µm long, 2-7 µm broad; spines 3.5-8 µm long.

13. ***Scenedesmus acuminatus*** (Lagerheim) Chodat (Fig. 3: 17)
Philipose 1967, P. 251, Fig. 161.

Colonies composed of 4-8 cells, forming a flat plate; external cellscurved, internal cells lunate, cell wall smooth and without teeth or spines; cells 12-48 µm long, 2-7 µm broad.

14. ***Scenedesmus bijugatus*** Kützing (Fig. 3: 18)
Philipose 1967, P. 252, Fig. 164 c, e, f.

Colonies composed of 2-4 cells, arranged in a single linear series; cells oblong or ellipsoidal to ovoid, end broadly rounded; cells 7-23 µm long, 3.5-7 µm broad.

15. ***Scenedesmus bijugatus*** var. ***alternanans*** (Fig. 3: 19)
Philipose 1967, P. 256, Fig. 164 g.

Colonies flat, composed of 4-8 cells, arranged in an alternating series; adjacent cells adnate to each other along a short portion of their length only; cells 13-16 µm long, about twice as long as broad.

16. ***Scenedesmus bijugatus*** var. ***gravenitzii*** (C.Bernard) Philipose (Fig. 3: 20)
Rai 2013, P.30, Fig. 5, 19.

Colonies flat, composed of 4-8 cells, arranged in alternate series with adjacent cells in contact only along a short portion of their length; cells fusiform, ellipsoid, oblong-ellipsoid to ovoid with obtuse apices, without teeth and spines; cells 11-16 µm long, 4.5-6.5 µm broad.

17. ***Scenedesmus quadricauda*** var. ***longispina*** (Chodat) (Fig. 3:21)
Rai 2013, P.35, Fig. 13.

Colonies flat, composed of 2-4 cells; cells ovoid to cylindrical, external cells with spines, internal cells without spines; spines slightly longer than the length of the cells; cell wall smooth; cells 15 µm long, 5 µm broad; spines 13-15 µm long.

18. ***Scenedesmus longus***Meyen (Fig. 3: 22)
Philipose 1967, P. 273, Fig. 180 a.

Colonies flat, composed of 2-4-8 cells,arranged in a single linear series;cells ovoid to oblong cylindrical with rounded or sometimes subacute poles; cells 8-19 µm long, 2.3-8 µm broad; spines1.5-15 µm long.

19. ***Scenedesmus* sp.** (Fig. 3: 23)
Wehr and Sheath 2003, P.298, Fig. 24B.

Colonies flat, composed of 2-4-8-16(-32) cells, arranged in linear or alternating series; cells ellipsoidal, ovoid, or crescent-shaped or tapering toward each end; cells not ending in spines.

20. ***Ankistrodesmus* cf. *falcatus*** (Corda) Ralfs (Fig. 3: 24)
Philipose 1967, P. 211, Fig. 221.

Colonies composed of fasciculate bundles of 2-4-8 cells; cells acicular to narrowly fusiform, with tapering to acute apices; cells 20-165 µm long, 1.5-7 µm broad.

21. ***Glaucocystis nostochinearum*** Itzigsohn (Fig. 3: 25)
Philipose 1967, P. 188, Fig. 101-102.

Colonies composed of 4-8 cells, enclosed within the old mother cell wall; cells oblong-ellipsoid, with a number (less than 20) of radiating chromatophore-like bodies inside; colonies 45-50 µm long, 15-20 µm broad; cells 15-30 µm long,10-18 µm broad.

22. ***Closterium ralfsii* Brébisson** ex Ralfs (Fig. 3: 26)
West and West 1904, P. 182, Pl. 24, Fig. 6, 7.

Cells large, moderately curved, yellow-brown or reddish-brown in colour; outer margin about 35° of arc, inner margin much inflated for over half the length of the cell; apices obtuse; cell wall finely striated, 28-33 striae visible across the cell; cells 6-8 times longer than their diameter.

23. ***Cosmarium impressulum*** Elfving (Fig. 3: 27)
Hirano 1969, P. 31 Pl. 2, figure 4; Croasdale and Flint 1988, P. 71, Pl. 40, Fig. 16-19.

Cells constricted; sinus deep and closed; semicells subcircular, with 8 even marginal crenae, the two basal crenae being in a straight line with those of the opposite semicell; apex retuse; cell wall fairly thick, often thicker at apex, closely punctate; cell 27-37 µm long, 13-18 µm broad; isthmus 3-10 µm thick.

24. ***Cosmarium* cf. *seelyanum*** Wolle (Fig. 3: 28)
Wolle 1883, P. 16, Pl. XXVII, Fig. 14.

Cell small, quadrangular, deeply constricted; sinus narrow, liner, with rounded notch in the middle of the sides; cell 25-30 µm in diameter; semicells twice as wide as long.

25. ***Cosmarium subprotumidum*** var. *gregoryi* West and G.S.West (Fig. 4: 29)
Mhaske and Talwankar 2018, P. 21, Fig. 2d.

Cell small, upper half narrowed to broadly truncate apex; cell wall with somewhat radially arranged granules within the margin in pair above but single further away, center with tumour above the isthmus, consisting of relatively larger granules disposed in irregular vertical series; cell 28 µm long, 21 µm broad.

26. ***Euastrum bidentatum*** Nägeli (Fig. 4: 30)
Rai and Mishra 2008, P. 49, Pl. 2, Fig. 4.

Cells deeply constricted; sinus slightly dilated at the extremity; apical angles with a short spine; lateral lobes bilated, marginal spines not distinct; semicells 3-lobed, polar lobe 19 µm broad with deep median incision, with 5 protuberances, one large just above the isthmus, one on each lateral lobe and one on each side of apical notch in the polar lobe; isthmus narrow; cells 44 µm long, 30 µm broad; isthmus 6-7 µm wide.

27. *Euastrum lacustre* (Messikommer) Coesel (Fig. 4: 31-33)

Reddy and Chaturvedi 2017, P. 32, Fig. 10.

Cells medium sized; sinus narrow linear with dilated apex; semicells nearly quadrangular, with deeply incised three lobed, polar lobe rectangular with rounded angles and truncate retuse apex, lateral lobes short with rounded angles and retuse margin, retuse margin some time more incised and giving appearance of two more lobes; cell wall smooth; cell 28-48µm long, 26-46 µm broad; Isthmus 9-10µm wide.

28. *Euastrum platycerum* Reinsch (Fig. 4: 34)

Rai and Mishra 2008, P.50, Pl. 2, Fig. 7.

Cells deeply constricted; semicells 3 lobed, polar lobes 12.5-13 µm broad, truncate without

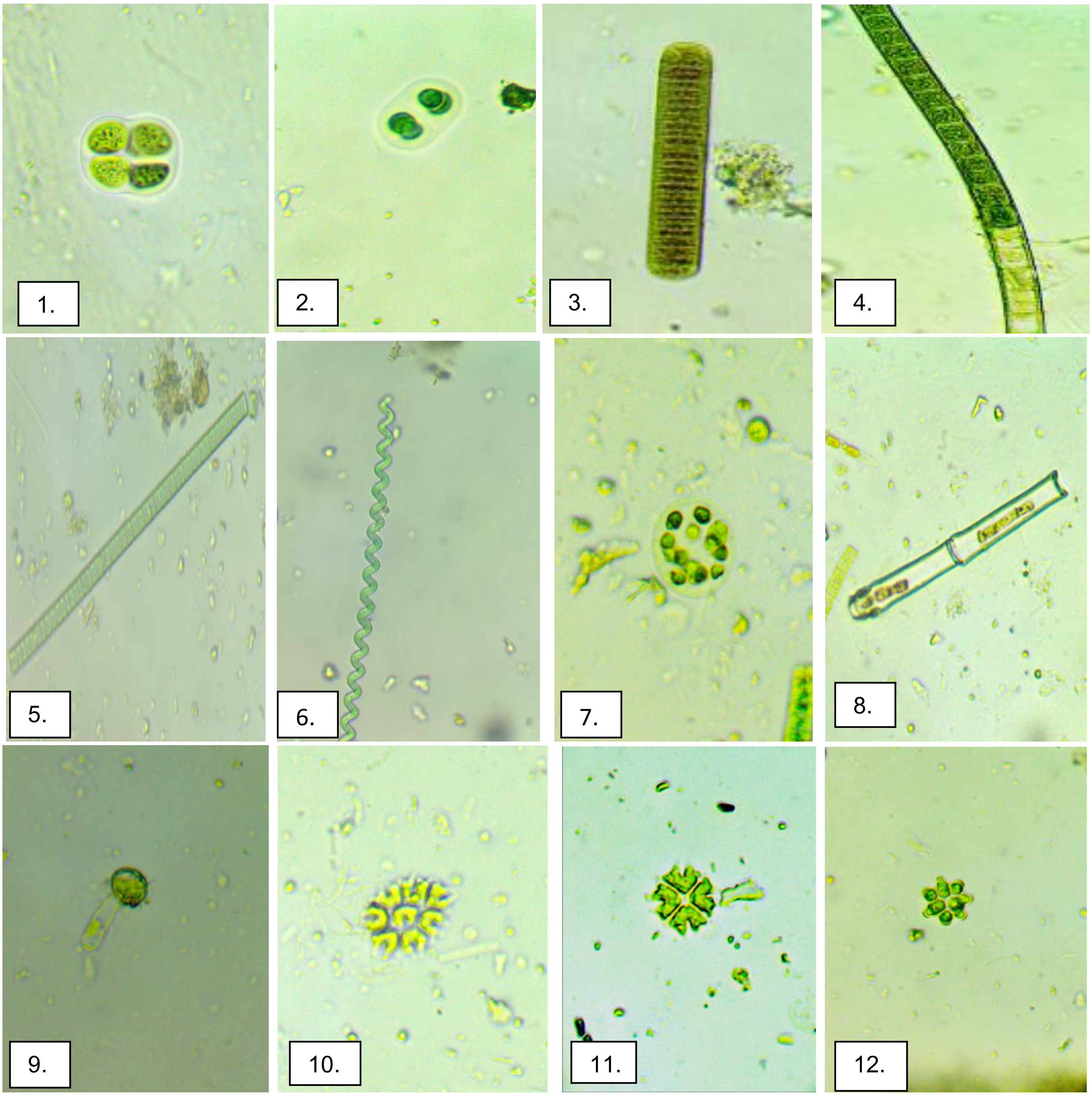

Figure 2: 1-2. *Chrococcus minutus*,3. *Oscillatoria princeps*,4. *Oscillatoria* sp., 5. *Phormidium* sp., 6. *Spirulina princeps*,7. *Sphaerocystis schroeteri*,8-9. *Oedogonium* sp., 10-11. *Pediastrum tetras*, and12. *Coelastrum astroideum*.

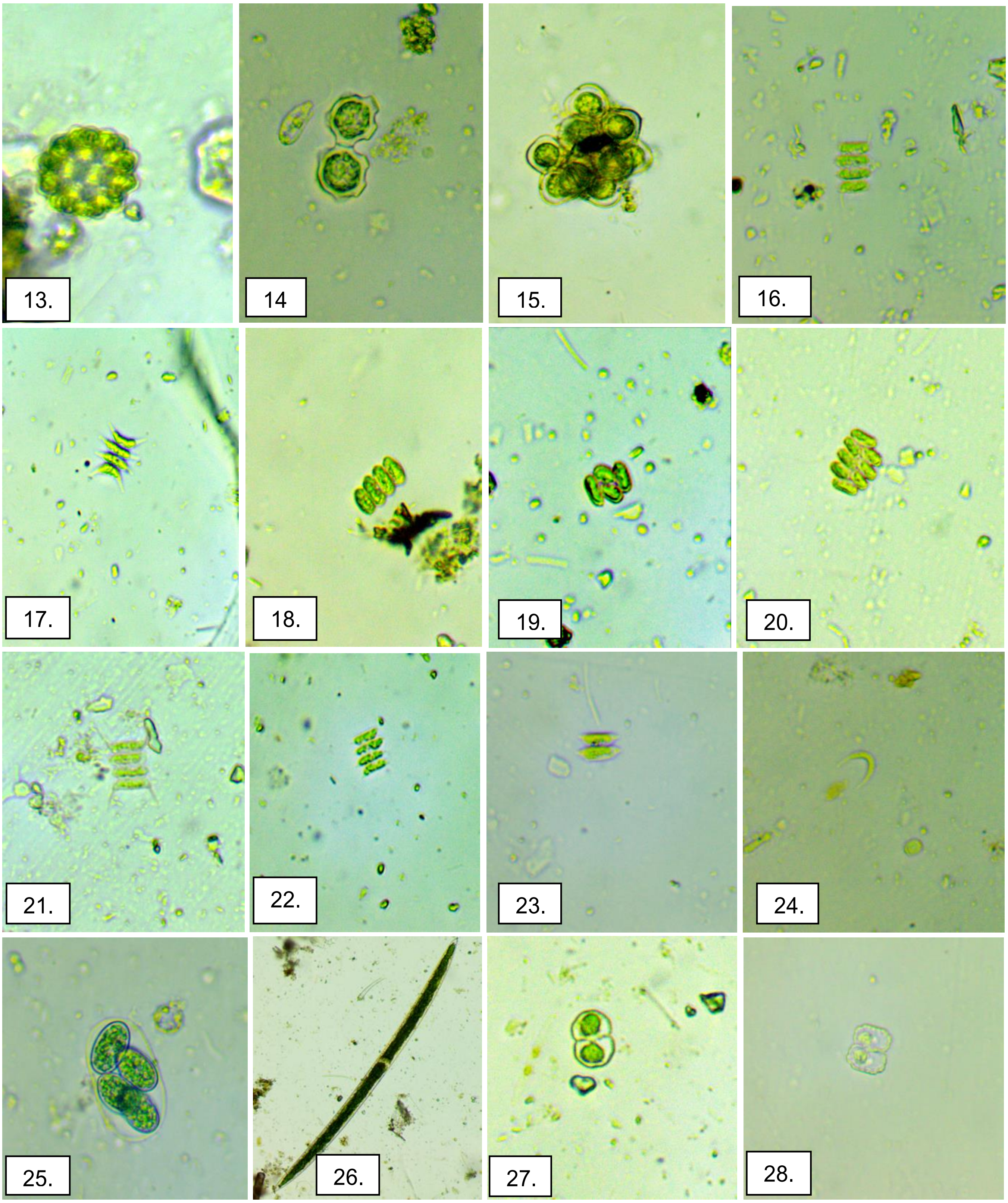

Figure 3: 13-14. *Coelastrum cambricum*, 15. *Coelastrum proboscideum*,16. *Scenedesmus abundans*,17. *Scenedesmus acuminatus*,18. *Scenedesmus bijugatus*,19. *Scenedesmus bijugatus* var. *alternanans*,20. *Scenedesmus bijugatus* var. *gravenitzii*, 21. *Scenedesmus quadricauda* var. *longispina*, 22. *Scenedesmus longus*,23. *Scenedesmus* sp., 24. *Ankistrodesmus* cf. *falcatus*, 25. *Glaucocystis nostochinearum*,26. *Closterium ralfsii*, 27. *Cosmarium impressulum*, and 28. *Cosmarium* cf. *seelyanum.*

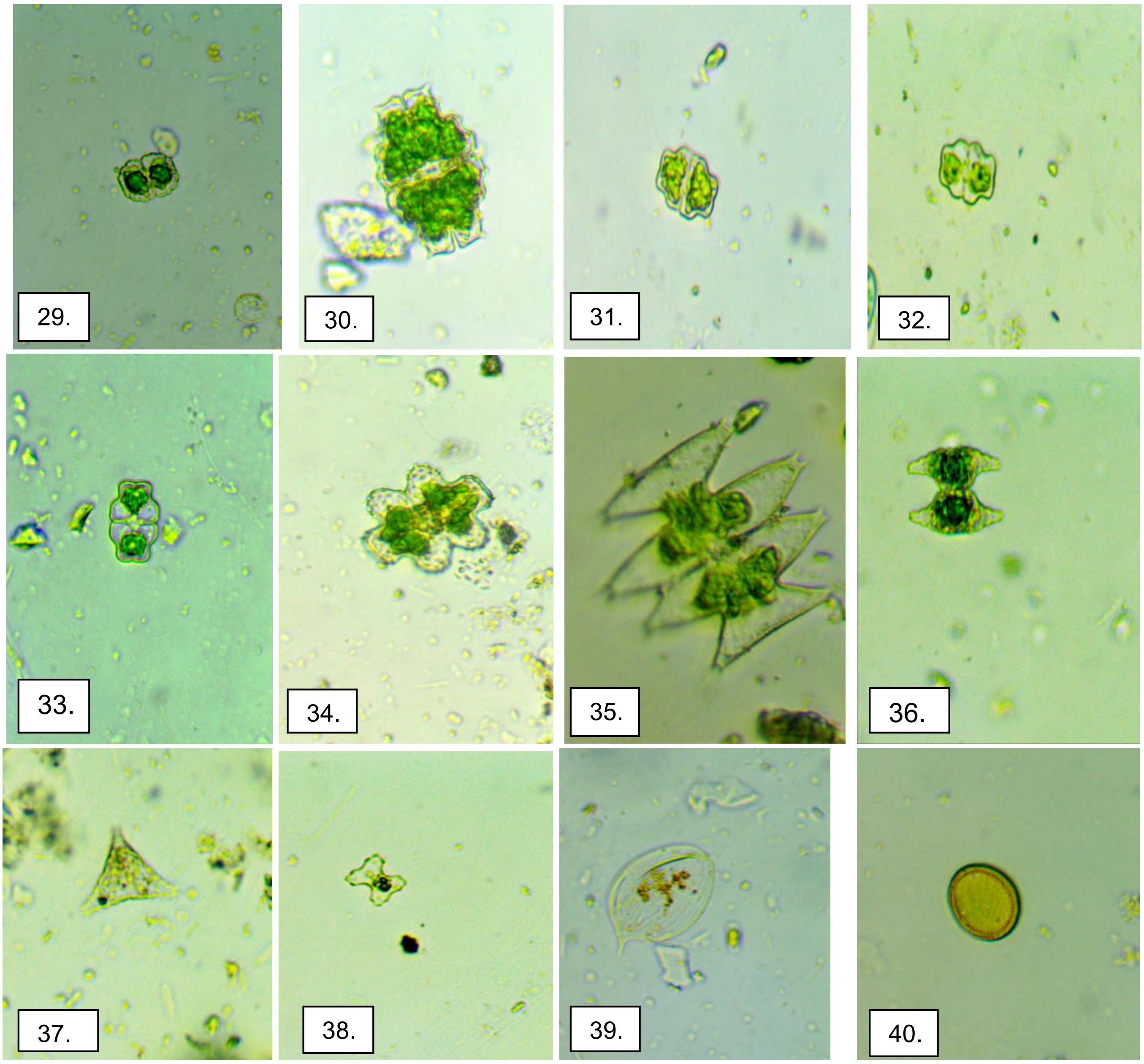

Figure 4: 29. *Cosmarium subprotumidum* var. *gregoryi*, 30. *Euastrum bidentatum*, 31-33. *Euastrum lacustre*, 34. *Euastrum platycerum*, 35. *Micrasterias pinnatifida*, 36. *Staurastrum manfeldtii*, 37-38. *Staurastrum* sp., 39. *Phacus* sp., and 40. *Trachelomonas* cf. *oblonga*.

median constriction, broadly rounded angles with 2 small marginal spines, lateral lobes broadly rounded with 5 small marginal spines; semicells with a rounded central protuberance just above the isthmus; cells 40-42 µm long, 37-37.5 µm broad; isthmus 8-10 µm wide.

29. ***Micrasterias pinnatifida*** Ralfs (Fig. 4: 35) Prescott and Scott 1952, P.244, Pl. 7, Fig. 6.

Polar lobe relatively broader at the apex, margin broadly convex and the lobules slightly converging, relatively less distance between apices and upper basal lobules; cells 60 µm long, 69 µm broad.

30. ***Staurastrum manfeldtii*** Delponte (Fig. 4: 36) Godar and Rai 2018, P. 9, Fig. 69-71.

Cellswith processes; semicells with swollen base; apex convex with row of emarginated verrucae; processes tapered, slightly convergent, spinulose end with 3-4 spines; cells 37-58 µm long, 33-100 µm broad; isthmus 13-15 µm wide.

31. ***Staurastrum* sp.** (Fig. 4: 37-38)
Wehr and Sheath 2003, P.377, Fig. 33, 34, 87–89, 92, and 93.

Cells are small to large, 2 to 12 radiate in end view, with a shallow or deep median constriction; semicells have long hollow processes.

32. ***Phacus* sp.** (Fig. 4: 39)
Wehr and Sheath 2003, P.413, Fig. 13.

Cells laterally flattened, leaf-shaped, and perhaps twisted as well.

33. ***Trachelomonas* cf. *oblonga*** Lemmermann (Fig. 4: 40)
Wo³owski and Grabowska 2007, P. 210, Fig. 23, 43, 44.

Lorica oblong, slightly narrowed at the posterior end, smooth; cells 12.5-13.0 µm long 10.5-12.6 µm wide; apical pore 1.5 µm diameter without collar.

Conclusion

This study is a pioneer work on the algal flora of Gajedi Lake which showed that the lake is rich in algal flora. But the data collected from this study alone was not sufficient to determine the richness of algal species in the area as well as their variation with different seasons. Therefore, further studies are necessary to documents common as well as interesting algae in the lake and to find out the seasonal variation of algae.

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References

Croasdale, H. & Flint, E.A. (1988). *Flora of New Zealand, Fresh water algae, chlorophyta, desmids with ecological comment on their habitats* (pp. 147). Vol. II. Botany Division, D.S.I.R., Christchurch, New Zealand.

Deshikachary, T.V. (1959). *Cyanophyta.* I.C.A.R. monograph in algae, New Delhi.

District Forest Office. (2073 B.S.). Annual Progress Review Report, Detailed introduction and Progress account fiscal Year 2072/073, Rupandehi, Nepal: Author.

Dhakal, K.S., Saud, D.S. & Joshi, N (2019). Wetland flora of Rupandehi District, Nepal. *Journal of Plant Resources*, *17(*1), 58-68.

Godar, K., & Rai, S.K. (2018). Freshwater green algae from Raja-Rani wetland, Bhogateni-Letang, Morang, Nepal. *Journal of Plant Resources*, *16*(1), 1-17.

Guiry, M.D., & Guiry, G.M. (2019). *Algae Base*. World-wide electronic publication, National University of Ireland, Galway. http://www.algaebase.org; searched on December 2019.

Halder, N. (2016). Taxonomy and Periodicity of *Chlorococcum* Fries, *Coelastrum* Näg. and *Scenedesmus* Meyen in Hooghly, West Bengal, India. *Mesop. Environ. J.*, *2*(2), 47-56.

Hirano, M. (1955). Fresh water algae. *In*: H. Kihara (Eds.), *Fauna and flora of Nepal Himalaya* (pp. 5- 42), Kyoto University, Kyoto, Japan: Fauna and Flora Research Society.

Hirano, M. (1963). Fresh water algae from the Nepal Himalaya, collected by a member of the Japanese Climbing Expedition. *Contr Biol Lab, Kyoto Univ*, Japan, *16*, 1-23.

Hirano, M. (1969). Fresh water algae from Langtang Himal, Nepal Himalaya. *Contr. Biol. Lab., Kyoto Univ., Japan*, *22,* 1-42.

Mhaske, T.K. & Talwankar D.S. (2018). Occurrence of *Cosmarium* species in Khadakpurna reservoir, Buldana district – Maharashtra, India. *GSC Biological and Pharmaceutical Sciences*, *5*(3), 20-24.

Phiipose, M.T. (1967). *Chlorococcales: Monograph on algae*. New Delhi, India: Indian Council of Agricultural Research.

Prescott, G.W. (1951). *Algae of the Western Great Lakes area* (pp. 977). Dubuque, Iowa: WM.C. Brown Publishers.

Prescott, G.W. & Scott, A.M. (1952). The algal flora of southeastern United States. V. Additions to our knowledge of the desmid genus *Micrasterias*. *Transactions of the American Microscopical Society*, *71*, 229-252.

Rai, S.K. & Misra P.K. (2008). On some desmids from Koshi Tappu wildlife reserve, Nepal. *Ecoprint*,*15,* 47-58.

Rai, S.K. (2013). Occurrence of genus *Scenedesmus* Mayen (Chlorophyceae) from East Nepal. *Nepalese Journal of Biosciences*, *3*, 26-37.

Rai, S.K. & Misra, P.K. (2010). Freshwater cyanophyceae from east Nepal. *Bangladesh J. of Plant Taxonomy,17*(2), 121-139.

Rai, S.K. & Paudel, S. (2019). Algal flora of Jagadishpur Tal, Kapilvastu, Nepal. *Journal of Plant Resources, 17*(1), 6-20.

Rai, S.K. & Rai, R.K. (2012). Some interesting freshwater algae from Chimdi Lake including a new record for Nepal. *Nepalese Journal of Biosciences, 2*, 118-125.

Rajopadhyaya, R., Joshi, S., Shrestha, S. & Rai, S.K. (2017). Some new and interesting cyanobacteria from Baghjhoda pond, eastern Nepal. *Himalayan J. of Science and Technology, 1*, 1-8.

Reddy, M. & Chaturvedi, A. (2017). Desmids from the rivers of Chandrapur district, Maharashtra. *An international journal of environment and biodiversity,8*(1), 25-34.

Shrestha, G. & Rai, S.K. (2017). Algal flora of Rajarani Lake, Dhankuta and their seasonal distribution. *Our Nature*, *15* (1-2), 44-54.

Thapa, M.S. & Poudel, G. (2018): Assessing the coverage of Urban Green Space in Butwal Sub-Metropolitan City, Nepal: A GIS based approach. *Journal of Institute of Forestry*, 15, 77-86.

Watanabe, M. (1995). Algae from lake Rara and its vicinities, Nepal Himalayas. *In*: H. Watanabe and H. Hagiwara (Eds.), *Crytogams of the Himalaya: Nepal and Pakistan* pp. 1-17, Vol. 3. Tsukub Japan: Department of Botany, National Science Museum.

Wehr, J. & Sheath, R.G. (2003). *Freshwater Algae of North America: Ecology and Classification* (pp. 917): Academic Press.

West, W. & West, G.W. (1904). *A monograph of the British desmidiaceae*. Vol. 1, London: Ray Society

Wolle, F. (1883). Fresh-water algae. VII. *Bulletin of the Torrey Botanical Club, 10*(2), 13-21.

Wo³owski, K. & Grabowska, M. (2007). *Trachelomonas* species as the main component of the euglenophyte community in the Siemianówka Reservoir (Narew River, Poland). *Ann. Limnol. - Int. J. Lim,43* (3), 207-218.

Table 2: Algal taxa reported from Gajedi Lake, Rupandehi district (Classification is based on Guiry & Guiry, 2019)

Phylum	Class	Order	Family	Genus	Algal taxa
Cyanobacteria	Cyanophyceae	Chroococcales	Chroococaceae	*Chroococcus*	1. *Chroococcus minutes* (Kützing) Nägeli
		Oscillatoriales	Oscillatoriaceae	*Oscillatoria*	2. *Oscillatoria princeps* Vaucher ex Gomont
					3. *Oscillatoria* sp.
			Oscillatoriaceae	*Phormidium*	4. *Phormidium* sp.
		Spirulinales	Spirulinaceae	*Spirulina*	5. *Spirulina princeps* West and G.S.West
Chlorophyta	Chlorophyceae	Chlamydomonadales	Sphaerocystidaceae	*Sphaerocystis*	6. *Sphaerocystis schroeteri* Chodat
		Oedogoniales	Oedogoniaceae	*Oedogonium*	7. *Oedogonium* sp.
		Sphaeropleales	Hydrodictyaceae	*Pediastrum*	8. *Pediastrum tetras* (Ehrenberg) Ralfs.
		Sphaeropleales	Scenedesmaceae	*Coelastrum*	9. *Coelastrum astroideum*De Notaris.
					10. *Coelastrum cambricum* W.Archer.
					11. *Coelastrum proboscideum* Bohlin
				Scenedesmus	12. *Scenedesmus abundans*(O.Kirchner) Chodat
					13. *Scenedesmus acuminatus* (Lagerheim) Chodat
					14. *Scenedesmus bijugatus* Kützing
					15. *Scenedesmus bijugatus* var. *alternanans*
					16. *Scenedesmus bijugatus* var. *gravenitzii* (C.Bernard) Philipose
					17. *Scenedesmusquadricauda* var. *longispina* (Chodat) G.M.Smith
					18. *Scenedesmus longus* Meyen
					19. *Scenedesmus* sp.
			Selenastraceae	*Ankistrodesmus*	20. *Ankistrodesmus* cf. *falcatus* (Corda) Ralfs
Glaucophyta	Glaucophyceae	Glaucocystales	Glaucocystaceae	*Glaucocystis*	21. *Glaucocystis nostochinearum* Itzigsohn
Charophyta	Conjugatophyceae	Desmidiales	Closteriaceae	*Closterium*	22. *Closterium ralfsii* Brébisson ex Ralfs
			Desmidiaceae	*Cosmarium*	23. *Cosmarium impressulum* Elfving

Phylum	Class	Order	Family	Genus	Algal taxa
					24. *Cosmarium* cf. *seelyanum* Wolle
					25. *Cosmarium subprotumidum* var. *gregoryi* West and G.S.West
				Euastrum	26. *Euastrum bidentatum* Nägeli
					27. *Euastrum lacustre* (Messikommer) Coesel
					28. *Euastrum platycerum* Reinsch
				Micrasterias	29. *Micrasterias pinnatifida* Ralfs
				Staurastrum	30. *Staurastrum manfeldtii* Delponte
					31. *Staurastrum* sp.
Euglenophyta	Euglenophyceae	Euglenida	Phacidae	*Phacus*	32. *Phacus* sp.
			Euglenidae	*Trachelomonas*	33. *Trachelomonas* cf. *oblonga* Lemmermann

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Post-Monsoon Macrofungal Diversity in Lumbini Collaborative Forest, Rupandehi District, Central Nepal

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Abstract

The study was carried out for higher fungi especially mushrooms, found in Lumbini collaborative forest, Rupandehi district, Central Nepal. Total of 31 mushroom species including both Ascomycetes (5 species) and Basidiomycetes (26 species) mushrooms were collected. Polyporales was found to be the dominant order in the study area with 16 species followed by Xylariales (5 species) and Hymenochaetales (4 species). *Terminalia alata* was found to be major host plant for harboring 10 different mushroom species (including 1 Ascomycetes and 9 Basidiomycetes species) followed by *Shorea robusta* (7 species).

Keywords: Ascomycetes, Basidiomycetes, Macrofungi, Mushroom, Substrate

Introduction

There are numerous macrofungi that generally produce fleshy or corky fruiting bodies commonly known as mushrooms and grow either above ground (epigenous) or underground (hypogenous) in nature (Pacioni, 1981) belonging to group Ascomycetes and Basidiomycetes. Ascomycetes, the Sac fungi, are the largest group of all the higher fungi having well developed, branched and septate mycelium like in Basidiomycetes. They produce sexual non motile spores i.e. ascospores (usually eight in number) endogenously enclosed within the microscopic sac like cell known as the ascus (pl. asci); the asci in most genera are arranged in a definite group within a fruiting body, the ascocarp. They are mostly terrestrial occurring as saprophytes or parasites (Alexopoulos & Mims, 1979). The cup fungi, the morels, and the truffles are among the best known examples of Ascomycetes. Basidiomycetes, the Club fungi, are thought to have evolved from an ascomycetous ancestor and are the most advanced of all the fungi (Alexopoulos & Beneke, 1962). They comprise the second biggest class of fungi which includes most of the large and conspicuous species mostly saprophytic found in fields and woods like mushroom & toadstools (collectively called agarics), jelly fungi, bracket fungi or polypores, puff-balls, coral fungi, earth-stars, bird's-nest fungi, stink-horns, etc. to micro-fungi like rusts and smuts which are obligate plant parasites and some parasitic (Acharya & Parmar, 2016). Mushrooms generally prefer to grow under humid condition. Different terms have been given to signify their habitats. For example: species growing on grasslands are known as praticolous; on woodland- silvicolous; on wood, woody debris, trees, stumps, rotten or burnt wood- lignicolous; on dung- coprophilous; amongst moss- muscicolous; on leaf litter- humicolous and so on (Purkaryastha & Chandra, 1985).

Generally post monsoon macrofungus are mostly the members of order Polyporales and some members of Ascomycetes. In Polyporaceae some species have a toadstool form, particularly species of *Boletus*, a large and abundant genus of fleshy toadstools. However, most of Polyporaceae are wood- inhabiting fungi forming sporophores as shelves or brackets on the trunks of living trees, or on dead branches of living tree or on fallen branches or log lying on the ground or on the stump. The fruiting bodies of Polyporaceae are commonly rather leathery or corky, and only rarely fleshy (Gold, 1975).

Nepal, a well famed country for mycodiversity, with its wide range in ecological conditions from the tropical Terai to the permanent snow at the highest elevation (phytogeographic factors) have played an interesting role in the distribution of diverse mycofloral components (Adhikari, 1994-95, 2000, 2009, 2014c). Till now, 34 endemic species of mushroom have been described from Nepal (Devkota & Aryal, 2020). So far, about 1,291 mushroom species have been recorded from Nepal ((Devkota & Aryal, 2020)). Among these about 159 species are said to be edible (Devkota & Aryal 2020) while 100 species are poisonous and 73 species have medicinal values (Adhikari, 2014). The Nepalese mycoflora are under process of exploration since the work of Lloyd (1808) and Berkely (1838) but still several parts of Nepal await their exploration, investigation, study and publication (Adhikari, 1999, 2000, 2009).

Intense mycological exploration and investigation, though, is carried out more in Central Nepal as compared to eastern and western regions of Nepal (Adhikari, 1999, 2000; Adhikari & Bhattarai, 2014), present study was carried out to document the higher fungi of both Ascomycetes and Basidiomycetes species from Lumbini collaborative forest, Rupandehi district (Central Nepal) which was still unexplored.

Materials and Methods

Study area

Lumbini collaborative forest (Terai sal forest) lie in Kanchan rural municipality-5, Fayarlayan, Rupandehi district. Rupandehi district is a part of the Terai region of Central Nepal and covers a total area of 130,522 ha in which 6,512 ha. lies in Terai and 18,593 ha. in Churiya region (Department of Forest Research and Survey [DFRS], 2015). It has about 73% land is agricultural land, urban areas, and roads, 23% forest, and the remaining 4% water resources (District Development Committee [DDC], 2007). Present research was carried out in one compartment (along east-west highway) of Lumbini collaborative forest, Fayarlayan, Rupandehi district. The forest is spreading over 1,321 ha where 204 ha. lies in steep churia range and remaining area in plane Terai region of Nepal. The forest is divided into eight compartments for scientific forest management. The forest is mainly dominated by Sal (*Shorea robutsa*) with its associated species like Saj (*Terminalia*

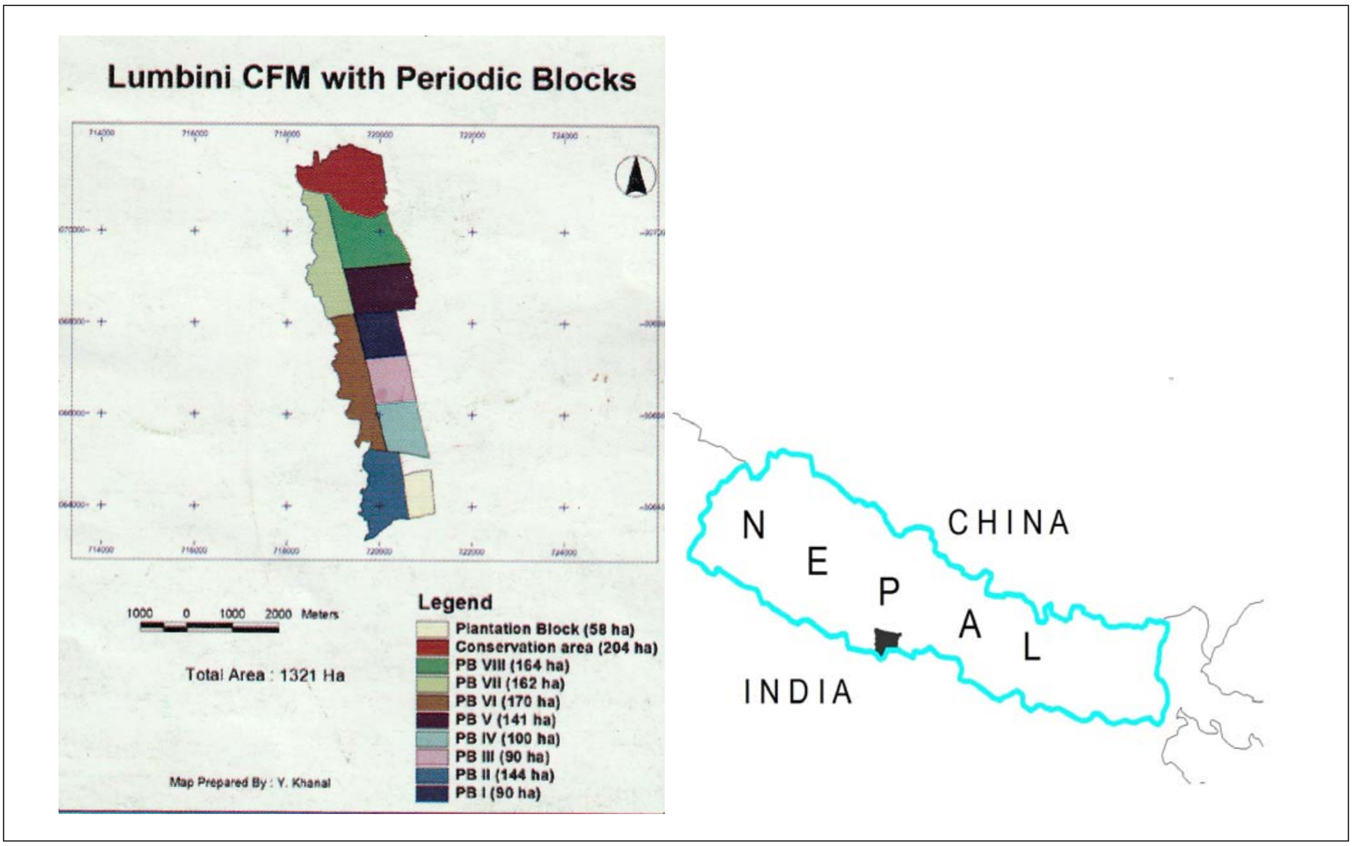

Figure 1: Map of the study area

alata), Banjhi (*Anogeissus latifolia*), *Adina cordifolia*, etc. The climate of the area is typically tropical dominated by the southeast monsoon. A hot climate generally prevails throughout the year except in the short winter.

Collection and identification

The study area was surveyed in October 2019. One compartment (along east-west highway) of Lumbini collaborative forest was extensively explored for Ascomycetes and Basidiomycetes mushroom from October 10 to 11, 2019. Altogether 31 species of fungal species including both Ascomycetes (5 species) and Basidiomycetes fungi (26 species) were collected from nature in the study area. The species collected were well air dried in the shade and packed in paper envelops with proper tag/collection numbers. The species found in soil were collected carefully by digging with the help of a digger. Other specimens which were found to grow on fallen or rotten branches/wooden logs, branches or trunks of dying or dead plants; or trunks of living plants were collected along with their host plant by cutting with the help of saw. During collection, at least one fruiting body (sporocarp) i.e. ascocarp or basidiocarp was left for their spore dispersal.

Photographs of all the mushroom specimens were taken in their natural habitat prior to collection. Date of collection, altitude, the nature of habitat/substrate, surrounding plant community especially trees, any distinctive odor, any change in color on cutting or bruising when fresh, color of latex (if present), whether growing solitary or in groups was recorded. Most of the host plants or substrates were identified in the field and for unidentified ones like piece or branch of wood or fallen leaf litter, etc. their sample were collected for identification in the herbarium. The paper envelops with the collected fungi were brought to National Herbarium and Plant Laboratories (KATH), Godawari for identification and making herbarium specimens. Identified mushroom specimens are housed in National Herbarium and Plant Laboratories, Godawari, Lalitpur. The identification was done following key identifying taxonomical characters of relevant literatures (Teng, 1939; Walting, 1973; Alexopoulous & Mims, 1979; Dickson & Lucas, 1979; Pacioni, 1981; Dennis, 1981; Svréek, 1983; Miller, 1984; Purkayastha & Chandra, 1985; Adhikari, 2014). It was also identified by tallying photographs of the relevant literatures and cross checking the collected specimens to that of identified herbarium specimens deposited at the herbarium. Some species were also identified seeking the help of expert of Mycology. The nomenclature of all the identified fungal species follows Adhikari (2012, 2014).

Enumeration of species

1. ***Daedaleopsis* sp.** [Polyporales: Polyportaceae]
   Log of *Terminalia alata* Roth, Lumbini collaborative forest, 165 m, 11 October 2019, collection no. 201933, collector- Rajendra Acharya & Bipin Khanal
2. ***Daedaleopsis* sp.** [Polyporales: Polyporaceae]
   Log of *Terminalia alata* Roth, Lumbini collaborative forest, 165 m, 11 October 2019, collection no. 201927, collector- Rajendra Acharya & Bipin Khanal
3. ***Daedaleopsis conchiformis*** Imazeki [Polyporales: Polyporaceae]
   Fallen branch of *Shorea robusta* Gaertn., Lumbini collaborative forest, 165m, 11 October 2019, collection no. 201931, collector- Rajendra Acharya & Bipin Khanal
4. ***Daedaleopsis confragosa*** (Bolt.: Fr.) Schr. var. *confragosa* [Polyporales: Polyporaceae]
   Log of *Anogeissus latifolia* (Roxb. ex DC.) Wall. ex Bedd., Lumbini collaborative forest, 165 m, 11 October 2019, collection no. 201926, collector- Rajendra Acharya & Bipin Khanal
5. ***Daldinia concentrica*** (Bolt.: Fr.) Ces. & De Not [Xylariales: Xylariaceae]
   Rotten log of unknown tree, Lumbini collaborative forest, 160 m, 10 October 2019, collection no. 20198, collector- Rajendra Acharya & Bipin Khanal
6. ***Ganoderma lucidum*** (Curt.: Fr.) Karst. [Polyporales: Ganodermataceae]
   On soil at the base of rotten wood, Lumbini collaborative forest, 165m, 11 Oct. 2019, collection no. 201922, collector- Rajendra Acharya & Bipin Khanal

7. ***Hexagonia* sp.** [Polyporales: Polyporaceae]
Wood of unknown tree, Lumbini collaborative forest, 160 m, 11 Oct. 2019, collection no. 201923, collector- Rajendra Acharya & Bipin Khanal
8. ***Inonotus* sp.** [Hymenochaetales: Hymenochaetaceae]
Stump of unknown tree, Lumbini collaborative forest, 165 m, 10 October 2019, collection no. 201915, collector- Rajendra Acharya & Bipin Khanal
9. ***Lenzites betulina*** (L.) Fr. [Polyporales: Polyporaceae]
Log of *Lagerstroemia parviflora* Roxb., Lumbini collaborative forest, 165 m, 11 October 2019, collection no. 201938, collector- Rajendra Acharya & Bipin Khanal
10. ***Laetiporus* sp.** [Polyporales: Polyporaceae]
Fallen branch of *Shorea robusta* Gaertn., Lumbini collaborative forest, 165 m, 11 October 2019, collection no. 201929, collector- Rajendra Acharya Bipin Khanal
11. ***Microporus xanthopus*** Fr. Kuntz [Polyporales: Polyporaceae]
Stump of *Melia azedarach* L., Lumbini collaborative forest, 160 m, 11 October 2019, collection no. 201924, collector- Rajendra Acharya & Bipin Khanal
12. ***Phellinus* gilvus** (Schw.) Pat. [Hymenochaetales: Hymenochaetaceae]
Log of *Shorea robusta* Gaertn., Lumbini collaborative forest, 165 m, 10 October 2019, collection no. 201910, collector- Rajendra Acharya & Bipin Khanal
13. ***Phellinus igniarius*** (L.) Fr. Quèl. [Hymenochaetales: Hymenochaetaceae]
Rotten log of *Shorea robusta* Gaertn., Lumbini collaborative forest, 160 m, 11 October 2019, collection no. 201937, collector- Rajendra Acharya & Bipin Khanal
14. ***Pleurotus* sp.** [Agaricales: Pleurotaceae]
On moist soil, Lumbini collaborative forest, 160 m, 11 October 2019, collection no. 201925, collector- Rajendra Acharya & Bipin Khanal
15. ***Polyporus* sp.** [Polyporales: Polyporaceae]
Log of *Shorea robusta* Gaertn., Lumbini collaborative forest, 165 m, 11 October 2019, collection no. 201921, collector- Rajendra Acharya & Bipin Khanal
16. ***Polyporus* sp.** [Polyporales: Polyporaceae]
Log of *Shorea robusta*, Lumbini collaborative forest, 160 m, 11 October 2019, collection no. 201930, collector- Rajendra Acharya & Bipin Khanal
17. ***Polystictus* sp.** [Hymenochaetales: Hymenochaetaceae]
Rotten log of unknown tree, Lumbini collaborative forest, 165 m, 10 October 2019, collection no. 20194, collector- Rajendra Acharya & Bipin Khanal
18. ***Pycnoporus cinnabarinus*** (Jacq.: Fr.) Karst. [Polyporales: Polyporaceae]
Log of *Terminalia alata* Roth, Lumbini collaborative forest, 165 m, 11 Oct. 2019, collection no. 201919, collector- Rajendra Acharya & Bipin Khanal
19. ***Schizophyllum commune*** (Fr.) Fr. [Agaricales: Schizophyllaceae]
Log of T*erminalia alata* Roth, Lumbini collaborative forest, 165 m, 10 Ocober 2019, collection no. 201912, collector- Rajendra Acharya & Bipin Khanal
20. ***Stereum* sp.** [Russulales: Stereaceae]
Log of *Lagerstroemia parviflora* Roxb., Lumbini collaborative forest, 165 m, 11 Ocober 2019, collection no. 201920, collector- Rajendra Acharya & Bipin Khanal
21. ***Stereum hirsutum*** (Willd.: Fr.) Gray [Russulales: Stereaceae]
Log of *Adina cordifolia* (Roxb.) Hook. f., Lumbini collaborative forest, 160 m, 11 October 2019, collection no. 201922, collector- Rajendra Acharya & Bipin Khanal
22. ***Thelephora* sp.** [Thelephorales: Thelephoraceae]
Log of unknown tree, Lumbini collaborative forest, 160 m, 10 October 2019, collection no. 20193, collector- Rajendra Acharya & Bipin Khanal
23. ***Trametes versicolor*** (L.: Fr.) Pilat [Polyporales: Polyporaceae]
Log of *Terminalia alata* Roth, Lumbini

collaborative forest, 160 m, 10 October 2019, collection no. 201912, collector- Rajendra Acharya & Bipin Khanal

24. ***Trametes lactinea*** (Berk.) Pat. [Polyporales: Polyporaceae]
Log of *Terminalia alata* Roth, Lumbini collaborative forest, 165 m, 11 October 2019, collection no. 201938, collector- Rajendra Acharya & Bipin Khanal
25. ***Trametes* sp.** [Polyporales: Polyporaceae]
Log of *Shorea robusta* Gaertn., Lumbini collaborative forest, 165 m, 10 October 2019, collection no. 20195, collector- Rajendra Acharya & Bipin Khanal
26. ***Tremella fusiformis*** Berk. [Tremellales: Tremellaceae]
Log of *Terminalia alata* Roth, Lumbini collaborative forest, 165 m, 10 October 2019, collection no. 201914, collector- Rajendra Acharya & Bipin Khanal
27. ***Trichaptum byssogenum*** (Jungh.) Ryv. [Polyporales: Polyporaceae]
Log of *Terminalia alata* Roth, Lumbini Collaborative forest, 165 m, 10 October 2019, collection no. 20197, collector- Rajendra Acharya & Bipin Khanal
28. ***Xylaria filiformis*** (Alb. et Schw.: Fr.) Fr. [Xylariales: Xylariaceae]
Leaf litter of *Terminalia alata* Roth, Lumbini collaborative forest, 160 m, 11 October 2019, collection no. 201932, collector- Rajendra Acharya & Bipin Khanal
29. ***Xylaria furcata*** Fr. [Xylariales: Xylariaceae]
Rotten log of *Terminalia alata* Roth, Lumbini collaborative forest, 165 m, 10 October 2019, collection no. 201917, collector- Rajendra Acharya & Bipin Khanal
30. ***Xylaria nigripes*** (KI.) Sacc. [Xylariales: Xylariaceae]
Rotten log of *Shorea robusta* Gaertn., Lumbini collaborative forest, 160 m, 10 October 2019, colletion no. 20199, collector- Rajendra Acharya & Bipin Khanal
31. ***Xylaria polymorpha*** (Pers.: Fr.) Grev. [Xylariales: Xylariaceae]
On soil at the base of rotten wood, Lumbini collaborative forest, 165 m, 10 October 2019, collection no. 201913, collector- Rajendra Acharya & Bipin Khanal

Results and Discussion

Altogether 31 species of fungi including Ascomycetes (5 species) and Basidiomycetes fungi (26 species) were collected from the study area. Out of total 31 identified fungal species, Ascomycetes species were from single order belonging to single family and two genera whereas Basidiomycetes species were from six orders belonging to 8 families and 18 genera.

The distribution of macro-fungal species is low in autumn season and so the exploration of Ascomycetes and Basidiomycetes fungi during autumn resulted in relatively fewer collections. The collected species of Ascomycetes fungi were the member of order: Xylariales with its corresponding family Xylariaceae (see in enumeration of mushroom species). In contrast, most of the collected Basidiomycetes fungi were the members of order Polyporales. Polyporales, the dominant order, in the study area with 15 species was followed by Hymenochaetales (4 species), Agaricales, Russulales (2 species each) (Figure 2). Similarly, Polyporaceae was found to be the dominant family represented by 15 species. It was followed by Hymenochaetaceae (4 species) and followed by Stereaceae (2 species each) and rest of the families was represented by only single Basidiomycetes species (Figure 3).

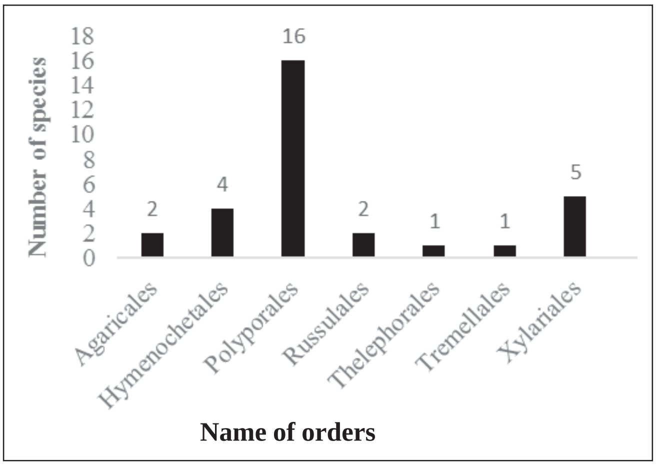

Figure 2: Orders representing number of species in the study area

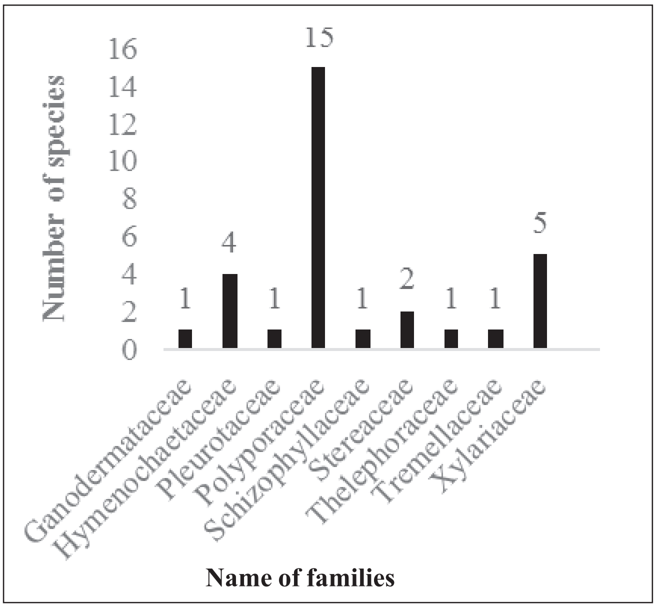

Figure 3: Families representing number of species in the study area

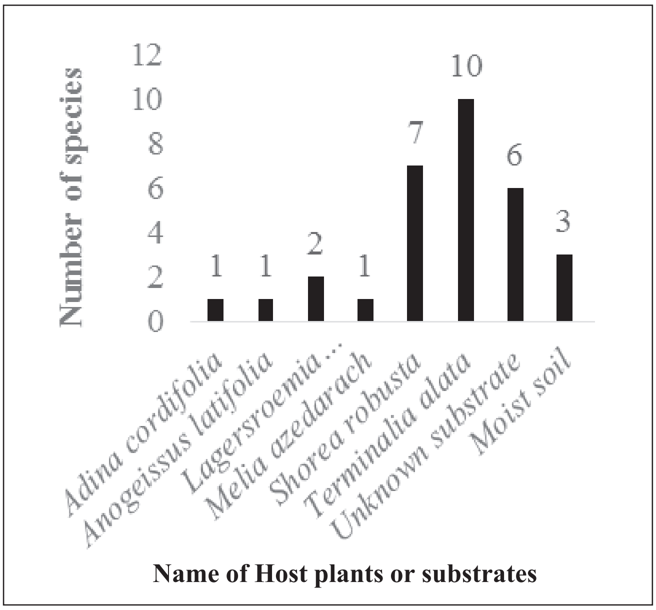

Figure 4: Host plants or substrates harboring number of fungal species in the study area

Polypores were the most common and were found to grow on dead woods, fallen logs, stumps, rotten branches and dead part of trunk and branches of living tree. Out of 31 fungal species, Ascomycetes species like *Daldinia concentrica* (Figure 5), *Xylaria nigripes* and Basidiomycetes species like *Daedaleopsis confragosa* (Figure 9), *Schizophyllum commune* (Figure 6), *Microporus xanthopus, Trametes versicolor* (Figure 7), *Pycnoporus cinnabarinus* (Figure 4) were found to be very common in the study area. Species of *Pycnoporus cinnabarinus, Ganoderma lucidum* and *Trametes hirsuta* reported in the present study area was also reported by Aryal & Budhathoki (2013) at Sankarnagar community forest, Rupandehi district (Central Nepal). *Pycnoporus cinnabarinus, Microporus xanthopus* and *Daldinia concentrica* reported in preset study area was also reported by Pokhrel (2017) at Amrite community forest, Kapilvastu district (Central Nepal).

Terminalia alata was found to be major host plant for harboring 10 different mushroom species (including one Ascomycetes and remaining nine Basidiomycetes species) which is followed by *Shorea robusta* (7 species), *Lagerstroemia parviflora* (2 species) and least by *Adina cordifolia*, *Buchanania latifolia* and *Melia azedarach* with one species only (Figure 4). All the host plants or substrates of the fungal species were identified except six since it was almost old tree stump and log with rotten bark. On the other hand, one species of Ascomycetes fungi belonging to the order Xylariales (Xylariaceae family) and two species of Basidiomycetes fungi belonging to the order Agaricales and Polyporales (Pleurotaceae and Ganodermataceae families respectively) were found to be grown on moist soil (see in enumeration of mushroom species).

Conclusion

A total of 31 fungal species from Ascomycetes (5 species) and Basidiomycetes fungi (26 species) were collected from Lumbini collaborative forest, Rupandehi district, Central Nepal. The identified Ascomycetes species were from single orders belonging to single family and two genera whereas Basidiomycetes species were from six orders belonging to 8 families and 18 genera. In overall, Polyporales and Polyporaceae were the dominant order and family respectively. *Terminalia alata* was found to be the major host plant for 10 different mushroom species (including 1 Ascomycetes and remaining 9 Basidiomycetes species). Beside post monsoon period further mycological exploration should be carried out in monsoon rainy season in all compartment to document the other macrofungal species for determining actual mycodiversity of that forest.

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References

Acharya, R. & Parmar, G. (2016). Preliminary documentation of Basidiomycetous fungi (Polypores and Mushrooms) found in Bardia national park and its buffer zone area, western Nepal. *Bulletin of the Department of Plant resources*, 38, 22–29.

Adhikari, M. K. (1999). Status of wild potential mushrooms in Nepal. Pp.1339–1350. In: *National Conference on Science and Technology March 8-11, 1999*: Nepal Academy of Science and Technology, Kathmandu, Nepal. Proceedings.

Adhikari, M.K. & Bhattarai, K.R. (2014). *Catalogue of fungi preserved in National Herbarium and Plant Laboratories (KATH), Mycology section, Godavary.* Lalitpur, Nepal: Department of Plant Resources.

Adhikari, M.K. (1994-95). Mycodiversity in Nepal: a glimpse. *Bull. Nat. Hist. Soc. Nep., 3-4* (1-4), 4-6.

Adhikari, M.K. (2000). Status and conservation of fungi in Nepal. *Journal of Natural History Museum, 19*, 117–133.

Adhikari, M.K. (2009). *Researches on the Nepalese mycoflora*: *revised account on the history of mycological explorations*. Kathmandu, Nepal: K.S. Adhikari.

Adhikari, M.K. (2012). *Researches on the Nepalese mycoflora–2: checklist of macrofungi (mushrooms)*. Kathmandu, Nepal: K.S. Adhikari.

Adhikari, M.K. (2014). *Mushrooms of Nepal*. (Eds. Durrieu, G. and Cotter, H.V.T.). Kathmandu, Nepal: K.S. Adhikari

Alexopoulos, C.J. & C.W. Mims. (1979). *Introductory Mycology*. New York. USA: John Willey and Sons Inc.

Alexopoulos, J.C. & Beneke, E.S. (1962). *Laboratory Manual for Introductory Mycology*. USA: Burgess Publishing Company.

Aryal, H. P. & Budhathoki, U. (2013). Mycodiversity at Sankarnagar community forest, Rupandehi district. *Nepal Journal of Science and Technology, 14*(1), 75–80.

Berkely, M.J. (1838). Description of exotic fungi in the collection of Sir W.J. Hooker from memories and notes of J.F. Hooker from memories and notes of J.F. Klotsch with addition and correction. *Ans. Natural History, 3*, 375–401.

Dennis, R.W.G. (1981). *Common British Fungi*. England: Saiga Publishing Co. Ltd.

Department of Forest Research and Survey. (2015). *State of Nepal's Forests*. Forest Resource Assessment (FRA). Kathmandu, Nepal: Author.

Devkota, S. & Aryal, H.P. (2020). Wild mushrooms of Nepal. Pp. 41–54. In: *Plant Diversity in Nepal* (Eds. Siwakoti, M., Jha, P.K., Rajbhandary, S. and Rai, S.K.)., Kathmandu, Nepal : Botanical Society of Nepal.

Dickson, C. & Lucas, J. (1979). *The Encyclopedia of Mushrooms*. London: Orbis Publishing Limited.

District Development Committee. (2007). *Rupandehi Jilla ek Parichaya* (In Nepali). Rupandehi District, Nepal: Author.

Gold, T.C. (1975). *The Biology of Fungi*. London: Hutchinson and Co-Ltd. and The English language Book Society.

Lloyd, C. G. (1808). *Mycological notes*. no. 1:75. Cincinnati, Ohio, USA: Lloyd Library & Museum.

Miller, O.K. Jr. (1984). *Mushrooms of North America*. New York, USA: E.P. Dutton, a Division of Elsevier-Dutton Publishing company.

Pacioni, G. (1981). *Simon & Schuster's Guide to Mushrooms*. (Ed. Lincoff, G.). USA: Simon & Schuster Inc.

Pokhrel, B. (2017). *Mushroom diversity of Amrite community forest, Kapilvastu district, western Nepal*. (MSc Thesis), Central Department of Botany, Kathmandu, Nepal.

Purkayastha, R. P. & Chandra, A. (1985). *Manual of Indian Edible Mushrooms*. New Delhi, India: Today's and Tomorrow's Printers and Publishers.

Svrèek, M. (1983). *The Hamlyn Book of Mushrooms and Fungi*. Artia, Prague, Czechoslovakia: The Hamlyn Publishing Group Limited.

Teng, S.C. (1939). *Higher Fungi of China*. China: National Institute of Zoology and Botany, Academia Sinica.

Walting, R. (1973). *Identification of the Larger Fungi*. Bucks, London: Hulton Educational Publications Ltd.

Figure 4: *Pycnoporus cinnabarinus* (Jacq.: Fr.) Karst.

Figure 5: *Daldinia concentrica* (Bolt.: Fr.) Ces. & De Not

Figure 6: *Schizophyllum commune* (Fr.) Fr.

Figure 7: *Trametes versicolor* (L.: Fr.) Pilat

Figure 8: *Hexagonia* sp.

Figure 9: *Daedaleopsis confragosa* (Bolt.: Fr.) Schr. var. *confragosa*

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Mycelial Growth of *Termitomyces albuminosus* (Berk.) R. Heim in vitro Culture

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Abstract

Termitomyces albuminosus (Berk.) R. Heim, commonly known as termite's mushroom, is a fungus belonging to Basidiomycetes. It grows on termite mound and has food and medicinal values. This research was carried out to determine the effect of various carbon sources, nitrogen sources, amino acids, vitamins and carbon-to-nitrogen ratios on the *in vitro* mycelial growth of this fungus. The specimens of this species were collected from Chitwan National Park, Central Nepal and inoculated into culture plates. The mycelia so obtained were subjected to culture process using semi solid and liquid media. One-way ANOVA, followed by Tukey HSD test was performed to compare the results of different treatments. Among the six carbon sources used, the best growth was observed in maltose with somewhat compact mycelial density while the least growth was observed in lactose ($p<0.05$). Among the nitrogen sources, the best growth was seen in yeast extract with compact mycelial density while the least growth was observed in urea. Glutamic acid was found to be the best amino acid for the mycelial growth with compact mycelial density while serine showed least growth with thin mycelial density. Out of 5 vitamins tested, thiamine showed the best growth stimulatory effect with compact mycelial density while folic acid showed the least growth effect with somewhat thin mycelial density. The carbon-to-nitrogen ratio of 5:1 favored the best growth with somewhat compact mycelial density while the least growth was observed in 1:1 ratio. Based on their popularity and capability to utilize lignocellulosic substrates, there is a need to pursue further work for cultivation of this mushroom.

Keywords: Artificial cultivation, Mycelial texture, Nutrients, Termitophilous fungi

Introduction

Termitophilous fungi comprise a monophyletic group of tropical gilled mushrooms with a single genus *Termitomyces*. They are unique, obligatory symbionts growing in close and intimate association with termites. They contain higher dry matter, protein and fiber, but contain lower amount of fat and carbohydrates. They are rich sources of important minerals, like phosphorus, potassium, calcium, copper, manganese, zinc, magnesium, sodium and iron. Besides nutrition, the species possess high medicinal value (Aryal, 2015). They are good sources of bioactive compounds (Aryal & Budhathoki, 2013). Due to high concentration of diverse phytochemicals, they are used in drug development (Aryal & Budhathoki, 2014).

Growth of fungi is measured in terms of changes in number of cells, in linear dimensions, in cell mass, in cell volume or in amount of some cellular components (Bilgrami & Verma, 1981). Various environmental and biochemical factors affect growth; among them nutrients have significant influence on mycelial growth (Hawker, 1957; Bilgrami & Verma, 1981; Kaul, 1999). Fungi receive their food either parasitically or saprotrophically. Nutritional requirements for growth and reproduction are carbon, nitrogen, sulphur, phosphorus, vitamins and certain metallic or trace elements sources (Kaul, 1999).

The mycelium accumulates food materials and synthesizes complex substances such as proteins, polysaccharides, fats and enzymes (Hawker, 1957). Thus, to achieve maximum yield, an optimum culture medium containing the nutrients in suitable amounts and combination should be formulated. Cultural characteristics optimization procedure aims at developing such a medium.

Nutritional components of a medium can be varied and the impact of these changes assessed in terms of fungal growth. All of these factors must be considered during optimization. The defined medium developed by such optimization process serves as a nutritional framework from which a production medium can be formulated. In a production medium, the nutritional components of the defined medium are replaced with low-cost, complex substrates. Use of this directed optimization method will be useful in developing production media for commercial mycelial production (Jackson, 1997). Thus, this study was conducted for the optimization of culture medium for mycelial growth of *Termitomyces albuminosus* (Berk.) R. Heim.

Materials and Methods

Study area

Exhaustive survey of various localities of Chitwan National Park, within the coordinate range of 27°35'08''N to 27°73'32''N latitude and 84°30'02''E to 84°59'27''E longitude, and altitudinal range of 220-330 m asl (Figure 1), was conducted for the collection of specimens of *Termitomyces albuminosus* (Berk.) R. Heim. The collected specimens were inoculated on culture plates and brought to the laboratory at Central Department of Botany (CDB), Tribhuvan University (TU) for further study.

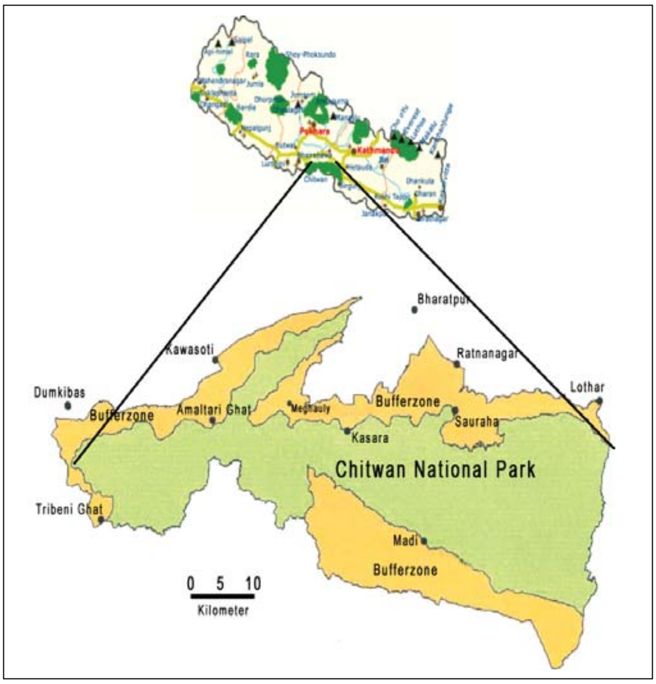

Figure 1: Chitwan National Park, Central Nepal.

Laboratory studies

The specimens were subjected to culture process using semi-solid and liquid media (Shim et al., 2005a) to observe the effects of various carbon sources, nitrogen sources, vitamins, amino acids and carbon-to-nitrogen ratios on mycelial growth at pH 6 and temperature 25±2°C. Measurements of linear growths were done on semi-solid media by means of a standard scale. Weighing of mycelial mass growth in liquid medium was done using an analytical balance.

Sterilization of instruments: Glasswares were wrapped in aluminium foil after cleaning with water and sterilized in a hot air oven at 160-170°C for 2 hrs. The culture media, cotton etc. were sterilized in an autoclave, at 121°C and 15 psi pressure for 30 min.

Isolation on potato dextrose agar (PDA) medium: Fresh and healthy fruiting body sample was surface sterilized by submerging in 0.4% sodium hypochlorite (NaOCl) for 1 min and was washed with sterilized water to remove residual NaOCl. Using a sterilized blade, its pileus and stipe were separated. The stipe was split longitudinally into two equal halves and approximately 3×6 mm pieces of tissue were taken from depth of $1/4^{th}$ thickness of the upper end of the stipe so that they contained neither outermost portion nor the central tissues of the stipe. The tissue pieces were inoculated on PDA plates. The plates were then sealed with parafilm tape and were covered with the help of aluminum foil.

The inoculated petriplates were incubated at 25±2°C in inverted position for seven days (Shim et al., 2005a; Shim et al., 2005b). At the end of the incubation period, mycelium was observed growing out of the inoculated tissue. Mycelial growth, colony diameter and colony texture were noted. Sub-cultures of the mycelium were done to obtain pure cultures. The pure cultures were transferred to PDA slants for further process.

Media preparation: Nutritional requirements for vegetative growth of mycelium of the species were studied by adopting the standard procedures

(Chandra & Purkayastha, 1977; Fasidi & Olorunmaiye, 1994; Shim et al., 2005a). The effects of variations in important components of culture media viz. carbon sources (dextrose, lactose, sucrose, fructose, mannitol, maltose and control), nitrogen sources (ammonium nitrate, peptone, urea, sodium nitrate, calcium nitrate, yeast extract and control), vitamins (ascorbic acid, nicotinic acid, folic acid, thiamine, D-priotine and control), amino acids (serine, leucine, valine, glutamic acid, arginine, aspartic acid and control) and C:N ratios were evaluated by maintaining five replicates for each treatment.

Statistical analysis

SPSS version 20.0 was used to analyze data. One-way analysis of variance (ANOVA) followed by Tukey's HSD test was performed to compare the results of different treatments.

Results and Discussion

Termitophilous mushrooms possess capability to produce lignocellulolytic enzymes, hence, has a potential to be efficient degrader of agro-wastes. Taprab et al. (2005) postulated that symbiotic fungi, viz. *Termitomyces* spp., produce laccases which are potentially involved in fungus combs and facilitate mushroom growth. Hence, for proper utilization of agro-wastes-biodegradation potential of *Termitomyces albuminosus* optimization of culture media for in vitro culture of this species would be beneficial. For the growth and reproduction of most fungi, the culture media must contain sources for carbon, nitrogen, sulphur, phosphorus, vitamins and certain trace or metallic elements (Kaul, 1999). Thus, various nutrients were compared to identify their efficiencies as the sources of these elements for the mycelial growth of *Termitomyces albuminosus*.

Effects of carbon sources

Linear mycelial growth varied significantly with change in carbon sources at 5% level of significance; *Termitomyces albuminosus* showed differential preferences for carbon sources for its metabolism (df = 6, 28; F = 25.88; P < .001). Utilization varied from one carbon source to another. The best linear growth was observed in maltose with somewhat compact mycelial density while the least growth was observed on lactose with compact mycelial density. However, in control medium, mycelial growth was better than in lactose. Dextrose, fructose and sucrose also stimulated good growth with somewhat compact mycelial density and better growth than control (Figure 2). However, both fresh weights (df = 6, 28; F = 1.28; P = .298), and dry weights (df = 6, 28; F = 0.914; P = .499) of mycelium in submerged culture did not vary significantly with change in carbon sources at 5% level of significance (Figure 3). It might be due to the lack of agitation, orbital incubation and continuous back up.

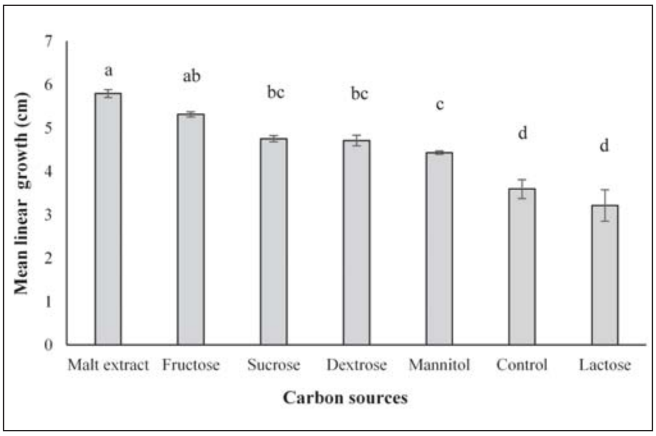

Figure 2: Linear mycelial growth on different carbon sources. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD test [$p < 0.05$].

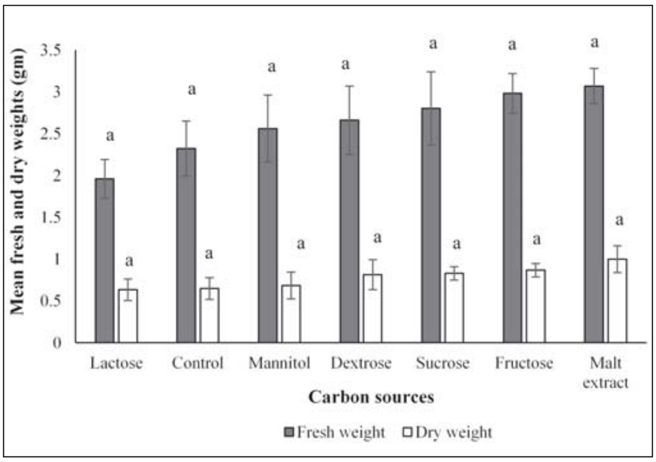

Figure 3: Mycelial growth on different carbon sources in submerged culture. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD test [$p < 0.05$].

According to Cochrane (1958), the ability of an organism to utilize carbohydrate depends on the types of enzymes produced by the organism. The results of the study showed that *Termitomyces albuminosus* produces enzymes which utilize maltose and fructose better than any other carbon sources. Shim et al. (2005b) reported that maltose was most suitable for mycelial growth in *Cystoderma amianthinum* var. *ruigosoreticulatum* while lactose was least suitable. According to Manjunathan and Kaviyarasan (2011), for *Lentinus tuber-regium*, dextrose was most effective while lactose was least effective. Ayodele (2008) found that, in *Psathyrella atroumbonata,* glucose was the best carbon sources followed by maltose, starch and mannitol in decreasing order. The least growth was observed in lactose and control. Subba (1975) reported that, among the carbon sources, sucrose caused the best growth while lactose supported poor growth in *Choanephora infundibulifera* Chandra and Purkayastha (1977) reported that most of the tropical edible macro-fungi were in favor of utilizing glucose than other carbon sources. Jayasinghe et al. (2008) found that dextrose was the best carbon source on *Ganoderma lucidum*. This was closely followed by galactose and fructose which were not considerably different from each other. Jonathan et al. (2006) found that in *Pleurotus florida,* the most supportive sugars were among the monosaccharides. Aldohexose (glucose) stimulated greater biomass yield than ketohexose (fructose) under the same conditions. Generally, complex sugar and sugar alcohols produced little biomass with the exception of dextrin and mannitol. He attributed the lower mycelial production with polysaccharides and sugar alcohols to their complex nature since hydrolytic enzymes would be required to convert polysaccharides and sugar alcohols to simple sugar before they will enter respiratory pathways. Mannitol (a sugar alcohol) also supported good biomass yield of *P. florida.* The best carbon source suitable for promoting mycelial growth of *Lignosus rhinoceros* was glucose but with somewhat compact mycelial density according to Lai et al. (2011). Additionally, fructose and mannose also recorded a high radial mycelial growth rate. The combination of these three carbon sources indicated that *L. rhinoceros* preferred monosaccharaides. Imtiaj et al. (2008) found that, in *Schizophyllum commune,* the suitable mycelial growth was found in dextrin and fructose. However, the lowest growth of mycelium was obtained in lactose, mannose and sorbitol. Poudel (2012) reported that maltose and sucrose, as sources of carbon, were most suitable for in vitro mycelial growth in *Volvariella taylorii*. Acharya (2012) reported malt extract as the best carbon source for the optimum growth of *Amanita chepangiana*.

Effects of nitrogen sources

Linear growth varied significantly with change in nitrogen source at 5% level of significance (df = 6, 28; F = 147.39; $P < .001$). The best linear growth was seen with compact mycelium density in yeast extract while the least growth with compact mycelial density was observed in urea (Figure 4). Similarly, both fresh weights (df = 6, 28; F = 29.87; $P < .001$), and dry weights (df = 6,28; F = 2.53; P = .043) of mycelium in submerged culture also varied significantly with change in nitrogen source at 5% level of significance. Maximum fresh weight was observed in yeast extract while minimum was observed in urea. However, in control treatment, mycelia had better growth than in urea with somewhat compact mycelial density (Figure 5).

Among the organic and inorganic nitrogen sources, *Termitomyces albuminosus* utilized organic nitrogen

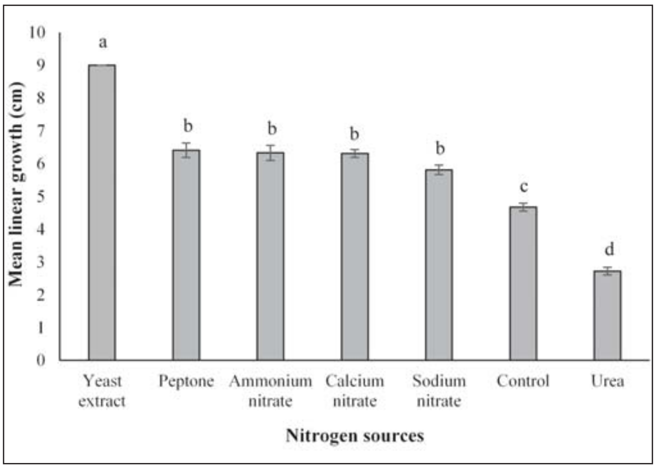

Figure 4: Linear mycelial growths on different nitrogen sources. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD Test [$p < 0.05$].

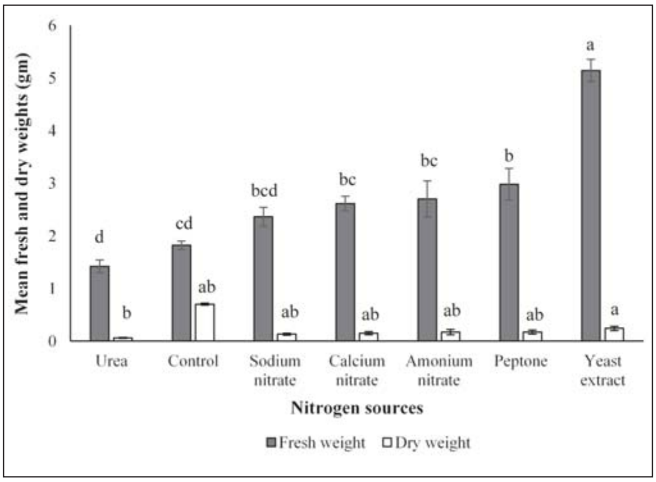

Figure 5: Mycelial growth on different nitrogen sources in submerged culture. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD Test [$p < 0.05$].

better than inorganic nitrogen. This finding was similar to that of Ayodele (2008), who reported that *Psathyrella atroumbonata* showed preference for organic nitrogen than inorganic nitrogen with the best yield in yeast extract and the least yields in sodium nitrate and ammonium nitrate. Manjunathan & Kaviyarasan (2011) also found that *Lentinus tuber-regium* utilized organic nitrogen better than inorganic nitrogen, yeast extract being the best for its mycelial growth in the same line. They suggested that the stimulatory effect of yeast extract in their study may have been due to its amino acids, protein and vitamins. According to Adebayo et al. (2011), comparatively, organic nitrogen supported optimum production of mycelium by *Pleurotus ostreatus* but when inorganic nitrogen was used, poor biomass growth was yielded. They reported urea as the best nitrogen source for biomass production by *P. ostreatus*. Jayasinghe et al. (2008) reported that *Ganoderma lucidum* showed optimum mycelial growth on ammonium acetate, glycine, arginine and calcium nitrate. They observed that inorganic nitrogen sources also enhanced the mycelial growth of *G. lucidium*. Subba (1975) found that, among the nitrogen sources, ammonium nitrogen and aspartic acid caused the best growth while nitrate nitrogen was found to stimulate moderate growth in *Choanephora infundibulifera* Jonathan et al. (2006) found that, in *Pleurotus florida*, inorganic compounds supported moderate biomass production. The best biomass yield was found with ammonium nitrate closely followed by potassium nitrate. Among the complex nitrogen compounds, yeast extract and casein hydrolysate supported significant biomass. Lai et al. (2011) observed that the best nitrogen source was potassium nitrate for promoting mycelial growth of *Lignosus rhinoceros*. Imtiaj et al. (2008) found that the most suitable nitrogen sources were calcium nitrate, potassium nitrate, and alanine, and the most unsuitable were ammonium phosphate, histidine, urea and arginine for growth of *Schizophyllum commune* Shim et al. (2005a) found that the best nitrogen source was glycine for *Macrolepiota procera*. However, in this study, urea, despite being an organic nitrogen source, showed the least support for the mycelial growth of the mushroom species under study. Poudel (2012) reported yeast extract and peptone as the most suitable sources of nitrogen for *in vitro* mycelial growth in *Volvariella taylorii*. Acharya (2012) observed sodium nitrate as optimum nitrogen source for the mycelial growth of *Amanita chepangiana*.

Effects of vitamins

Linear growth varied significantly with change in vitamin in the culture medium at 5% level of significance (df = 5, 24; F = 6.37; $p < .001$). Out of five vitamins tested, the highest liner growth was found in thiamine and the lowest in folic acid ($p < .05$). Ascorbic acid and D-priotine also stimulated good growth with somewhat thin mycelia density and better growth than control (Figure 6).

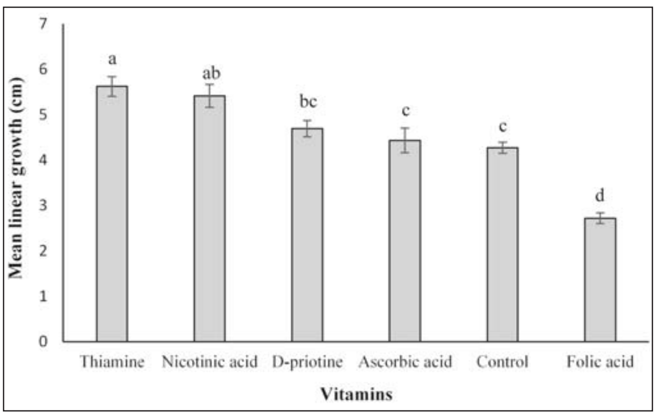

Figure 6: Linear mycelial growths on different vitamins. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD Test [$p < 0.05$].

Likewise, fresh weight of mycelium in liquid medium also varied significantly with change in vitamin sources at 5% level of significance (df = 5,24; F = 8.87; $p < .001$). Maximum weight was observed in D-priotine while minimum weight was observed in ascorbic acid. However, dry weight of mycelium in submerged culture did not vary significantly with change in vitamins source at 5% level of significance (df = 5, 24; F = 2.28; $p = 0.078$; Figure 7).

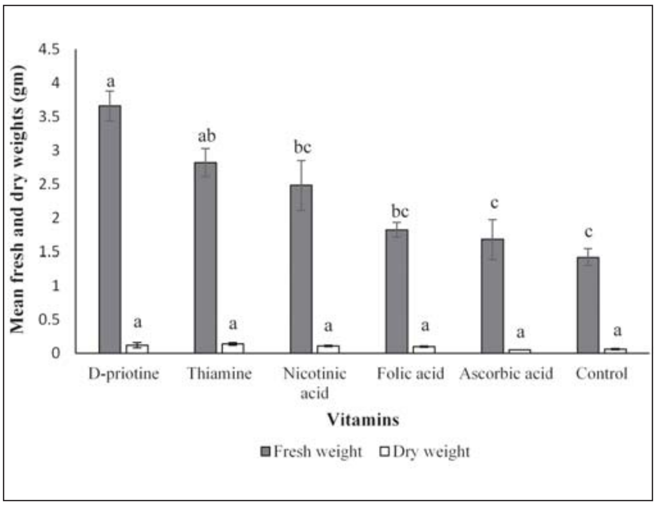

Figure 7: Mycelial growth on different vitamins in submerged cultures. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD Test [$p < 0.05$].

Among different vitamins in this study, thiamine was observed to produce the best, linear growth in *Termitomyces albuminosus.* Control treatment with somewhat compact mycelia density, ascorbic acid with somewhat thin mycelial density and folic acid with somewhat thin mycelial density showed the least growth (Figure 4). This finding was in line with the report of Ayodele (2008), who proved that thiamine was the best vitamin, followed by nicotinic acid and riboflavin for *Psathyrella atroumbonata* while the least growth was observed in folic acid. This observation was also in line with the report of Landers (1964) who found that thiamine stimulated mycelial growth of *Cercospora arachidicola* in liquid culture. Manjunathan and Kaviyarasan (2011) also proved that thiamine was the best among the vitamins followed by biotin and tocopherol for *Lentinus tuber-regium* Adebayo et al. (2007) found that riboflavin and pyridoxine promoted the mycelial growth for *Schizophyllum commune*. Adebayo et al. (2011) observed that among the vitamins tested, ascorbic acid and folic acid had the highest stimulatory effect on mycelia of *Pleurotus ostreatus*. Pokhrel et al. (2009a) reported that riboflavin and thiamine were the most stimulatory vitamins for mycelial growth of *Lyophyllum decastes*. Poudel (2012) observed that serine, as vitamin, caused the best in vitro mycelial growth in *Volvariella taylorii*. Acharya (2012) reported that, in *Amanita chepangiana*, ascorbic acid caused optimum mycelial growth.

Effects of amino acids

Linear growth varied significantly with change in amino acid in the culture medium at 5% level of significance (df = 6, 28; F = 11.56; $p < .001$). Among them, glutamic acid proved to stimulate the maximum linear growth with somewhat thin mycelial density while folic acid caused the least growth with thin mycelial density. The control treatment showed intermediate growth with somewhat thick mycelial density. Leucine, valine, aspartic acid and serine showed medium linear growths with somewhat thin mycelial densities (Figure 8). However, fresh weights (df = 6, 28; F = 0.958; $p = .471$) and dry weights (df = 6, 28; F = 0.409; $p = .866$) of mycelia in submerged cultures did not vary significantly with change in amino acid source at 5% level of significance (Figure 9).

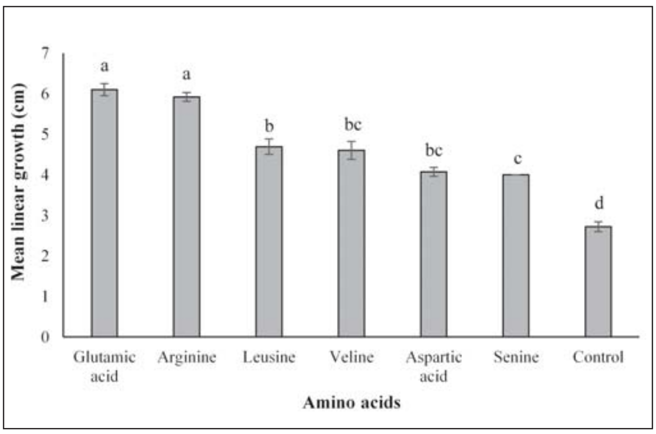

Figure 8: Linear mycelial growths on different amino acids. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD Test [$p < 0.05$].

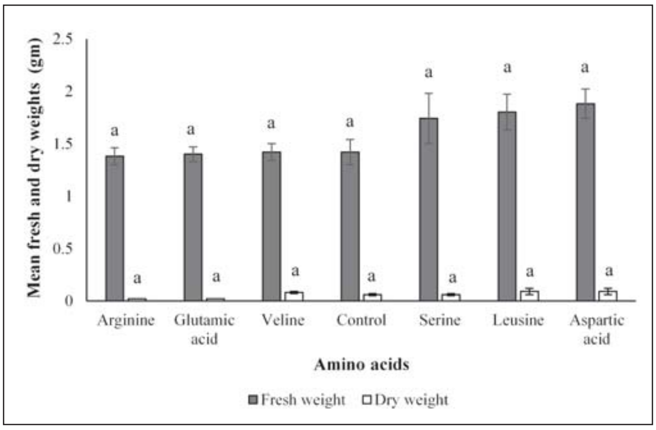

Figure 9: Mycelial growth on different amino acids in submerged culture. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD Test [$p < 0.05$].

According to Ayodele (2008), asparagine proved to be the best amino acid followed by aspartic acid for *Psathyrella atroumbonata*. According to Chandra & Purkayastha (1977), amides such as asparagine and aspartic acid have been employed in increasing mycelial growth and fruiting body production in *Agaricus bisporus*. Hayes (1981) observed that both higher and lower concentrations of amino acids were found to be either ineffective or inhibitory for the growth of mycelia. According to Manjunathan & Kaviyarasan (2011) glycine proved to be the best amino acid followed by L-ornithine monohydrochloride for the mycelial growth of *Lentinus tuber-regium* Hayes (1981) reported that higher and lower concentrations of these amino acids were found to be either ineffective or inhibitory for growth. Pokhrel et al. (2009b) investigated the effects of light, moisture, amino acids, vitamins and mineral nutrients on mycelial growth of *Lyophyllum decastes* on solid media. Glutamic acid was the best amino acid among the eight amino acids tested. Adebayo et al. (2011) observed that some of the amino acids had a stimulatory effect on mycelial yield of *Pleurotus ostreatus*. Optimum mycelial growth was recorded when glycine was used. Jonathan et al. (2006) found that in *Pleurotus florida*, although all the twelve amino acids used in his study enhanced biomass production, the most stimulatory amino acid was tryptophan. Pokhrel et al. (2009b) found that proline and glutamic acid showed significant growth than other amino acids tested in *Lyophyllum decastes*. Poudel (2012) concluded that serine was most suitable amino acid for mycelial growth of *Volvariella taylorii*. According to Acharya (2012), arginine was found to cause maximum in vitro mycelia growth in *Amanita chepangiana*.

Effects of carbon-to-nitrogen (C:N) ratios

The effects of different carbon-to-nitrogen ratios on linear growths varied significantly with the change in carbon-to-nitrogen ratio in the culture medium at 5% level of significance (df = 9,40; F = 8.656; $p < .001$). The linear growth rate increased with the proportional increase in carbon up to C:N ratio of 5:1. C:N ratio of 1:1 showed the least linear growth with somewhat compact and thin mycelial densities respectively. In the same manner, when the ratio of nitrogen was increased, linear growth rate also increased gradually up to the C:N ratio of 1:5. Hence, the best linear growths were observed when the C:N was 5:1 and 1:5. However, better growth was observed in control than in 1:1 ratio (Figure 10). Similarly, both fresh weights (df = 9,40; F = 7.44; $p < .001$) and dry weights (df = 9,40; F = 2.876; $p = .010$) of mycelium in submerged culture also varied significantly with change in carbon-to-nitrogen ratio at 5% level of significance (Figure 11). Maximum fresh weight was observed in C:N ratio of 5:1 while minimum was observed in 1:1 ratio. However, mycelial fresh weight in control was observed to be better than in C:N ratios of 1:1, 1:2 and 1:5 while it was equal with 2:1 and 3:1 ratio. Similarly, maximum dry weight was observed in C:N ratio of 1:2 and minimum in 1:1 ratio.

It was observed that the mycelial growth of the fungi under study increased with the proportional increase of carbon in comparison to nitrogen up to C:N ratio of 5:1. Control treatment and C:N ratio of 1:1 showed the least growth with somewhat compact and thin mycelial density respectively. When the ratio of nitrogen was increased, mycelial growth was also found to increase gradually in the same manner, up to C:N ratio 1:4. However, the best growth observed was when the C:N ratio was 5:1 (Figure 10). Lai et al. (2011) reported that C:N ratio of 10:1 was favorable for mycelial growth of *Lignosus* *rhinoceros*. Notably, no growth occurred at 40:1 C:N ratio. Shim et al. (2003) observed that the optimum C:N ratio suitable for favorable growth of *Paecilomyces fumosoroseus* were on culture media which were adjusted to C:N ratio of 40:1. Shim et al. (2005b) found the C:N ratio of 30:1 was the best for the mycelial growth of *Cystoderma amianthinum* var. *ruigosoreticulatum*. Chandra & Purkayastha (1977) observed C:N ratio of 3:1, 5:1 and 1:3 were favorable for the growth of *Lentinus subnudus*, *Volvariella volvacea* and *Termitomyces eurrhizus* respectively. According to Manjunathan & Kaviyarasan (2011), carbon-to-nitrogen ratios of 1:3 and 1:5 were the best for the growth of *Lentinus tuber-regium* (Fr.) Fr. Ayodele (2008) found that the C:N ratios of 4:1 and 1:4 supported the best growth

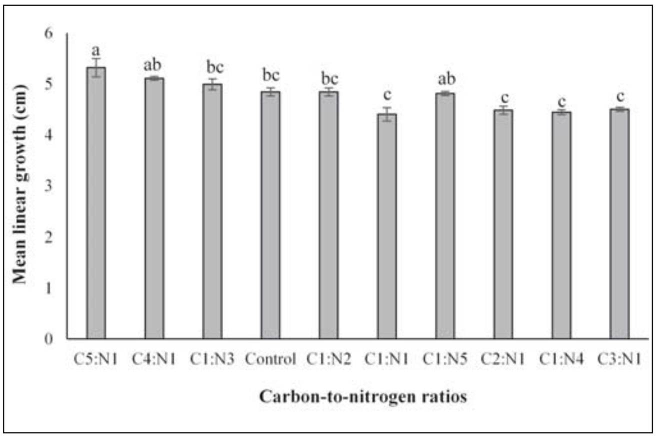

Figure 10. Linear mycelial growths on different carbon-to-nitrogen ratios. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD Test [$p < 0.05$].

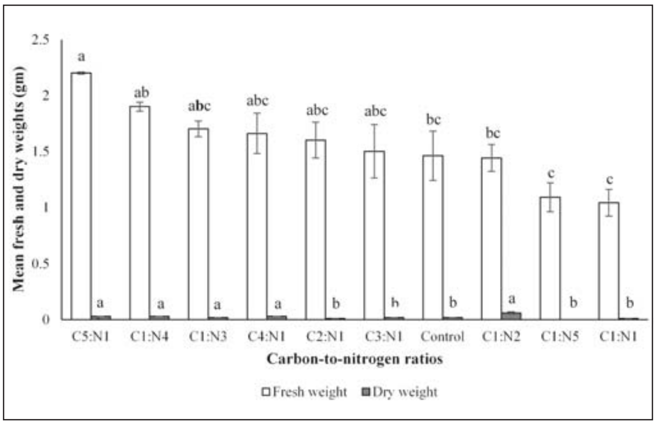

Figure 11. Mycelial growth on different amino acids in submerged culture. Different letters above the error bars indicate significant differences in mycelial growths among the treatments as determined by Tukey's HSD Test [$p < 0.05$].

of *Psathyrella atroumbonata*. According to Poudel (2012), C:N ratios of 5:1, 1:4, and 1:5 induced optimum mycelial growths in *Volvariella taylorii* (Berk. & Broome) Singer. Acharya (2012) has concluded 4:1 as the optimum carbon-to-nitrogen ratio for the growth of *Amanita chepangiana* Tulloss & Bhandary.

Conclusion

In view of the popularity of *Termitomyces albuminosus* with rural masses and its capability to utilize lignocellulosic substrates, there is need to pursue further work for the cultivation of this mushroom species, so as to utilize its biological efficiency in converting agro-forestry by-products and other such substrates available in plenty in different parts of the country. Through domestication and bulk production, it can be made easily available to the consumers. This can generate revenue to the country and, at the same time, will reduce collection pressure on natural habitat. This is how the dual purpose of meeting the human demand and conservation aspects can be targeted simultaneously which is so vital for conservation of the natural ecosystem and sustainability. Therefore, there is need to domesticate this mushroom species. In this regard, the findings of this research indicate that the optimal medium, comprising of maltose as carbon source, yeast extract as nitrogen source, thiamine as vitamin, serine as amino acid and C:N ratio of 5:1, may be suitable for in vitro mycelial growth of this mushroom species.

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References

Acharya, K. (2012). *A study on mycelial growth of Amanita chepangiana, a wild edible mushroom, on different nutrient sources* (Master's thesis). Central Department of Botany, Tribhuvan University, Nepal.

Adebayo, T.B.C., Ogunjobi, A.A. & Afolabi, O.O. (2007). Physiochemical studies on *Schizophyllum commune* Fr. a Nigerian edible fungus. *World Applied Sciences Journal*, *2*(1), 73-76.

Adebayo, T.B.C., Jonathan, S.G., Popoola, O.O. & Egbomuche, R.C. (2011). Optimization of growth conditions for mycelial yield and exopolysaccharide production by *Pleurotus ostreatus* cultivated in Nigeria. *African Journal of Microbiology Research*, *5*(15), 2130-2138.

Aryal, H.P. (2015). *Termitomyces R. Heim (Trichlomataceae) in Nepal: Diversity, Nutrients and Growth* (Ph.D. thesis). Central Department of Botany, Tribhuvan University, Kathmandu, Nepal.

Aryal, H.P., & Budhathoki, U. (2013). Phytochemical screening of termite's mushroom in Nepal. *Journal of Natural History Museum*, *27*, 107-119.

Aryal, H.P., & Budhathoki, U. (2014). Ethnomycology of *Termitomyces* spp. R. Heim for its medicinal importance in Nepal. *An International Journal of Medicinal Plants*, *6*(2), 128-137.

Ayodele, S.M. (2008). Studies on the vegetative growth requirements of *Psathyrella atroumbonata* Pegler, an edible mushroom in Nigeria. *African Journal of Plant Science*, *2*(5), 028-033.

Bilgrami, K.S., & Verma, R.N. (1981). *Physiology of fungi*. New Delhi, India: Vikas Publishing House Pvt. Ltd.

Chandra, A., & Purkayastha, R.P. (1977). Physiological studies on Indian mushrooms. *Transactions of British Mycological Society*, *69*, 63-70.

Cochrane, V.W. (1958). *Physiology of fungi*. New York, USA: John Wiley and Sons.

Fasidi, I.O., & Olorunmaiye, K.S. (1994). Studies on the requirements for vegetative growth of *Pleurotus tuber-regium* (Fr.) Singer, a Nigerian mushroom. *Food Chemistry*, *50*(4), 397-401.

Hawker, E.L. (1957). *Physiology of reproduction in fungi*. London, UK: Cambridge University Press.

Hayes, W.A., & Hand, P. (1981). Interrelated studies of physical, chemical and biological factors in casing soil and relationships with productivity in commercial culture of *Agaricus biosporus* Lange. *Mushroom Science*, *11*, 177-181.

Imtiaj, A.C.J., Lee, G.W., Kim, H.Y., Shim, M.J., Rho, H.S., Lee, H.S., Hur, H., Lee. M.W., Lee, U.Y., & Lee. T.S. (2008). Physiochemical requirement for the vegetative growth of *Schizophyllum commune* collected from different ecological origins. *The Korean Society of Mycology*, *36*(1), 34-39.

Jackson, M.A. (1997). Optimizing nutritional conditions for the liquid culture production of effective fungal biological control agents. *Journal of Industrial Microbiology & Biotechnology*, *19*, 180-187.

Jayasinghe, C., Imtiaj, A., Hur, H., Lee, G.W., Lee, T.S., & Lee. U.Y. (2008). Favorable culture conditions for mycelial growth of Korean wild strains in *Ganoderma lucidium*. *The Korean Society of Mycology*, *36*(1), 28-33.

Jonathan, G., Ayodele, S., & Damilola. A. (2006). Optimization of sub-merged culture conditions for biomass production in *Pleurotus florida* (Mont.) Singer, a Nigerian edible fungus, *African Journal of Biotechnology*, *5*(16), 1464-1469.

Kaul, T.N. (1999). *Introduction to mushroom science (systematics)*. New Delhi, India: Oxford & IBH Publishing Co. Pvt. Ltd.

Lai, W.H., Murni, M.J.S., Fauzi, D., Abas, O.M., &. Saleh, V.N.M. (2011). Optimal culture conditions for mycelial growth of *Lignosus rhinoceros*. *The Korean Society of Mycology*, *39*(2), 92-95.

Landers, K.E. (1964). Growth of *Cercospora arachidicola* in a glucose-agar-asparagine-thiamine medium. *Phytopathology*, *54*, 1236-1239.

Manjunathan, J., & Kaviyarasan, V. (2011). Optimization of mycelial growth and antimicrobial activity of new edible mushroom, *Lentinus tuber-regium* (Fr.) Fr. Tamil Nadu, India. *International Journal of Pharm Tech Research*, *3*(1), 497-504.

Pokhrel, C.P., Yoshimoto, H., Iida, S., & Ogha, S. (2009a). Mycelial growth and fruit body formation of *Lyophyllum decastes* in Livestock Compost. *Journal of Faculty of Agriculture, Kyushu University*, *49*(2), 273-282.

Pokhrel, C.P., Yadav, R.K.P., & Obga, S. (2009b). Effect of physical factors and synthetic media on mycelial growth of *Lyophyllum decastes*. *Journal of Ecobiotechnology*, *1*(1), 46-50.

Poudel, R. (2012). *Optimization of cultural media for mycelial growth of Volvariella taylorii (Berk. & Broome) Singer, an edible mushroom of Nepal* (Master's thesis). Central Department of Botany, Tribhuvan University, Kathmandu, Nepal.

Shim, S.M., Lee, K.L., Im, K.H., Lee, U.Y., Lee, M.W., & Lee. T.S (2003). The characteristics of cultural conditions for the mycelial growth and fruiting body formation of *Paecilomyces sinclairii*. *The Korean Society of Mycology*, *31*(1), 8-13.

Shim, S.M., Oh, Y.H., Lee, K.R., Kim, S.H., Im. K.H., Kim, J.W., Lee, J.U., Shim, Y.O., Shim, M.J., Lee, M.W., Ro, H.S., Lee, H.S., & Lee, T.S. (2005a). The characteristics of cultural conditions for the mycelial growth of *Macrolepiota procera*. *The Korean Society of Mycology*, *33*(1), 15-18.

Shim, S.M., Oh, Y.H., Lee, K.R., Kim, S.H., Im, K.H., Kim, J.W., Lee, U.Y., Shim, J.O., Shim, M.J., Lee, M.W., Ro, H.S., Lee, H.S., & Lee, T.S. (2005b). The cultural conditions affecting the optical mycelial growth of *Cystoderma amianthinum*. *The Korean Society of Mycology*, *33*(1), 15-18.

Subba, S.K. (1975). *Effect of carbon and nitrogen sources on growth of Choanephora infundibulifera (Curry) D.D. Cunn* (Master's thesis). Central Department of Botany, Tribhuvan University, Kathmandu, Nepal.

Taprab, Y., Johjima T., Maeda, Y., Moriya, S., Trakulnaeamsai, S., Naparatnaraporn, N., Ohkuma, M., & Kudo, T. (2005). Symbiotic fungi produce laccases potentially involved in fungus combs of fungus growing termites in Thailand. *Applied and Environmental Microbiology*, *71*(12), 7696-7704.

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Institutional Perspective of Yarsagumba (*Ophiocordyceps sinensis*) Collection in Kailash Sacred Landscape, Nepal and India

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Abstract

Ophiocordyceps sinensis (Yarsagumba) is a valuable non-timber forest product found in between 3000-5000 m elevation range throughout the Hindu-Kush Himalayan range. Kailash Sacred Landscape, Darchula of Nepal and Dharchula of India are good habitats of this species. People have been involving in collection and selling activities due to its high economic value. Governments of both countries also recognize its high value so that the royalty of Yarsagumba is assigned the highest one among all forest products for same quantity. However, due to its habitat in remote areas as well as diverse people involved in collection, there are several challenges in collection and trading within the site. Moreover, having two different set of rules for different countries but same eco-cultural landscape in collection and royalty payments, regulations on collection and trading has further increased the challenges. This paper tried to explore the opportunities and challenges in Yarsagumba collection and onsite trading in Darchula district of Nepal and Pithauragadh district of India from institutional perspective.

Keywords: Collection challenges, Management, Policy, Sustainability

Introduction

Landscape level conservation initiative in Kailash Sacred Landscape (KSL) was started in 2009. Program lunched since 2012 in first phase. Area coverage by the landscape is in largest proportion in Nepal (13289 sq. km), followed by China (10873 sq. km) and remaining parts in India (International Centre for Integrated Mountain Development [ICIMOD], 2019). Four district of Nepal (Humla, Bajhang, Baitadi and Darchula) and Pithauragadh district of India fall in the landscape. Biodiversity is an important resource in the Kailash Sacred Landscape. It provides numerous ecosystem services, ranging from the provision of food, fuel and shelter to cultural service including religious pilgrimages and tourism for other purpose (ICIMOD, 2019). The landscape is habitat to numerous species of plants, including many valuable medicinal plants such as Panchaule (*Dactylorhiza hatagirea*), Jatamansi (*Nardostachys grandiflora*), Kutki (*Neopicrorhiza scrophulariiflora*) and Yarsagumba (*Ophiocordyceps sinensis*) (Api-Nampa Conservation Area [ANCA], 2018).

Yarsagumba (*Ophiocordyceps sinensis*) is a caterpillar fungus that grows naturally in the northern alpine grassland of Bhutan, India, Nepal and the Tibetan plateau of China at an altitude of 3000 to 5000 m (Zhang et al., 2009). However, Yarsagumba is distributed around 4000 m to 5500 m altitude in high-altitude grassland and on sloppy land in Darchula district in and around Bhagawati, Ghunsha, Byas and Rapla areas (Uprety et al., 2016). Recognized for its medicinal value (Gupta & Manvitha, 2017), it provides high economic returns to mountain communities living in subsistence economy (Shrestha & Bawa, 2014). As a result, Yarsagumba trade has become an important economic activity in the Kailash Sacred Landscape with its major market in China. A number of researchers found that it has a number of benefits. Researches range from medicinal effects (Shiao et al.,1997; Koh et al., 2003), chemical composition (Li et al., 2001; Shiao et al., 2002; Li et al., 2003), structural features (Kiho et al., 1999), quality control for quality products (Koh et.al., 2002; Li et al., 2006) to the scientific rediscovery of Yarsagumba (Zhu et al., 1998) in the researcher field. But no research yet focus on the collection challenges on same eco-cultural landscape.

Sustainable harvesting, environmental management and its trade across the border have become pertinent trans-boundary issues of concern in the KSL. Conservation salvage of *Ophiocordyceps sinensis* collection in the Himalayan mountains is neglected (Zhu et al., 1998). The ever-increasing harvesting pressure raises the question of sustainability. The fact that *Ophiocordyceps* has been collected for centuries and is still common argues for its resilience, but the lack of harvest studies for *O. sinensis* precludes a definite answer as to whether the harvest can be sustained at its current level (Winkler, 2008). National governments within the landscape are introducing guidelines and policies for sustainable harvesting and management practices, but policies and regulations for collection and trade of Yarsagumba differ from Nepal to India. This result will be very useful for the governments from where the Yarsagumba is collected and traded. Thus, this research aimed to identify the status of Yarsagumba management in two countries and to explore major challenges in Yarsagumba management and find out the possible way out. Therefore, this paper tries to explore the policy differences between two countries, because of this reason; are there any issues and challenges in collection of Yarsagumba in the same eco-cultural landscape? And suggest out the possible solutions for those issues raised during collection and trading (district level) at site level.

Materials and Methods

An embedded case study approach (Scholz & Tietje, 2002) was followed to carry out this study in a natural setting using a variety of methods as suggested by Collis & Hussey (2009) and Gerring (2007). Further, this study relied upon multiple sources of evidence using both qualitative and quantitative techniques (Yin, 2014). The study was based on descriptive and explorative social science research design. As suggested by Maxwell & Miller (2008) and Maxwell (2009), a single case study was deemed appropriate here, as phenomena about which little is known were studied in the case. Moreover, it is only such kind of forest management paradigm in the Karnali and Sudurpaschim Province of Nepal. This article is based on archival records of management plans, group-operating guideline, reports, meeting minutes and other official documents, together with focused interviews, interaction program, direct field observation and participant observation (Kawulich, 2005). The findings were further substantiated and validated through a wide range of stakeholders who were directly or indirectly involved in Yarsagumba collection, trading, collectors' security personnel and the forestry staffs who are involved in the processes.

The primary information was collected through the semi-structured interview-schedule for key-informants based on their experience along with content analysis of archive (documents and reports) and direct field observation. Personal interviews were arranged with members of Management council of Api Nampa Conservation Area (ANCA), officials from ANCA, general users, Non Timber Forest Products (NTFP) and Medicinal and Aromatic Plants (MAP) concessionaires/contractors, local units (government) and Non Government Organization (NGO) personnel. Moreover, information and views of different stakeholders were collected during focus group discussion, key informant interviews, general assembly, interaction program and monthly meetings.

Further, the secondary data were obtained from various published and unpublished information sources i.e., relevant literatures, library study, research reports, annual reports, official documents, journals, magazines, newspapers, online sources, books, archival records of Api-Nampa Conservation Area.

Quality standard and credibility of the research

Qualitative inquiry is becoming more popular (Anney, 2014), where trustworthiness with reliability and credibility are considered as vital factors. To accomplish this, as suggested by Anney, (2014) some criteria as: (a) credibility (in preference to internal validity); (b) transferability (in preference to external validity/generalizability); (c) dependability (in preference to reliability) and (d) conformability (in preference to objectivity) were incorporated during each and every step for the study. Considering this, the following methods were employed in every step of data collection and analysis throughout the study to increase the quality standards and credibility of the research.

1. Un-obstructive measures (for triangulation)
2. In-depth interview (as an overall strategy)
3. Observational checklist (just not "hanging out")
4. Participant observation (involving in social world: meeting, office, field work/operation)
5. Elite interview (with conceptualization of the problem through thoughtful questioning).

Results and Discussion

Status of Yarsagumba collection

Official record of the Division Forest Offices showed that around 3500 people were involved in Yarsagumba collection in the KSL area. People involved with their sheep herds during Yarsagumba collection that deteriorate the resource. In Fiscal year 2073/074, around 3500 people involved in Yarsagumba collection (formal record).In Nepal side, only Nepali can enter and collect the Yarsagumba**.** They need to have a citizenship card (age above 16 years). Problem is that school remained closed during the season. Around 10-15 years school children are involvedin the collection activity. They have full eye-sight to find a Yarsagumba. Village Development Committees (VDCs) also impose to charge entry fees for the collectors in this system, collector need to pay double fees. Difficult to regulate, very remote for

Table 1: Perceptions of different stakeholders

Stakeholder	Perceptions
Political parties / local bodies	• Council and committees should be punished if they did wrong activities. • Local right synchronizes by council and Api-Nampa CA. • Permission and insurance program should be lunched effectively. • Local people should be provided with enough information on collection time and procedure • Local peoplefocused program should be implemented, even orientation about the Yarsagumba, collection period, its importance etc. • There should be involvement of local bodies in Yarsagumba regulation. • Management committee and ANCA need to coordinate with local bodies while regulation the Yarsagumba.
Warden	• Yarsagumba collection area is the habitat of Red panda, Snow leopard, Kasturi etc. • In BS 2074-1-8, collection directives come into enforcement. • Almost 7 quintals of Yarsagumba per year is collected. • Collection time is around one month. • Directives enforce only inside the protected areas. • Pits are needed for waste management. • Ownership development is needed in the local people. • Sustainable development of ANCA is required • There is some place where local level and ANCA have some confrontation. • Transparency of budget is required. • Changing working and collaboration willingness is necessary.
Api-Nampa Conservation Area Management Council	• Heavily soil digging by collectors that threats to its sustainable management. • Large and remote areas hindering its monitoring. • Better to issue collection license to the local people as they are responsible to their local environment, thereby ensuring its sustainable management. • Awareness rising to collectors and traders. • Developing sustainable harvesting / collection (management) practice ensuring its sustainability. • Women and children are heavily involved (50% or more some places) in collection but get little benefit (not property rights) • Children are good collector as they easily see Yarsagumba than older or matured persons. • Problem of plastic and other bottle pollution • Problem of grazing (sheep, goat and mull) and their dung is also harmful for Yarsagumba production, which seems chemically harmful so that reducing its production day by day.

Table 2: Comparisons of existing institutional arrangements and policies between Nepal and India

	Nepal	India
Legislative provision	According the Forest Act, 2019 the right to issue the license for collecting the Non-Timber Forest Products (NTFPs) including Yarsagumba is vested with divisional Forest Officer. Likewise, National Parks and Wildlife Conservation Act, 1973 provide right to Warden for issuing NTFPs collection license.	Central government has right to issue the permits for collection.
Policy objectives	Forest policy, 2019 and Forestry Sector Strategy (2016-2025) emphasize on sustainable management of NTFPs resources.	There is no specific plan for collection. There is no limitation for collection of the Yarsagumba.
Institutional arrangement	Interested traders need to register an application in DFO in case of outside protected area and in Warden office if the collection area is located inside the protected area. DFO and Warden grant the license for collecting Yarsagumba after completing necessary steps not exceeding the amount specified in the District Forest and Protected Area Management Plan.	Interested traders needs to register in officials of central government only.
Collection and trading practices	Local people collect Yarsagumba from forest and sell to local traders. Local traders sell to regional and national traders. Local level can make separate rules.	All interested people throughout the country get permission for collections throughout the country.
Strength	Harvesting based on annual allowable harvest. Due process is followed.	There is free trade system throughout country. Even a single letter permits to carry throughout the country.
Weakness	Blanket approach of collection rule in all areas. Ecologically unsustainable collection. Traders have monopoly in Yarsagumba collection and trading.	There is no tracking system during the collection and trade. Due to open border towards Nepal, people can enter into Nepal throughout the border.

supervisors and officials to stay all the time in Yarsagumba collection areas.

Horse, donkey, mule, goat, sheepgrazing degrade the site and hider the regeneration of Yarsagumba. Illegal shops are established inside collection area, which also damage the plants. The illegal harvesting of the forest products also deteriorates the population of Yarsagumba. Double budgeting system, duplication of works between the different government agencies are also causes of destruction of Yarsagumba populations.

Institutional arrangement for Yarsagumba collection

According the Forest Act, 2019, the right to issue the license for collecting the Non-Timber Forest Products (NTFPs) including Yarsagumba is vested with Divisional Forest Officer (DFO). Likewise, National Parks and Wildlife Conservation Act, 1973, provided right to warden for issuing NTFPs collection license within the protected areas system. Based on these legislative provisions, DFO and Warden used to grant the license for collecting Yarsagumba after completing necessary steps. DFO can grant permission for collection not exceeding the amount specified in the five-year forest management plan, whereas warden also grant collection license not exceeding the amount specified in the management plan of protected area.

Interested traders need to register an application in DFO in case of outside protected area and in Warden Office if the collection area is located inside the protected area.

Major issues explored for Nepal

Despite having the provision in Yarsagumba Management (Collection and Trade) Directives, 2073 B.S. brought into practice by Government of

Nepal, Department of National Parks and Wildlife Conservation, the major issues raised by the stakeholder during data collection in institutional aspect for Nepalese side were following highlights.

- In sufficient regulatory enforcement and supervision despite the guidance in place.
- Pollution in the collection side and deforestation for fuel wood purpose.
- Different level of treatment in entry fees implies by buffer zone management committee and local units for same nationals - Nepalese (within the local unit - NRs 500.00, with in the same district - NRs. 2000.00 and rest of all Nepalese - NRs. 3000.00)
- The operational plan of CFUGs inside the Api-Nampa Conservation Area need to be revised considering the updated context and provisions.
- Very short period of collection in Nepal where, collection allows only for one month.

Major issues explored for India

There are a number of provisions in Indian guidelines that create implications in legalization in Yarsagumba collection for the Kailash region, which are as follows:

- Low royalty rate in India than in Nepal as Rs. 10,000.00 Indian Currency for per Kg whereas Nepal assigned for same quantity to NRs. 30,000.00
- Long period of collection in India i.e. more than 3 months.
- Total volume production might be increased thereby lowering down the market price of Yarsagumba in international market due to high rate of collection potentiality but without considering the perspective of sustainability.
- Issues of trans-boundary in and out flow of Yarsagumba.
- Due to long period of collection regulatory allowance and framework deteriorate the sustainability of Yarsagumba rotation for natural growth and maturity, sprouting and sporting for next generation.

Common understanding between stakeholders of Nepal and India

There were several points that the both side Yarsagumba stakeholders come in conclusion but till the date, no more initiation were made for commonality in collection and trade, institutional management issues regarding Yarsagumba.

- Resource inventory program for Yarsagumba is immediate need. This would help to increase the value of commercialization and value addition.
- Governance and institutional aspect of Yarsagumba management need to be strengthened. These would help to manage site specific Api-Nampa Conservation Area own guideline is needed for collection and permit. Indian is also allowed to collect the Yarsagumba but Nepali (local) people are banned in some area inside Nepal. Such system should be revised and eliminate the discriminatory portion for equality to all citizens of Nepal.

There is also lack of monitoring from Api-Nampa Conservation Area Council because of their limited capacity (technical, human and other resources). Also, there is lack of coordination among stakeholders due to different perception of different stakeholders. Cultural degradation alcohol consumption, cards playing in cash or kind (Yarsagumba), loud speakers (film), noise pollution. These situations also increased the quarrelling and fighting in collection time/place. Moreover, poaching and unsustainable and unscientific collection of other medicinal and aromatic plan together with Yarsagumba like Wild onion, Satuwa, Jatamansi are also being threatened.

Conclusion

Stakeholders are agreed on cooperation and collaborative work including cross border issues and sectors working in KSL area. However, until and unless the prevailing policies can not be harmonized for commonly shared resources and ecological region, it would not translate well into practice. Local units in coordination with Buffer zone management coordinating committee and Api-Nampa Management

Authorities needs to work very closely. For security reasons, fines-penalties and rewards, entry permits, social crimes controlling and gender related issues, sanitary and waste management, reducing deforestation and reforestation, among many other issues during Yarsagumba collection areas and along the route for sustainability of its production is to be considered simultaneously. Collective and collaborative efforts could be synergizes the action one after others. It has been found that production of Yarsagumbais decreasing each year.

Participation, gender equity and social inclusionsare needed for the proper harvesting management of Yarsagumba. Gender equality dimension of Yarsagumba and its value chain engendering of gender (in terms of benefit of Yarsagumba to the women) is to be highly considered. Women violence increased due to workload, monitory work load, social disorders, alcoholism, vandalism, playing cards in collection site. Such activities should be discourage by initiating innovative policies, programs and activities through collective and integrated approaches.

Other negative implication of Yarsagumba (e.g. cultural deterioration and harmony, antisocial activities, poaching, unsustainable/unscientific collection of other MAP species), hampering on the education of the student, heavily dependent only on Yarsagumba collection that threat to long term sustainability of their livelihoods and socio-cultural scenarios. For socio-economic issues resolution as aforementioned, respective strategies and strategic actions to be taken from local level to the national. Assurance of market of collected Yarsagumba is immediate action to be taken.

Future research areas

The following major recommendations are made from this study.

Policy: Government of Nepal needs to coordinate with Government of India to harmonize the similar policy in Yarsagumba collection especially royalty and time length- duration of collection.

Management: Entry fees and other regulating sustainability for sustain production to be left some areas - left uncollected in alternative years.

Sustainability: Life-cycle and its ecology are to be duly studied in reference to rotation of Yarsagumba.

***Contribution*:** Livelihoods and poverty alleviation contribution carried out by Yarsagumba is another field for contributing national goals and sustainable development agendas.

***Production*:** Quality production of Yarsagumba in relation to sites, collection seasons, and districts is to be explored.

***Constituents*:** Dietary components of varies field's Yarsagumba in Nepalese case is to be an area for doing research.

Security: A security check post of armed police force should be established in border site between Nepal, India and China to control illegal collection and transportation of all sorts of NTFPs including Yarsagumba.

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References

Api-Nampa Conservation Area. (2018). Annual Progress Report of for FY 2017/2018. Darchula, Khalanga, Nepal: Author.

Anney, V.N. (2014). Ensuring the Quality of the Findings of Qualitative Research: Looking at Trust worthiness Criteria. *Journal of Emerging Trends in Educational Research and Policy Studies*, *5*(2), 272-281.

Collis, J & Hussey, R. (2009). *A Practical Guide for Undergraduate and Postgraduate Students, Business & Economics*, (pp. 420) UK: Palgrave Macmillan.

Gerring, J. (2007). Case Study Research Principles and Practices, Cambridge, UK: Cambridge University Press.

Gupta, G. & Manvitha, K. (2017). A miracle mushroom its history, cultivation, phytopharmacology and medicinal uses. *International Journal of Herbal Medicine*, *5*(2), 69-72.

International Centre for Integrated Mountain Development. (2019). Kailash Sacred Landscape Conservation and Development Initiative (KSLCDI). Retrieved January 22, 2019, from http://www.icimod.org/9457

KohJonk-Ho, KWang-Won YU, Hyung-Joo SUH, Yand-Moon CHOI & Tae-SeokAHN (2002). Activation of Macrophages and the Intestinal Immune System by an Orally Administered Decoction from Cultured Mycelia of *Cordyceps sinensis*: Bioscience, Biotechnology, and Biochemistry, *66* (2). Retrieved December 31, 2018, from https://www.tandfonline.com/doi/abs/10.1271/bbb.66.407

Koh,Jong-Ho, Jin-Man Kim, Un-Jae Chang & Hyung-Joo Suh.(2003). Hypocholesterolemic Effect of Hot-Water Extract from Mycelia of *Cordyceps sinensis*. Retrieved December 31, 2018

Kawulich, B.B. (2005) Participant Observation as a Data Collection Method. *Qualitative Social Research*, *6*, 43.

Kiho, T., Ookubo, K., Usui, S., Ukai, S., & Hirano, K. (1999). Structural Features and Hypoglycemic Activity of a Polysaccharide (CS-F10) from the Cultured Mycelium of *Cordycepssinensis*. *Biological and Pharmaceutical Bulletin*, *22*(9), 966-970. https://doi.org/10.1248/bpb.22.966

Li, S. P., Li, P., Dong, T. T. X., & Tsim, K. W. K. (2001). Anti-oxidation activity of different types of natural *Cordycepssinensis* and cultured *Cordyceps mycelia*. *Phytomedicine*, *8*(3), 207-212. https://doi.org/10.1078/0944-7113-00030.

Li, S. P., Yang, F. Q. & Tsim, K. W. K. (2006). Quality control of *Cordyceps sinensis*, a valued traditional Chinese medicine. *Journal of Pharmaceutical and Biomedical Analysis*, *41*(5), 1571-1584. https://doi.org/10.1016/j.jpba.2006.01.046.

Li, Shao P, Zhao, K. J., Ji, Z. N., Song, Z. H., Dong, T. T. X., Lo, C. K. & Tsim, K. W. K. (2003). A polysaccharide isolated from *Cordyceps sinensis*, a traditional Chinese medicine, protects PC12 cells against hydrogen peroxide-induced injury. *Life Sciences*, *73*(19), 2503-2513. https://doi.org/10.1016/S0024-3205 (03)00652-0.

Maxwell, J. A., & Miller, B. A. (2008). Categorizing and connecting strategies in qualitative data analysis. In P. Leavy & S. Hesse-Biber (Eds.), *Handbook of emergent methods* (pp. 461-477). New York: Guilford Press.

Maxwell, J.A. (2009). Designing a Qualitative Study, Qualitative Research, 214-153.

Scholz, R. W., & Tietje, O. (2002). Embedded case study methods: Integrating quantitative and qualitative knowledge: Sage Publications.

Shiao,Y.J., Lee, S.S., & Wang, S.Y. (1997). Effect of *Cordyceps sinensis* on the proliferation and differentiation of human leukemic U937 cells. *Life Sciences*, *60*(25), 2349-2359. https://doi.org/10.1016/S0024-3205(97)00291-9

Shiao,T. H., L.H., Hsieh, C., & Chang, D. M. (2002). A comparison of the chemical composition and bioactive ingredients of the Chinese medicinal mushroom DongChongXiaCao, its counterfeit and mimic, and fermented mycelium of *Cordyceps sinensis*. *Food Chemistry*, *78*(4), 463-469. https://doi.org/10.1016/S0308-8146 (02)00158-9

Shrestha, U.B. & K.S. Bawa. (2014). Economic Contribution of Chinese caterpillar fungus to the livelihoods of mountain communities in Nepal. *Biological conservation*, *177*, 194-202. http://doi.org/10.1016/j.biocon.2014.06.019.

Uprety,Y., Poudel, RC., Chaudhary, RP., Oli, BN., Bhatta, LD. & Baral, SP. (2016). *Sustainable Utilization and Conservation of Non-Timber Forest Products: Major Species of Kailash Sacred Landscape Nepal*. Kathmandu, Nepal: Ministry of Forests and Soil Conservation, Research Centre for Applied Science and Technology & International Centre for Integrated Mountain Development.

Winkler, D. (2008). Yartsa Gumbu (*Cordyceps sinensis*) and the Fungal Commodification of Tibet's Rural Economy. *Economic Botany*, *62*(3), 291-305. https://doi.org/10.1007/s12231-008-9038-3

Yin, R.(2014). *Case Study Research: Design and Methods*.(5rd ed.) : Sage Publication.

Zhu, J.-S., Halpern, G. M., & Jones, K. (1998). The Scientific Rediscovery of an Ancient Chinese Herbal Medicine: *Cordyceps sinensis* Part I. *The Journal of Alternative and Complementary Medicine*, *4*(3), 289-303. https://doi.org/10.1089/acm.1998.4.3-289

Zhang Y, Xu Lui & Zhang S. (2009). Genetic diversity of *Ophiocordyceps sinensis*: a medicinal fungus endemic to the Tibetan Plateau: implication for its evolution and conservation. *Evolutionary Biology*, *9*, 290. DOI: 101186/1471-2148-9-290

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Notes on the First Sino-Nepal Joint Botanical Expedition to Bajhang, West Nepal

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Abstract

As part of Flora of Pan-Himalaya project, the first-ever Sino-Nepal Joint Botanical Expedition was carried out in Bajhang district of West Nepal. Here we summarize the objectives and achievements of this collaborative venture. This joint work included field visit in West Nepal supplemented by herbarium consultation in Nepal and China. During the fieldwork, herbarium specimens of 625 species were collected.The specimens are preserved at National Herbarium and Plant Laboratories (KATH) and Chinese National Herbarium (PE). Of the total collection, a list of 503 species is presented here.*Itea nutans* Royle (Iteaceae), collected during the expedition, is reported as a new generic and family record for the flora of Nepal. The description, distribution notes and collection details of the newly reported taxon are provided.

Keywords: Pan-Himalaya,Flora, *Itea nutans*, New record

Introduction

The Himalaya is one of the most extensive, beautiful and revered ranges on earth, and its plants are equally as enchanting as the ranges themselves. The Pan-Himalaya (the Himalaya and adjacent regions) forms a natural geographic unit, which includes the northeastern corner of Afghanistan, northern Pakistan, northern India, Nepal, Bhutan, northern Myanmar, and southwest China (Hong, 2015). Plants of some of the countries are reasonably well known, while those of the others are poorly known, and the historical efforts to bring the available information together and improve its quality by trans-boundary cooperation are equally important. To fill the knowledge gap and to usher in an era of close cooperation among the stakeholders, The Flora of Pan-Himalaya (FLPH) project was initiated; it is an international collaboration project to document the plant biodiversity within this region. The mission of FLPH is to better understand plant diversity in the Pan-Himalaya and toconserve the rich and precious plant diversity in this region. Coordinated by Chinese botanists, it was started in the year 2011 and intends to be a long term project. The flora will cover ca. 20,000 species of vascular plants in 50 volumes (ca. 80 books).

Pan-Himalaya forms a natural phytogeographical unit, ranging from the Vakhan Corridor in the west to the Hengduan Mountains in the east through the Karakoram and the Himalaya in between. This region is divided into 17 subregions: Vakhan, N Pakistan, Jammu & Kashmir, U Ganga & Indus, U Yarlung Zangbo, W Nepal, C Nepal, E Nepal, Sikkim & Darjiling, Bhutan, M Yarlung Zangbo, L Yarlung Zangbo, Yarlung Zangbo-Brahmaputra, Tangut, N Hengduan, S Hengduan, and U Irrawaddy (Figure 1)(Hong, 2015).

A number of botanical expeditions has been organized in Nepal focusing different ecological regions over the years either from within the country or jointly with international organizations; Rajbhandari (2016) has summarized the historical account of the explorations carried out in West Nepal highlighting the significant contributions made by the teams led by many prominent botanists and collectors including Scully (1876) in Mahakali valley, Duthie (1884-1886) in Garhwal, Kumaon to West Nepal (Nampa gadh), Bis Ram, K.N. Sharma

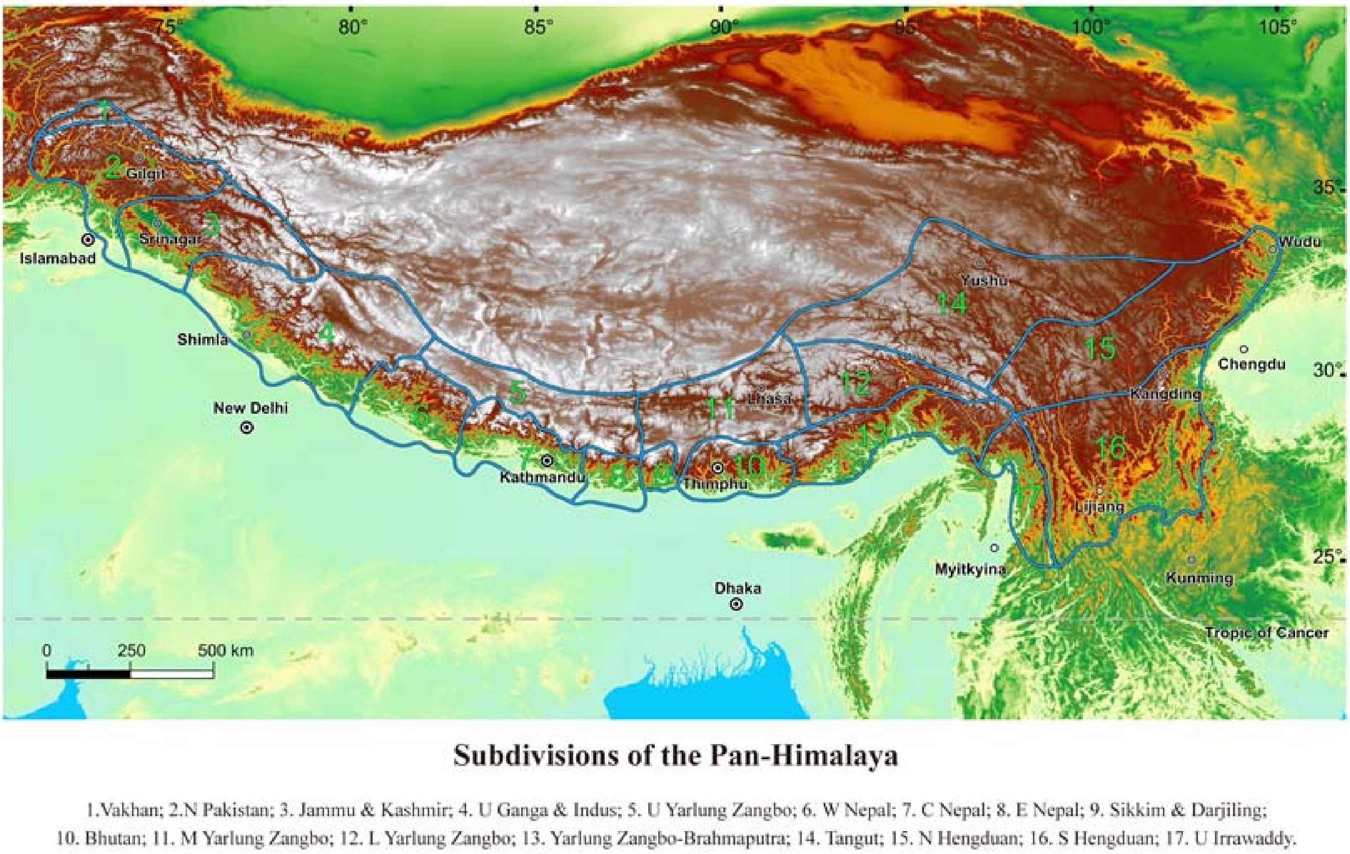

Figure 1: Map showing the geographical range of the Pan-Himalaya

& B.L. Gupta (1929-1935) in Khaptad region, K.R. Rajbhandari (1983) in Mahakali zone, Suzuki (1991) in Bheri, Karnali and Seti zone, Ikeda (2009, 2012) in Khaptad and Api area and many others.

Despite the many botanical explorations that have been carried out in Nepal since the late 18th century to this date by many foreign and national agencies (Rajbhandari, 1976; Rajbhandari, 2016) amassing a bulk of specimens deposited in the herbaria within and outside the country, the bilateral collaborative research with the northern neighbor has just begun.

As it is evident, Nepal occupies the three important subregions of Pan-Himalaya, providing crucial habitats for the unique plant species. In an initiative to bolster the bilateral cooperation between China and Nepal in the field of botany and to document the plant wealth from the W Nepal, the first-ever Sino-Nepal Joint Botanical Expedition to Bajhang, Far West Nepal was organized and successfully carried out in 2017. The expedition was conducted from 9th – 21st September 2017, jointly by researchers from the Institute of Botany, Chinese Academy of Sciences (IBCAS) in collaboration with the Department of Plant Resources (DPR) and Research Centre for Applied Science and Technology, Tribhuvan University (RECAST, TU) from Nepal. In total, six researchers from the three institutes participated in the expedition led by Prof. Hai-nin Qin from IBCAS (Figure2), institutional affiliations of the participants is provided in Table 2.The primary focus of the expedition was to explore the Seti river valley, Bajhang district; however, some plant species along the Bhimdutt Highway and Jaya Prithvi Bahadur Singh Highway were also included, thus increasing the spatial coverage.

The herbarium specimens collected during the expedition were identified in Nepal and China, and a new record for Nepal was found. This paper aims to summarize the output of the expedition in terms of botanical novelties for the region and flora of Nepal.

Figure 2: Research team members with the supporting staffs at Dhalaun camp, Bajhang

Materials and Methods

Study area

The study area consisted mostly of Bajhang district of Seti zone, however, actual exploration started right from the lowland area of Kailali district. Specimens were collected alongside the roads, rivers and foot trails, mainly through the Bhimdutt Highway from Godawari (Kailali) to Khodpe (Baitadi), Jaya Prithvi Bahadur Singh Highway from Khodpe to Chainpur (Bajhang), from Chainpur along the Seti river upstream to Agara and thence forward along the foot trails to Dhalaun and adjoining areas. Details of the study area and exploration route are outlined in the map (Figure 3).

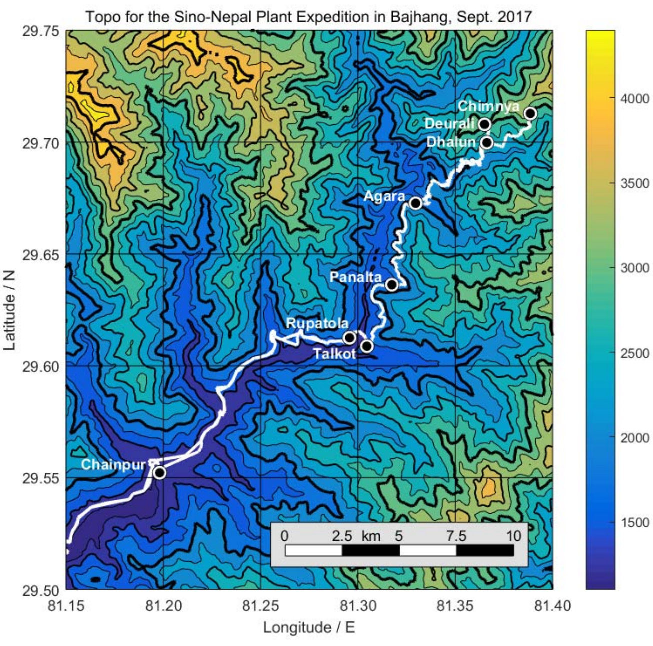

Figure 3: Collection route in Bajhang district (black dots in the map represent the major settlements and the camping sites)

Based on the already worked out plan, plant specimens were collected for herbarium preparation following the standard protocol (Forman & Bridson, 1989). For each specimen, maximum five duplicates were collected as per the availability, intended to be deposited at five important herbaria for Flora of Pan-Himalaya and Flora of Nepal, i.e. National Herbarium and Plant Laboratories (KATH), Chinese National Herbarium (PE), Tribhuvan University Central Herbarium (TUCH), Royal Botanical Garden Edinburgh Herbarium (E) and Herbarium of the University of Tokyo (TI). For thenationally protected and CITES-listed plants, only two specimens were collected, one each for KATH and TUCH. Following the fieldwork, specimens were identified through diverse means: expert opinion, use of standard literature (Hara et al., 1978; Polunin & Stainton, 1981; Stainton, 1984; Press et al., 2000; Wu & Raven, 2001) and herbarium consultation (KATH & PE).

Results and Discussion

During the expedition, total collection number reached 625, of which here we present a list of 503 identified species so far (Table 1), while the process of identification for the rest of the collected specimens is still underway.The list consists of enumeration of all the collections made during the field work irrespective of their recurrences as they represent different localities.

Among the observed species, 3 endemic plants- *Cirsium phulchokiense*, *Strobilanthes bheriense* and *Arenaria mukerjiana* are also reported from the region. Similarly, the study has shed light on the occurrence of invasive plants including *Ageratum haustonianum*, *Ageratum conyzoides*, *Erigeron karvinskianus*, *Bidens pilosa*, *Bidens biternata* and *Lantana camara*, that will help to study their distribution extent and impacts in future. Besides, this study has reinforced Bajhang district as a rich habitat for many medicinal plants that includes

Bergenia ciliata, *Swertia chirayita*, *Panax pseudoginseng*, *Senna tora*, *Taxus contorta*, *Artemisia dubia*, *Artemisia indica*, *Zanthoxylum armatum*, *Astilbe rivularis*, *Phyllanthus emblica* and *Calotropis gigantea*.

Going through the identification process, a shrub species was found intriguing and did not match with any of the previously reported species (Hara et al., 1979; Press et al., 2000, Shrestha et al., 2018; Rajbhandari & Rai, 2019) from Nepal. Later, the specimen was identified as *Itea nutans* Royle (Iteaceae).

Itea nutans was first described by Royle from Dehradun, Western Himalaya (Royle, 1839) as a member of family Saxifragaceae. Subsequently, Hooker (1879) mentioned the species as occurring in Garwhal and Kumaon at the elevation of 3000-5000 ft. Interestingly Royal Botanic Garden Edinburgh Herbarium (E) contains one specimen of *Itea nutans* collected from Nepal (Bhaskar Adhikari pers. comm. December 3, 2019). However, there is no mention of distribution of Iteaceae or *Itea* in the published volume of Flora of Nepal in the family account Saxifragaceae (Akiyama et al.,2011), neither the recently published literatures (Rajbhandari et al., 2011; Rajbhandari et al., 2015; Shrestha et al., 2018; Rajbhandari & Rai, 2019) has revealed the occurrence of the species in the country. Therefore, this paper is the first attempt to report the distribution of Iteaceae in Nepal.

Taxonomic treatment

Family: Iteaceae

Itea L. Sp. Pl. 1:199. 1753.

Shrubs to tree. Leaves alternate, petiolate, simple. Inflorescence terminal or axillary, racemose. Flowers pentamerous; ovary superior, bicarpellate, stigma capitate. Fruit a capsule.

About 27 species in the world, distributed from N America to Japan and South-East Asia through the Himalaya; one species in Nepal.

Itea nutans Royle, Illust. Bot. Himal. Mount. 226. 1835.Type: N. W. Himalaya, Dehra Dun, 2000 m, *Royle s.n.* (holotype: LIV).

Shrub, 3-5m tall. Stem with dark-grey bark. Leaves alternate, 5-15 cm long, 2-6 cm broad, ovate to elliptic-oblong, slightly tapering with acuminate apex; denticulate, adaxial surface glabrous, abaxialpubescent. Flowers greenish-white, ca. 2 mm long, borne in fascicles of 2-7, 10-20 cm long drooping racemes. Sepals ca. 1 mm long, as long as the calyx tube, linear, with acute apex, densely pubescent, persistent. Petals ca. 2.5 mm long, linear-oblong, erect, tips inflexed. Filaments 1.5 mm long, subulate. Anthers dorsifixed exerted. Ovary bilocular; styles grooved. Capsules 4 mm long, septicidally 2-valved.

Phenology: floweringApril to July, fruiting July to October.

Habitat: on moist slopes; 500-2000 m.

Distribution: W Himalaya; from Pakistan, Jammu & Kashmir (India) to W Nepal.

Specimen observed: W Nepal, Bajhang, Above Deuthala Kalanga River, 1000 m, 2017.9.20, Sino-Nepal Joint Expedition –H.N. Qin, P. Bhandari, T.R. Pandey, B.R. Subedee, Y.C. Yang & S.R. Zhang 620 (KATH) (Figure 4).

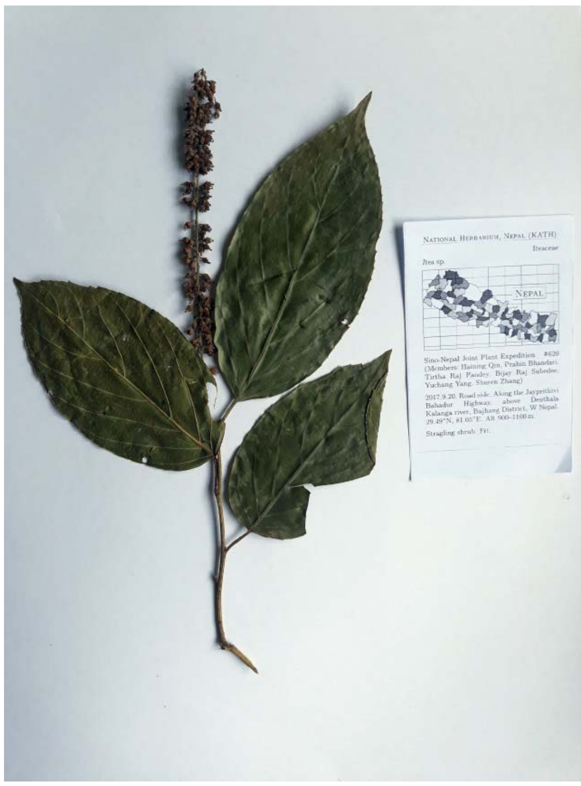

Figure 4: Herbarium specimen of *Itea nutans* (KATH)

Conclusion

This joint work has significantly contributed to represent the plant diversity of West Nepal in terms of total number of herbarium specimen collection, discovering new habitats and geographical range of endemic species, knowing the occurrence and expansion of invasive species and prospects of medicinal plants availability in the area.

Bilateral and multilateral cooperation among the botanical institutions is of key importance for better understanding the plant wealth of any region, and for the documentation and publication of flora account, periodic collection of herbarium specimens and reconfirming their taxonomic identity is undeniable. Exploring even the already represented region may often result in new records and may help to gather useful information regarding their distribution range, phenology, ecology, conservation status and impacts of climate change. The first Sino-Nepal Joint Expedition has therefore contributed to the better understanding of the flora of west Nepal. Also, one new record for Nepal has been added through the Sino-Nepal expedition, and it has paved the way forward for the prospects of many collaborative researches and scientific exchanges in the future.

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References

Akiyama, S., Gornall, R.J., Adhikari, B., Pendry C.A. & Watson, M.F.(2012). Watson et al. (Eds.) Flora of Nepal, Saxifragaceae web edition. http://www.floraofnepal.org/sys/scripts/accountlibrary/getaccount.php. (Accessed on 2019.12.7).

Forman, L. & Bridson, D. Eds. (1989). *The Herbarium Handbook*. Kew, UK: Royal Botanic Gardens

Hara, H., Stearn, W.T. & Williams, L.H.J. (1978). *An Enumeration of the Flowering Plants of Nepal*, British Museum (Natural History): Trustees of British Museum (Natural History).

Hong, D.-Y. (2015). *Flora of Pan-Himalaya*, Vol. 47. Beijing: Science Press.

Hooker, J.D. (1879). *The Flora of British India*, Vol. II: 408. London.

Polunin, O. & Stainton, A. (1981). *Flowers of the Himalaya*. New Delhi, India: Oxford University Press.

Press, J.R., Shrestha. K.K. & Sutton. D.A. (2000). Annotated checklist of the flowering plants of Nepal, London: The Natural History Museum.

Rajbhandari, K.R. (1976). History of botanical explorations in Nepal. *Journal of Bombay Natural History Society*, *73*(1).

Rajbhandari, K.R. (2016). History of Botanical Explorations in Nepal: 1802-2015. In P. K. Jha, M. Siwakoti & S. Rajbhandary (Eds.), *Frontiers of Botany*. Kathmandu, Nepal: Central Department of Botany, Tribhuvan University.

Rajbhandari, K.R., Bhattarai, K.R. & Baral, S.R. Eds. (2011). *Catalogue of Nepalese Flowering Plants-II*. Lalitpur, Nepal; National Herbarium and Plant Laboratories.

Rajbhandari, K.R., Bhatt, G.D., Chhetri, R. & Rai, S.K. (2015). *Catalogue of Nepalese Flowering Plants Supplement 1*. Lalitpur, Nepal: National Herbarium and Plant Laboratories.

Rajbhandari, K.R. & Rai, S.K. (2019). *A Handbook of the Flowering Plants of Nepal*, Vol. 2., Kathmandu, Nepal: Department of Plant Resources.

Royle, J. F. (1839). *Illustrations of the Botany and other branches of the natural history of the Himalayan Mountains*, Vol.1, 226. London.

Shrestha, K.K., Bhattarai, S. & Bhandari, P. (2018). *Handbook of Flowering plants of Nepal*, vol. 1. Jodhpur, India: Scientific Publishers,

Stainton, A. (1984). *Flowers of the Himalaya, A Supplement*. New Delhi, India: Oxford University Press.

Wu, Z.Y. & Raven, P.H. Eds. (2001). *Flora of China*. Vol. 8 (Brassicaceae through Saxifragaceae), Beijing: Science Press. & St. Louis, Missouri Botanical Garden Press.

Table 1: List of Plant species collected during the expedition

S.N.	Collection Number	Family	Scientific name	Habit
1	1	Begoniaceae	*Begonia picta* Wall.	Herbs
2	2	Cyperaceae	*Fimbristylis dichotoma* (L.) Vahl	Herbs
3	3	Droseraceae	*Drosera peltata* Thunb.	Herbs, Insectivorous
4	6	Cyperaceae	*Bulbostylis barbata* (Rottb.) C.B. Clarke	Herbs
5	7	Polypodiaceae	*Lepisorus bicolor* (Takeda) Ching	Herbs
6	8	Commelinaceae	*Cyanotis axillaris* (L.) D. Don ex Sweet	Herbs
7	9	Cyperaceae	*Fimbristylis dichotoma* (L.) Vahl	Herbs
8	10	Myricaceae	*Myrica esculenta* Buch.-Ham. ex D. Don	Trees, Edible fruits
9	11	Lamiaceae	*Scutellaria discolor* Colebr.	Herbs
10	12	Cyperaceae	*Kyllinga brevifolia* Rottb.	Herbs
11	13	Fabaceae	*Flemingia strobilifera* (L.) W.T.Aiton	Herbs
12	15	Asteraceae	*Ageratum houstonianum* Mill.	Herbs, Invasive
13	16	Asteraceae	*Ageratum conyzoides* L.	Herbs, Invasive
14	17	Asteraceae	*Erigeron karvinskianus* DC.	Herbs, Invasive
15	18	Asteraceae	*Bidens pilosa* L.	Herbs, Invasive
16	19	Zingiberaceae	*Hedychium spicatum* Sm.	Herbs
17	21	Urticaceae	*Girardinia diversifolia* (Link) Friis	Herbs, Fibre yielding
18	22	Commelinaceae	*Commelina maculata* Edgew.	Herbs
19	23	Fabaceae	*Butea minor* Buch.-Ham. ex Baker	Shrubs
20	24	Lentibulariaceae	*Utricularia bifida* L.	Herbs, Insectivorous
21	26	Cyperaceae	*Eriophorum comosum* (Wall.) Nees	Herbs
22	28	Cyperaceae	*Carex vesiculosa* Boott	Herbs
23	29	Cyperaceae	*Carex filicina* Nees	Herbs
24	30	Cyperaceae	*Fimbristylis complanata* (Retz.) Link	Herbs
25	31	Cyperaceae	*Fimbristylis dichotoma* (L.) Vahl	Herbs
26	32	Urticaceae	*Girardinia diversifolia* (Link) Friis	Herbs
27	33	Begoniaceae	*Begonia dioica* Buch.-Ham. ex D. Don	Herbs
28	36	Balsaminaceae	*Impatiens urticifolia* Wall.	Herbs
29	37	Cyperaceae	*Carex filicina* Nees	Herbs
30	39	Pteridaceae	*Adiantum philippense* L.	Herbs
31	40	Amaranthaceae	*Achyranthes aspera* L.	Herbs
32	41	Commelinaceae	*Commelina diffusa* Burm.f.	Herbs
33	42	Cyperaceae	*Cyperus compressus* L.	Herbs
34	43	Cyperaceae	*Cyperus compressus* L.	Herbs
35	44	Cyperaceae	*Fimbristylis ovata* (Burm.f.) J.Kern	Herbs
36	45	Fabaceae	*Senna tora* (L.) Roxb.	Shrubs, Medicinal
37	46	Lamiaceae	*Scutellaria repens* Buch.-Ham. ex D.Don	Herbs
38	47	Vitaceae	*Leea asiatica* (L.) Ridsdale	Herbs
39	48	Fabaceae	*Mimosa rubicaulis* Lam.	Herbs
40	49	Asteraceae	*Tridax procumbens* (L.) L.	Herbs
41	50	Commelinaceae	*Murdannia nudiflora* (L.) Brenan	Herbs
42	51	Fabaceae	*Desmodium laxiflorum* DC.	Shrubs
43	52	Vitaceae	*Ampelocissus divaricata* (Lawson) Planch.	Climbers
44	53	Convolvulaceae	*Ipomoea indica* (Burm.) Merr.	Herbs
45	54	Malvaceae	*Sida rhombifolia* L.	Shrubs
46	55	Boraginaceae	*Cynoglossum lanceolatum* Forssk.	Herbs
47	56	Buxaceae	*Sarcococca pruniformis* Lindl.	Herbs
48	57	Balsaminaceae	*Impatiens edgeworthii* Hook. f.	Herbs
49	58	Fumariaceae	*Corydalis chaerophylla* DC.	Herbs
50	59	Polygonaceae	*Polygonum recumbens* Royle ex Bab.	Herbs
51	60	Taxaceae	*Taxus contorta* Griff.	Trees, Medicinal

S.N.	Collection Number	Family	Scientific name	Habit
52	62	Poaceae	*Setaria parviflora* (Poir.) M.Kerguelen	Herbs
53	63	Asteraceae	*Artemisia indica* Willd.	Subshrubs, Medicinal
54	64	Asteraceae	*Xanthium strumarium* L.	Herbs
55	65	Polygonaceae	*Polygonum abbreviatum* Kom.	Herbs
56	67	Polygonaceae	*Fagopyrum acutatum* (Lehm.) Mansf. ex K.Hammer	Herbs
57	68	Asteraceae	*Galinsoga quadriradiata* Ruiz & Pav.	Herbs
58	69	Asteraceae	*Artemisia gmelinii* Weber ex Stechm.	Subshrubs
59	70	Ericaceae	*Lyonia ovalifolia* (Wall.) Drude	Trees
60	71	Betulaceae	*Alnus nepalensis* D. Don	Trees
61	72	Moraceae	*Ficus palmata* Forssk.	Shrubs
62	73	Poaceae	*Eleusine coracana* (L.) Gaertn.	Herbs, Cultivated
63	74	Urticaceae	*Boehmeria rugulosa* Wedd.	Shrubs
64	75	Urticaceae	*Boehmeria platyphylla* Buch.-Ham. ex D. Don	Herbs
65	76	Lamiaceae	*Callicarpa macrophylla* Vahl	Shrubs
66	79	Poaceae	*Arundinella setosa* Trin.	Herbs
67	80	Fabaceae	*Aeschynomene indica* L.	Herbs
68	81	Ranunculaceae	*Clematis roylei* Rehder	Climbers
69	82	Polygonaceae	*Persicaria barbata* (L.) H.Hara	Herbs
70	83	Melastomataceae	*Osbeckia stellata* Buch.-Ham. ex Ker Gawl.	Shrubs
71	84	Solanaceae	*Physalis angulata* L.	Herbs
72	85	Acanthaceae	*Barleria cristata* L.	Herbs
73	86	Urticaceae	*Urtica ardens* Link	Subshrubs
74	87	Solanaceae	*Datura stramonium* L.	Herbs
75	88	Gesneriaceae	*Rhynchoglossum obliquum* Blume	Herbs
76	89	Orchidaceae	*Vanda cristata* Wall. ex Lindl.	Herbs, Epiphytic
77	90	Acanthaceae	*Dicliptera bupleuroides* Nees	Herbs
78	91	Poaceae	*Oplismenus compositus* (L.) P.Beauv.	Herbs
79	92	Fabaceae	*Crotalaria prostrata* Willd.	Herbs
80	93	Mazaceae	*Mazus surculosus* D. Don	Herbs
81	95	Cyperaceae	*Cyperus cuspidatus* Kunth	Herbs
82	96	Linderniaceae	*Lindernia ciliata* (Colsm.) Pennell	Herbs
83	98	Linderniaceae	*Lindernia abyssinica* Engl.	Herbs
84	99	Fabaceae	*Desmodium triflorum* (L.) DC.	Herbs
85	100	Rubiaceae	*Oldenlandia corymbosa* L.	Herbs
86	102	Fabaceae	*Desmodium heterocarpon (*L.) DC.	Shrubs
87	103	Caryophyllaceae	*Drymaria cordata* (L.) Willd. ex Schult.	Herbs
88	105	Malvaceae	*Triumfetta rhomboidea* Jacq.	Herbs
89	106	Rosaceae	*Pyrus pashia* Buch.-Ham. ex D.Don	Trees
90	107	Cornaceae	*Alangium alpinum* (C.B. Clarke) W.W. Sm. & Cave	Trees
91	108	Asteraceae	*Cirsium phulchokiense* Kitam.	Herbs, Endemic
92	109	Balsaminaceae	*Impatiens scabrida* DC.	Herbs
93	110	Poaceae	*Echinochloa crus* galli (L.) P.Beauv.	Herbs
94	111	Poaceae	*Sporobolus fertilis* (Steud.) Clayton	Herbs
95	112	Cyperaceae	*Cyperus iria* L.	Herbs
96	113	Pteridaceae	*Adiantum philippense* L.	Herbs
97	114	Orobanchaceae	*Lindenbergia indica* Vatke	Herbs
98	115	Fabaceae	*Parochetus communis* D.Don	Herbs
99	116	Cyperaceae	*Kyllinga brevifolia* Rottb.	Herbs
100	117	Cyperaceae	*Kyllinga squamulata* Vahl	Herbs
101	118	Malvaceae	*Abelmoschus moschatus* Medik.	Subshrubs
102	119	Vitaceae	*Parthenocissus semicordata* (Wall.) Planch.	Climbers
103	120	Lamiaceae	*Isodon coesta* (Buch.-Ham. ex D.Don) Kudo	Herbs
104	121	Fabaceae	*Desmodium podocarpum* DC.	Herbs

S.N.	Collection Number	Family	Scientific name	Habit
105	122	Gesneriaceae	*Henckelia pumila* (D.Don) A.Dietr.	Herbs
106	124	Fagaceae	*Quercus lanata* Sm.	Trees
107	125	Fagaceae	*Quercus lanata* Sm.	Trees
108	127	Orchidaceae	*Malaxis abieticola* Salazar & Soto Arenas	Herbs, Terrestrial
109	128	Piperaceae	*Peperomia tetraphylla* (G.Forst.) Hook. & Arn.	Herbs
110	129	Poaceae	*Drepanostachyum falcatum* (Nees) Keng f.	Shrubs
111	130	Ericaceae	*Rhododendron arboreum* Sm. var. arboreum	Trees
112	131	Cyperaceae	*Carex filicina* Nees	Herbs
113	132	Cyperaceae	*Bulbostylis densa* (Wall.) Hand.-Mazz.	Herbs
114	134	Betulaceae	*Carpinus faginea* Lindl.	Trees
115	135	Poaceae	*Eragrostis minor* Host	Herbs
116	137	Asteraceae	*Pseudognaphalium hypoleucum* (DC.) Hill. &Burtt	Herbs
117	138	Cyperaceae	*Pycreus sanguinolentus* (Vahl) Nees	Herbs
118	140	Rubiaceae	*Leptodermis kumaonensis* R. Parker	Shrubs
119	141	Commelinaceae	*Cyanotis cristata* (L.) D. Don	Herbs
120	142	Polygonaceae	*Persicaria capitata* (Buch.-Ham. ex D.Don) H.Gross	Herbs
121	144	Poaceae	*Pseudopogonatherum contortum* (Brongn.) A. Camus	Herbs
122	146	Cyperaceae	*Fimbristylis dichotoma* (L.) Vahl	Herbs
123	148	Dryopteridaceae	*Polystichum squarrosum* (D. Don) Fée	Herbs
124	150	Sinopteridaceae	*Aleuritopteris albomarginata* (C.B. Clarke) Ching	Herbs
125	153	Rubiaceae	*Galium acutum* Edgew.	Herbs
126	154	Asteraceae	*Anaphalis busua* (Buch.-Ham.) DC.	Herbs
127	155	Lamiaceae	*Isodon scrophularioides* (Wall. ex Benth.) Murata	Herbs
128	156	Boraginaceae	*Cynoglossum wallichii* var. *glochidiatum* (Wall. ex Benth.) Kazmi	Herbs
129	157	Scrophulariaceae	*Scrophularia urticifolia* Wall. ex Benth.	Herbs
130	158	Hypoxidaceae	*Hypoxis aurea* Lour.	Herbs
131	159	Rosaceae	*Agrimonia pilosa* Ledeb.	Herbs
132	160	Hypericaceae	*Hypericum elodeoides* Choisy	Herbs
133	161	Polypodiaceae	*Lepisorus bicolor* (Takeda) Ching	Herbs
134	162	Lamiaceae	*Salvia plebeia* R.Br.	Herbs
135	163	Asteraceae	*Anaphalis adnata* DC.	Herbs
136	164	Fabaceae	*Rhynchosia falconeri* Baker	Herbs
137	165	Asteraceae	*Anaphalis chlamydophylla* Diels	Herbs
138	166	Fabaceae	*Indigofera dosua* D.Don	Shrubs
139	167	Gentianaceae	*Swertia angustifolia* Buch.-Ham. ex D. Don	Herbs
140	168	Caprifoliaceae	*Valeriana hardwickii* Wall.	Herbs
141	169	Asteraceae	*Duhaldea cappa* (Buch.-Ham.ex D.Don) Pruski &Anderb.	Herbs
142	170	Lamiaceae	*Salvia coccinea* Buc'hoz ex Etl.	Herbs
143	171	Polypodiaceae	*Drynaria propinqua* (Wall. ex Mett.) Bedd.	Herbs
144	172	Onagraceae	*Epilobium cylindricum* D.Don	Herbs
145	174	Orchidaceae	*Satyrium nepalense* D.Don	Herbs, Terrestrial
146	175	Scrophulariaceae	*Verbascum thapsus* L.	Herbs
147	177	Droseraceae	*Drosera peltata* Thunb.	Herbs, Insectivorous
148	178	Urticaceae	*Lecanthus peduncularis* (Wall. ex Royle) Wedd.	Herbs
149	180	Hypericaceae	*Hypericum japonicum* Thunb.	Herbs
150	182	Dioscoreaceae	*Dioscorea bulbifera* L.	Herbs, Climber
151	183	Fabaceae	*Smithia ciliata* Royle	Herbs
152	184	Polygalaceae	*Salomonia cantoniensis* Lour.	Herbs
153	185	Primulaceae	*Androsace sarmentosa* Wall.	Herbs
154	186	Poaceae	*Oplismenus burmanni* (Retz.) P.Beauv.	Herbs
155	189	Poaceae	*Arthraxon hispidus* (Thunb.) Makino	Herbs
156	191	Poaceae	*Chamabainia cuspidata* Wight	Herbs

S.N.	Collection Number	Family	Scientific name	Habit
157	193	Amaranthaceae	*Cyathula tomentosa* (Roth) Moq.	Shrubs
158	194	Asteraceae	*Tagetes minuta* L.	Herbs
159	196	Aceraceae	*Acer oblongum* Wall. ex DC.	Trees
160	197	Anacardiaceae	*Toxicodendron wallichii* (Hook. f.) Kuntze	Shrubs
161	198	Myricaceae	*Myrica esculenta* Buch.-Ham. ex D. Don	Trees, Edible fruits
162	199	Fabaceae	*Rhynchosia falconeri* Baker	Herbs
163	201	Rutaceae	*Zanthoxylum armatum* DC.	Shrub, Medicinal
164	202	Urticaceae	*Girardinia diversifolia* (Link) Friis	Herbs, Fiber yielding
165	204	Zingiberaceae	*Hedychium spicatum* Sm.	Herbs
166	205	Orobanchaceae	*Pedicularis bifida* (Buch.-Ham.) Pennell	Herbs
167	206	Ericaceae	*Rhododendron arboreum* Sm. var. arboreum	Trees
168	207	Amaranthaceae	*Achyranthes bidentata* Blume	Herbs
169	208	Fabaceae	*Desmodium microphyllum* (Thunb.) DC.	Herbs
170	209	Fabaceae	*Indigofera heterantha* Brandis	Shrubs
171	210	Lamiaceae	*Leucas lanata* Benth.	Herbs
172	211	Fabaceae	*Desmodium multiflorum* DC.	Shrubs
173	212	Acanthaceae	*Strobilanthes bheriensis* (Shakya) J.R.I.Wood	Herbs, Endemic
174	213	Polygalaceae	*Polygala persicariifolia* DC.	Herbs
175	214	Campanulaceae	*Campanula pallida* Wall.	Herbs
176	215	Primulaceae	*Lysimachia alternifolia* Wall.	Herbs
177	216	Acanthaceae	*Justicia japonica* Thunb.	Herbs
178	217	Caprifoliaceae	*Valeriana hardwickii* Wall.	Herbs
179	219	Convolvulaceae	*Ipomoea hederifolia* L.	Herbs
180	220	Rubiaceae	*Rubia alata* Wall.	Herbs
181	221	Dioscoreaceae	*Dioscorea belophylla* (Prain) Voigt ex Haines	Herbs, Climber
182	223	Lamiaceae	*Origanum vulgare* L.	Herbs
183	224	Fabaceae	*Desmodium elegans* DC.	Shrubs
184	225	Oleaceae	*Ligustrum compactum* (Wall. ex G.Don) Hook.f. & Thomson ex Brandis	Shrubs
185	227	Berberidaceae	*Berberis glaucocarpa* Stapf	Herbs
186	228	Fabaceae	*Albizia chinensis* (Osbeck) Merr.	Trees
187	229	Toricelliaceae	*Toricellia tiliifolia* DC.	Trees
188	230	Rutaceae	*Boenninghausenia albiflora* (Hook.) Rchb. ex Meisn.	Herbs
189	232	Rutaceae	*Zanthoxylum acanthopodium* DC.	Shrubs
190	233	Cucurbitaceae	*Solena amplexicaulis* (Lam.) Gandhi	Herbs
191	234	Caryophyllaceae	*Drymaria cordata* (L.) Willd. ex Schult.	Herbs
192	235	Lamiaceae	*Isodoncoesta* (Buch.-Ham. ex D.Don) Kudo	Herbs
193	236	Urticaceae	*Elatostema monandrum* (Buch.-Ham. ex D.Don) H.Hara	Herbs
194	237	Lamiaceae	*Isodon coetsa* (Buch.-Ham. ex D.Don) Kudô	Herbs
195	238	Urticaceae	*Chamabainia cuspidata* Wight	Herbs
196	239	Caprifoliaceae	*Viburnum mullaha* Buch.-Ham. ex D. Don	Shrubs
197	240	Asteraceae	*Conyza japonica* (Thunb.) Less. ex Less.	Herbs
198	241	Begoniaceae	*Begonia picta* Wall.	Herbs
199	242	Lamiaceae	*Origanum vulgare* L.	Herbs
200	244	Asteraceae	*Erigeron bellidioides* (Buch.-Ham. ex D.Don) Benth. ex C.B.Clarke	Herbs
201	245	Lamiaceae	*Elsholtzia blanda* (Benth.) Benth.	Herbs
202	246	Urticaceae	*Pilea scripta* (Buch.-Ham. ex D. Don) Wedd.	Herbs
203	247	Thymelaeaceae	*Daphne bholua* Buch.-Ham. ex D.Don	Shrubs
204	248	Rosaceae	*Rosa moschata* Herrm.	Shrubs
205	249	Caprifoliaceae	*Viburnum mullaha* Buch.-Ham. ex D. Don	Shrubs
206	250	Oleaceae	*Jasminum officinale* L.	Shrubs
207	251	Smilacaceae	*Smilax aspera* L.	Climbers

S.N.	Collection Number	Family	Scientific name	Habit
208	252	Rosaceae	*Prinsepia utilis* Royle	Shrubs
209	253	Cuscutaceae	*Cuscuta reflexa* Roxb.	Climbers, Parasitic
210	254	Dipsacaceae	*Dipsacus inermis* Wall.	Herbs
211	256	Asteraceae	*Senecio graciliflorus* (Wall.) DC.	Herbs
212	258	Crassulaceae	*Sedum multicaule* Wall. ex Lindl.	Herbs
213	259	Geraniaceae	*Geranium nepalense* Sweet	Herbs
214	260	Begoniaceae	*Begonia picta* Wall.	Herbs
215	261	Hypericaceae	*Hypericum hookerianum* Wight & Arn.	Shrubs
216	262	Violaceae	*Viola canescens* Wall.	Herbs
217	263	Asteraceae	*Lactuca brunoniana* (DC.) Wall. ex C.B.Clarke	Herbs
218	264	Dennstaedtiaceae	*Microlepia firma* Mett. ex Kuhn	Herbs
219	265	Piperaceae	*Peperomia heyneana* Miq.	Herbs
220	266	Lamiaceae	*Elsholtzia ciliata* (Thunb.) Hyl.	Herbs
221	268	Rubiaceae	*Galium asperifolium* Wall.	Herbs
222	270	Lamiaceae	*Leucas cephalotes* (Roth) Spreng.	Herbs
223	271	Campanulaceae	*Lobelia nicotianifolia* Roth ex Schult.	Herbs
224	272	Fabaceae	*Senna occidentalis* (L.) Link	Shrubs
225	273	Fabaceae	*Flemingia strobilifera* (L.) W.T.Aiton	Shrubs
226	274	Saxifragaceae	*Astilbe rivularis* Buch.-Ham. ex D. Don	Herbs, Medicinal
227	275	Fabaceae	*Indigofera heterantha* Brandis	Shrubs
228	276	Ranunculaceae	*Clematis gouriana* Roxb. ex DC.	Herbs
229	277	Zingiberaceae	*Hedychium spicatum* Sm.	Herbs
230	278	Ranunculaceae	*Anemone vitifolia* Buch.-Ham. ex DC.	Herbs
231	279	Gentianaceae	*Gentiana capitata* Buch.-Ham. ex D.Don	Herbs
232	280	Cyperaceae	*Cyperus squarrosus* L.	Herbs
233	281	Malvaceae	*Urena repanda* Roxb. ex Sm.	Herbs
234	282	Polygalaceae	*Polygala crotalarioides* Buch.-Ham. ex DC.	Herbs
235	283	Asteraceae	*Anaphalis triplinervis* (Sims) C.B. Clarke	Herbs
236	285	Anacardiaceae	*Brucea javanica* (L.) Merr.	Trees
237	286	Vitaceae	*Ampelocissus rugosa* (Wall.) Planch.	Climber
238	287	Euphorbiaceae	*Leptopus cordifolius* Decne.	Shrubs
239	288	Rosaceae	*Spiraea canescens* D.Don	Shrubs
240	289	Orchidaceae	*Satyrium nepalense* D.Don	Herbs, Terrestrial
241	290	Begoniaceae	*Begonia picta* Wall.	Herbs
242	291	Solanaceae	*Nicandra physalodes* (L.) Gaertn.	Herbs
243	292	Fagaceae	*Quercus lanata* Sm.	Trees
244	293	Zingiberaceae	*Cautleya spicata* (Sm.) Baker	Herbs
245	294	Menispermaceae	*Stephania glabra* (Roxb.) Miers	Herbs
246	295	Urticaceae	*Boehmeria polystachya* Wedd.	Herbs
247	296	Asteraceae	*Gynura bicolor* (Roxb. ex Willd.) DC.	Herbs
248	299	Poaceae	*Dichanthium annulatum* (Forssk.) Stapf	Herbs
249	300	Equisataceae	*Equisetum arvense* L.	Herbs
250	301	Balsaminaceae	*Impatiens edgeworthii* Hook. f.	Herbs
251	302	Lamiaceae	*Origanum vulgare* L.	Herbs
252	303	Cyperaceae	*Bulbostylis densa* (Wall.) Hand.-Mazz.	Herbs
253	305	Polygonaceae	*Aconogonum molle* (D. Don) H. Hara	Herbs
254	306	Lentibulariaceae	*Utricularia scandens* Benj.	Herbs, Insectivorous
255	307	Poaceae	*Saccharum spontaneum* L.	Herbs
256	308	Zingiberaceae	*Hedychium spicatum* Sm.	Herbs
257	309	Poaceae	*Drepanostachyum falcatum* (Nees) Keng f.	Shrubs
258	310	Polypodiaceae	*Polypodiodes lachnopus* (Wall. ex Hook.) Ching	Herbs
259	311	Poaceae	*Festuca gigantea* (L.) Vill.	Herbs
260	312	Cyperaceae	*Carex myosurus* Nees	Herbs

S.N.	Collection Number	Family	Scientific name	Habit
261	313	Eriocaulaceae	*Eriocaulon nepalense* Prescott ex Bong.	Herbs
262	314	Equisataceae	*Equisetum arvense* L.	Herbs
263	315	Asteraceae	*Sigesbeckia orientalis* L.	Herbs
264	316	Lamiaceae	*Prunella vulgaris* L.	Herbs
265	318	Ranunculaceae	*Anemone rivularis* Buch.-Ham. ex DC.	Herbs
266	319	Ranunculaceae	*Ranunculus laetus* Wall. ex Hook. f. & J.W. Thomson	Herbs
267	320	Urticaceae	*Lecanthus peduncularis* (Wall. ex Royle) Wedd.	Herbs
268	321	Saxifragaceae	*Bergenia ciliata* Sternb.	Herbs
269	322	Polygonaceae	*Persicaria lapathifolia* (L.) Delarbre	Herbs
270	323	Rosaceae	*Prinsepia utilis* Royle	Shrubs
271	326	Lamiaceae	*Phlomoides macrophylla* (Benth.) Kamelin & Makhm.	Herbs
272	327	Pteridaceae	*Notholaena himalaica* Fraser-Jenk.	Herbs
273	328	Asteraceae	*Artemisia dubia* Wall. ex Besser	Subshrubs
274	329	Rubiaceae	*Galium asperifolium* Wall.	Herbs
275	331	Asteraceae	*Bidens biternata* (Lour.) Merr. & Sherff	Herbs, Invasive
276	332	Gentianaceae	*Swertia chirayita* (Roxb.) Buch.-Ham. ex C.B.Clarke	Herbs, Medicinal
277	334	Asteraceae	*Adenostemma lavenia* (L.) Kuntze	Herbs
278	336	Polypodiaceae	*Polypodiodes lachnopus* (Wall. ex Hook.) Ching	Herbs
279	338	Urticaceae	*Elatostema monandrum* (Buch.-Ham. ex D.Don) H.Hara	Herbs
280	339	Liliaceae	*Disporum cantoniense* (Lour.) Merr.	Herbs
281	341	Symplocaceae	*Symplocos paniculata* (Thunb.) Miq.	Trees
282	342	Hydrangeaceae	*Philadelphus tomentosus* Wall. ex G. Don	Shrubs
283	343	Rhamnaceae sp.	*Rhamnus virgatus* Roxb.	Shrubs
284	346	Caryophyllaceae	*Stellaria monosperma* Buch.-Ham. ex D. Don	Herbs
285	347	Gesneriaceae	*Platystemma violoides* Wall.	Herbs
286	348	Brassicaceae	*Cardamine flexuosa* With.	Herbs
287	349	Asteraceae	*Bidens biternata* (Lour.) Merr. & Sherff	Herbs, Invasive
288	350	Polypodiaceae	*Microsorum membranaceum* (D. Don) Ching	Herbs
289	351	Apiaceae	*Chaerophyllum reflexum* Aitch.	Herbs
290	352	Cornaceae	*Cornus macrophylla* Wall.	Trees
291	353	Aquifoliaceae	*Ilex dipyrena* Wall.	Trees
292	354	Aceraceae	*Acer cappadocicum* Gled.	Trees
293	355	Gesneriaceae	*Didymocarpus albicalyx* C.B. Clarke	Herbs
294	356	Theaceae	*Eurya acuminata* DC.	Trees
295	357	Elaeagnaceae	*Elaeagnus umbellata* Thunb.	Shrubs
296	358	Fagaceae	*Quercus glauca* Thunb.	Trees
297	359	Cyperaceae	*Carex condensata* Nees	Herbs
298	360	Rosaceae	*Spiraea canescens* D.Don	Shrubs
299	361	Rosaceae	*Pyracantha crenulata* (Roxb. ex D.Don) M.Roem.	Shrubs
300	362	Polygonaceae	*Polygonum recumbens* Royle ex Bab.	Herbs
301	363	Orchidaceae	*Habenaria intermedia* D.Don	Herbs
302	365	Loranthaceae	*Scurrula parasitica* L.	Shrubs
303	366	Rosaceae	*Cotoneaster frigidus* Wall. ex Lindl.	Shrubs
304	369	Araceae	*Arisaema erubescens* (Wall.) Schott	Herbs
305	370	Poaceae	*Bromus himalaicus* Stapf	Herbs
306	373	Gentianaceae	*Swertia paniculata* Wall.	Herbs
307	374	Ranunculaceae	*Clematis connata* DC.	Herbs, Climber
308	376	Malvaceae	*Urena lobata* L.	Herbs
309	377	Cyperaceae	*Pycreus sanguinolentus* (Vahl) Nees	Herbs
310	378	Campanulaceae	*Codonopsis viridis* Wall.	Herbs
311	380	Apocynaceae	*Ceropegia longifolia* Wall.	Herbs, Climber
312	382	Balsaminaceae	*Impatiens urticifolia* Wall.	Herbs
313	383	Polypodiaceae	*Lepisorus bicolor* (Takeda) Ching	Herbs

S.N.	Collection Number	Family	Scientific name	Habit
314	384	Fabaceae	*Uraria lagopus* DC.	Herbs
315	385	Poaceae	*Heteropogon contortus* (L.) P.Beauv. ex Roem. & Schult.	Herbs
316	388	Cyperaceae	*Cyperus cyperoides* (L.) Kuntze	Herbs
317	389	Cyperaceae	*Fimbristylis dichotoma* (L.) Vahl	Herbs
318	390	Rosaceae	*Cotoneaster microphyllus* Wall. ex Lindl.	Shrubs
319	391	Moraceae	*Ficus sarmentosa* Buch.-Ham. ex Sm.	Shrubs
320	392	Pinaceae	*Tsuga dumosa* (D.Don) Eichler	Trees
321	393	Rosaceae	*Cotoneaster bacillaris* Wall. ex Lindl.	Shrubs
322	394	Asteraceae	*Synotis wallichii* (DC.) C.Jeffrey & Y.L.Chen	Herbs
323	395	Asteraceae	*Carpesium abrotanoides* L.	Herbs
324	396	Plantaginaceae	*Wulfeniopsis amherstiana* (Benth.) D.Y. Hong	Herbs
325	397	Ericaceae	*Gaultheria nummularioides* D.Don	Herbs
326	398	Onagraceae	*Circaea repens* Wall. ex Asch. & Magnus	Herbs
327	399	Urticaceae	*Elatostema pusillum* C.B. Clarke ex Hook. f.	Herbs
328	402	Asteraceae	*Anaphalis triplinervis* (Sims) C.B. Clarke	Herbs
329	403	Athyriaceae	*Athyrium pectinatum* (Wall. ex Mett.) T. Moore	Herbs
330	404	Polypodiaceae	*Drynaria mollis* Bedd.	Herbs
331	405	Rosaceae	*Potentilla lineata* Trevir.	Herbs
332	406	Cannabaceae	*Cannabis sativa* L.	Herbs
333	407	Gentianaceae	*Swertia nervosa* (Wall. ex G. Don) C.B. Clarke	Herbs
334	408	Gentianaceae	*Halenia elliptica* D.Don	Herbs
335	409	Rosaceae	*Rubus nepalensis* (Hook. f.) Kuntze	Herbs
336	410	Lamiaceae	*Elsholtzia fruticosa* (D.Don) Rehder	Herbs
337	411	Plantaginaceae	*Plantago asiatica* subsp. erosa (Wall.) Z.Yu Li	Herbs
338	412	Polygonaceae	*Rumex nepalensis* Spreng.	Herbs
339	414	Asteraceae	*Taraxacum mitalii* Soest	Herbs
340	415	Poaceae	*Capillipedium parviflorum* (R.Br.) Stapf	Herbs
341	416	Plantaginaceae	*Hemiphragma heterophyllum* Wall.	Herbs
342	417	Polygonaceae	*Bistorta amplexicaulis* (D. Don) Greene	Herbs
343	418	Asteraceae	*Senecio cappa* Buch.-Ham. ex D.Don	Herbs
344	419	Asteraceae	*Lactuca violifolia* (Decne.) C.B.Clarke	Herbs
345	420	Poaceae	*Poa annua* L.	Herbs
346	423	Cyperaceae	*Isolepis setacea* (L.) R.Br.	Herbs
347	424	Papaveraceae	*Corydalis stipulata* Lidén	Herbs
348	425	Plantaginaceae	*Veronica cana* Wall. ex Benth.	Herbs
349	427	Cyperaceae	*Carex filicina* Nees	Herbs
350	428	Gentianaceae	*Swertia nervosa* (Wall. ex G. Don) C.B. Clarke	Herbs
351	429	Caryophyllaceae	*Silene kumaonensis* F.N.Williams	Herbs
352	431	Poaceae	*Themeda triandra* Forssk.	Herbs
353	432	Saxifragaceae	*Saxifraga parnassifolia* D. Don	Herbs
354	433	Fabaceae	*Lespedeza gerardiana* Maxim.	Herbs
355	436	Rubiaceae	*Galium acutum* Edgew.	Herbs
356	438	Urticaceae	*Pilea umbrosa* Blume	Herbs
357	439	Apiaceae	*Sanicula elata* Buch.-Ham. ex D.Don	Herbs
358	441	Crassulaceae	*Rhodiola chrysanthemifolia* (H. Lév.) S.H. Fu	Herbs
359	442	Apocynaceae	*Vincetoxicum hirundinaria* Medik.	Herbs
360	443	Fabaceae	*Vicia bakeri* Ali	Herbs
361	445	Poaceae	*Stipa roylei* (Nees) Duthie	Herbs
362	446	Cyperaceae	*Carex vesiculosa* Boott	Herbs
363	447	Rosaceae	*Cotoneaster bacillaris* Wall. ex Lindl.	Shrubs
364	450	Crassulaceae	*Rhodiola chrysanthemifolia* (H. Lév.) S.H. Fu	Herbs
365	451	Orobanchaceae	*Pedicularis scullyana* Prain ex Maxim.	Herbs
366	452	Asteraceae	*Senecio raphanifolius* Wall. ex DC.	Herbs

S.N.	Collection Number	Family	Scientific name	Habit
367	453	Campanulaceae	*Campanula pallida* Wall.	Herbs
368	455	Polypodiaceae	*Drynaria propinqua* (Wall. ex Mett.) Bedd.	Herbs
369	458	Rosaceae	*Dasiphora fruticosa* (L.) Rydb.	Herbs
370	459	Ericaceae	*Rhododendron lepidotum* Wall. ex G. Don	Shrubs
371	461	Orchidaceae	*Satyrium nepalense* D.Don	Herbs, Terrestrial
372	462	Euphorbiaceae	*Euphorbia sikkimensis* Boiss.	Shrubs
373	465	Asteraceae	*Senecio raphanifolius* Wall. ex DC.	Herbs
374	467	Campanulaceae	*Cyananthus microphyllus* Edgew.	Herbs
375	468	Ericaceae	*Rhododendron arboreum* Sm. var. arboreum	Trees
376	469	Rosaceae	*Rosa webbiana* Wall. ex Royle	Shrubs
377	470	Rosaceae	*Potentilla griffithii* Hook.f.	Herbs
378	472	Balsaminaceae	*Impatiens puberula* DC.	Herbs
379	473	Pinaceae	*Pinus wallichiana* A.B.Jacks.	Trees
380	474	Ericaceae	*Rhododendron barbatum* Wall. ex G. Don	Trees
381	475	Fagaceae	*Quercus semecarpifolia* Sm.	Trees
382	476	Asteraceae	*Lactuca violifolia* (Decne.) C.B.Clarke	Herbs
383	477	Caryophyllaceae	*Silene kumaonensis* F.N.Williams	Herbs
384	478	Rosaceae	*Sorbus lanata* (D.Don) S.Schauer	Trees
385	479	Aceraceae	*Acer acuminatum* Wall. ex D. Don	Trees
386	480	Taxaceae	*Taxus contorta* Griff.	Trees, Medicinal
387	481	Caprifoliaceae	*Viburnum cotinifolium* D. Don	Shrubs
388	482	Pinaceae	*Abies spectabilis* (D. Don) Spach	Trees
389	483	Hypericaceae	*Hypericum choisianum* Wall. ex N.Robson	Shrubs
390	484	Caprifoliaceae	*Leycesteria formosa* Wall.	Shrubs
391	487	Acanthaceae	*Strobilanthes urticifolia* Wall. ex Kuntze	Herbs
392	488	Polypodiaceae	*Drynaria propinqua* (Wall. ex Mett.) Bedd.	Herbs
393	489	Athyriaceae	*Athyrium pectinatum* (Wall. ex Mett.) T. Moore	Herbs
394	491	Polygonaceae	*Salomonia cantoniensis* Lour.	Herbs
395	492	Liliaceae	*Polygonatum verticillatum* (L.) All.	Herbs
396	494	Hydrangeaceae	*Hydrangea robusta* Hook. f. & Thomson	Shrubs
397	495	Caryophyllaceae	*Arenaria mukerjeeana* H. Hara	Herbs, Endemic
398	496	Cyperaceae	*Bulbostylis densa* (Wall.) Hand.-Mazz.	Herbs
399	498	Smilacaceae	*Smilax menispermoidea* A.DC.	Herbs
400	500	Salicaceae	*Salix denticulata* Andersson	Shrubs
401	501	Fagaceae	*Quercus semecarpifolia* Sm.	Trees
402	502	Schisandraceae	*Schisandra grandiflora* (Wall.) Hook.f. & Thomson	Shrubs
403	503	Balsaminaceae	*Impatiens sulcata* Wall.	Herbs
404	504	Poaceae	*Bromus himalaicus* Stapf	Herbs
405	505	Hypericaceae	*Hypericum choisianum* Wall. ex N.Robson	Shrubs
406	507	Liliaceae	*Lilium nepalense* D.Don	Herbs
407	509	Asparagaceae	*Maianthemum fuscum* (Wall.) LaFrankie	Herbs
408	510	Lamiaceae	*Ajuga macrosperma* Wall. ex Benth.	Herbs
409	511	Lamiaceae	*Prunella vulgaris* L.	Herbs
410	512	Papaveraceae	*Corydalis stipulata* Lidén	Herbs
411	513	Araliaceae	*Panax pseudoginseng* Wall.	Herbs, Medicinal
412	515	Aceraceae	*Acer caesium* Wall. ex Brandis	Trees
413	516	Asparagaceae	*Asparagus filicinus* Buch.-Ham. ex D. Don	Herbs
414	517	Lamiaceae	*Orthosiphon incurvus* Benth.	Herbs
415	519	Cyperaceae	*Cyperus squarrosus* L.	Herbs
416	520	Cyperaceae	*Kyllinga squamulata* Vahl	Herbs
417	521	Urticaceae	*Laportea bulbifera* (Siebold & Zucc.) Wedd.	Herbs
418	522	Apiaceae	*Pimpinella diversifolia* DC.	Herbs
419	523	Asteraceae	*Taraxacum parvulum* DC.	Herbs

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420	525	Primulaceae	*Androsace geraniifolia* Watt	Herbs
421	527	Pteridaceae	*Aleuritopteris bicolor* (Roxb.) Fraser-Jenk.	Herbs
422	529	Loranthaceae	*Scurrula elata* (Edgew.) Danser	Shrubs
423	530	Lamiaceae	*Phlomoides spectabilis* (Falc. ex Benth.) Kamelin & Makhm.	Herbs
424	531	Oleaceae	*Jasminum polyanthum* Franch.	Shrubs
425	532	Betulaceae	*Betula alnoides* Buch.-Ham. ex D. Don	Trees
426	533	Amaranthaceae	*Celosia argentea* L.	Herbs
427	535	Elaeagnaceae	*Elaeagnus umbellata* Thunb.	Shrubs
428	536	Solanaceae	*Solanum americanum* Mill.	Herbs
429	537	Cuscutaceae	*Cuscuta reflexa* Roxb.	Climbers, Parasitic
430	538	Larzidabalaceae	*Holboellia latifolia* Wall.	Climbers
431	539	Caprifoliaceae	*Lonicera lanceolata* Wall.	Shrubs
432	540	Rosaceae	*Malus baccata* (L.) Borkh.	Trees
433	541	Rosaceae	*Cotoneaster adpressus* Bois	Shrubs
434	543	Cyperaceae	*Carex nubigena* D.Don ex Tilloch & Taylor	Herbs
435	545	Ranunculaceae	*Thalictrum alpinum* L.	Herbs
436	546	Cyperaceae	*Kyllinga brevifolia* Rottb.	Herbs
437	547	Ranunculaceae	*Anemone obtusiloba* D. Don	Herbs
438	548	Amaranthaceae	*Cyathula capitata* Moq.	Herbs
439	549	Ranunculaceae	*Thalictrum chelidonii* DC.	Herbs
440	550	Ranunculaceae	*Clematis grata* Wall.	Herbs, Climber
441	551	Rosaceae	*Prunus venosa* Koehne	Trees
442	552	Lamiaceae	*Orthosiphon incurvus* Benth.	Herbs
443	553	Asteraceae	*Lactuca brunoniana* (DC.) Wall. ex C.B.Clarke	Herbs
444	554	Poaceae	*Digitaria ciliata* Lag.	Herbs
445	555	Asteraceae	*Tricholepis furcata* DC.	Herbs
446	558	Onagraceae	*Epilobium wallichianum* Hausskn.	Herbs
447	559	Poaceae	*Agrostis micrantha* Steud.	Herbs
448	560	Liliaceae	*Cardiocrinum giganteum* (Wall.) Makino	Herbs
449	562	Hydrangeaceae	*Hydrangea macrophylla* (Thunb.) Ser.	Shrubs
450	563	Sabiaceae	*Meliosma dilleniifolia* (Wall. ex Wight & Arn.) Walp.	Trees
451	564	Betulaceae	*Carpinus viminea* Wall. ex Lindl.	Trees
452	565	Fabaceae	*Desmodium elegans* DC.	Shrubs
453	566	Phytolaccaceae	*Phytolacca acinosa* Roxb.	Herbs, Vegetable
454	569	Caryophyllaceae	*Stellaria himalayensis* Majumdar	Herbs
455	570	Poaceae	*Drepanostachyum intermedium* (Munro) Keng f.	Shrubs
456	571	Lauraceae	*Lindera pulcherrima* (Nees) Hook. f.	Trees
457	572	Fagaceae	*Quercus semecarpifolia* Sm.	Trees
458	573	Urticaceae	*Lecanthus peduncularis* (Wall. ex Royle) Wedd.	Herbs
459	574	Lamiaceae	*Isodon scrophularioides* (Wall. ex Benth.) Murata	Herbs
460	575	Lamiaceae	*Elsholtzia ciliata* (Thunb.) Hyl.	Herbs
461	576	Gesneriaceae	*Platystemma violoides* Wall.	Herbs
462	577	Sapindaceae	*Aesculus indica* (Wall. ex Cambess.) Hook.	Trees
463	578	Pinaceae	*Abies pindrow* (Royle ex D. Don) Royle	Trees
464	580	Asteraceae	*Carpesium scapiforme* F.H.Chen & C.M.Hu	Herbs
465	581	Urticaceae	*Urtica ardens* Link	Herbs
466	582	Lamiaceae	*Scutellaria scandens* D.Don	Herbs
467	585	Zingiberaceae	*Roscoea purpurea* Sm.	Herbs
468	586	Oleaceae	*Jasminum dispermum* Wall.	Shrubs
469	587	Solanaceae	*Solanum viarum* Dunal	Herbs
470	588	Geraniaceae	*Geranium nepalense* Sweet	Herbs
471	589	Phrymaceae	*Phryma leptostachya* L.	Herbs
472	590	Urticaceae	*Pouzolzia sanguinea* (Blume) Merr.	Herbs

S.N.	Collection Number	Family	Scientific name	Habit
473	591	Apocynaceae	*Cynanchum auriculatum* Royle ex Wight	Herbs
474	592	Apocynaceae	*Cynanchum dalhousiae* Wight	Herbs
475	593	Rubiaceae	*Argostemma verticillatum* Wall.	Herbs
476	594	Fabaceae	*Desmodium multiflorum* DC.	Shrubs
477	595	Amaranthaceae	*Achyranthes bidentata* Blume	Herbs
478	596	Ranunculaceae	*Clematis montana* Buch.-Ham. ex DC.	Herbs, Climber
479	597	Cyperaceae	*Kyllinga brevifolia* Rottb.	Herbs
480	598	Cyperaceae	*Kyllinga brevifolia* Rottb.	Herbs
481	599	Araliaceae	*Hedera nepalensis* K.Koch	Climbers
482	600	Gesneriaceae	*Aeschynanthus parviflorus* (D. Don) Spreng.	Herbs
483	601	Fagaceae	*Quercus glauca* Thunb.	Trees
484	605	Commelinaceae	*Commelina maculata* Edgew.	Herbs
485	606	Gesneriaceae	*Corallodiscus lanuginosus* (Wall. ex DC.) B.L.Burtt	Herbs
486	607	Tiliaceae	*Grewia optiva* J.R.Drumm. ex Burret	Trees
487	608	Orchidaceae	*Vanda cristata* Wall. ex Lindl.	Herbs, Epiphytic
488	609	Fabaceae	*Erythrina stricta* Roxb.	Trees
489	611	Euphorbiaceae	*Excoecaria acerifolia* Didr.	Shrubs
490	612	Gentianaceae	*Tripterospermum volubile* (D. Don) H. Hara	Herbs
491	613	Asteraceae	*Ligularia fischeri* (Ledeb.) Turcz.	Herbs
492	614	Phyllanthaceae	*Phyllanthus emblica* L.	Trees, Medicinal
493	615	Lythraceae	*Punica granatum* L.	Trees, Cultivated
494	616	Myrtaceae	*Psidium guajava* L.	Trees, Cultivated
495	617	Anacardiaceae	*Rhus parviflora* Roxb.	Shrubs
496	618	Apocynaceae	*Calotropis gigantea* (L.) Dryand.	Shrubs, Medicinal
497	619	Verbenaceae	*Lantana camara* L.	Shrubs, Invasive
498	620	Iteaceae	*Itea nutans* Royle	Shrubs
499	621	Sapindaceae	*Sapindus mukorossi* Gaertn.	Trees
500	622	Caprifoliaceae	*Cornus oblonga* Wall.	Shrubs
501	623	Balsaminaceae	*Impatiens scabrida* DC.	Herbs
502	624	Fabaceae	*Mimosa rubicaulis* Lam.	Shrubs
503	625	Fabaceae	*Indigofera atropurpurea* Hornem.	Shrubs

Table 2: Details of the research team and their responsibilities

S.N.	Name	Designation	Institution	Remarks
1	Hai-Nin Qin	Professor	Institute of Botany, Chinese Academy of Sciences, Beijing, China	Team leader
2	Shu-Ren Zhang	Associate Professor	Institute of Botany, Chinese Academy of Sciences, Beijing, China	Member
3	Yu-Chang Yang	Graduate Student	Institute of Botany, Chinese Academy of Sciences, Beijing, China	Member
4	Prabin Bhandari	Researcher	RECAST, Tribhuvan University, Kirtipur, Kathmandu, Nepal	Member
5	Bijay Raj Subedee	Lecturer	RECAST, Tribhuvan University, Kirtipur, Kathmandu, Nepal	Member
6	Tirtha Raj Pandey	Research Officer	National Herbarium and Plant Laboratories, Godawari, Lalitpur, Nepal	Member & liaison officer

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A Checklist of Flowering Plants of Jaljalâ Mountain and adjoining Areas, Rolpâ District, Western Nepal

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Abstract

Jaljalâ mountain falling under the lesser Himalayas in the mid-mountain zone is surrounded by Thabâng rural municipality (previous VDCs; Thabâng, Mirul and Uwâ), Sunchhahâri rural municipality (Jelbang and Serum), and Rolpâ municipality (Dhabâng). The high topographic and seasonal variation allows this mountain with diverse flowering plant species. The present research work was undertaken with the aim of documenting the flowering plant species from the region. Plant specimens were collected along an elevation gradient of 2000-3580 m asl during three field visits during different seasons in 2014-2017. Herbarium specimens were prepared and identified by consulting relevant literatures and by comparing the specimens with specimens deposited in TUCH and KATH. In this study, a total of 560 species belonging to 329 genera and 98 families of flowering plants have been documented. Among them, 138 species were reported for the first time from western Nepal. Asteraceae (45 species) was the largest family followed by Rosaceae (41), Orchidaceae (38), Poaceae (36), Lamiaceae (24) and Ranunculaceae (24) in terms of species. Of the total species, 9 were endemic to Nepal, 86 were Himalayan endemic, 206 were Eastern Asiatic and 204 had broad distribution. Similarly, the area is highly influenced by Eastern Asiatic elements. Upon segregation by availability status, 226 species were abundant, 200 species were common, and 133 species were rare. Therefore, this study revealed that the area is rich in flowering plant diversity.

Keywords: Endemic, Flora, Lesser Himalayas, Mid-mountain

Introduction

Flora is one of the important components of biodiversity. It is crucial for the exploration of vegetation (Georgieva et al., 2013), as it helps to identify important elements of plant diversity, protect and preserve threatened plant species thereby to monitor and provide effective management for the particular vegetation type (Sahu & Dhal, 2012; Akinyemi & Oke, 2014).

In Nepal, floristic work was initiated since early nineteenth century from east Nepal when Buchanan-Hamilton visited Kathmandu valley in 1802 (Rajbhandari, 1976). However, it was started only by late nineteenth century in western Nepal after 1880 by Duthei from Doti and Baitadi districts during his collection along the Mahakali river along the border of Kumaon. Most of the major floristic works have been conducted in eastern and central Nepal and is still continuing along with publishing flora of Nepal. However, the western Nepal is relatively less explored in comparison to central and eastern Nepal (Rajbhandary et al., 1994). Among the very few works conducted in western Nepal, most have been either in Himalayan region or in lowlands; only a very few works have been carried out in the mid-mountain zone. The botanical explorations in western Nepal from 1880 onwards are: Duthei (1880-1884) collected plant specimens from Doti and Baitadi, few collections from Valley of Karnali River in the neighborhood of Simikot during 1927-1949 under the leadership of Major Lal Dhwoj, British expedition team (1952) under the leadership of three botanists Williams, Polunin and Sykes explored Karnali to Kali Gandaki of west Nepal in Jumla, Humla, Jajarkot and Salyan, Tyson

(1953) explored the vicinity of Api, Jest (1961) explored north west of Nepal in the region of Dolpo, Itoh and Rajbhandary (1963) collected specimens from Ghurchi and Khaptad (Rajbhandari, 1976) and the floristic expedition team collected plant specimens from different parts of west Nepal during flora of Nepal expedition 2012-2014 (Alliott, 2014). Regarding Rolpa, only few collections were made by national botanists; Manandhar, Bhattarai and Baral in 1982 and 1988 from the Sothern lower zone (record from herbarium specimens). Recently, a paper on medicinal plant species from the Northern highlands including Jaljalâ Mountain of the district have been published as the continuation of this project (Budha-Magar et al., 2020). Hence, this research work was conducted with the aim of documenting flowering plant species in Jaljalâ Mountain area which represents one of the mid-mountain physiographic zones in western Nepal.

Materials and Methods

Study area

The study area 'Jaljalâ Mountain' lies in Rolpa district, Mid-western Nepal. This is one of the proposed conservation areas by Government of Nepal (Basnet, 2010). The area is surrounded by Thabâng Rural municipality (previous VDCs; Thabâng, Mirul and Uwâ) and Sunchhahri Rural municipality (Jelbang and Serum), and Rolpâ Municipality (Dhabâng) (Central Bureau of Statistics [CBS], 2011). But the present study was mainly focused in three villages: Thabâng and Mirul of Thabâng Rural Municipality, and Jelbang of Sunchhahari Rural Municipality (Figure 1). Geographically, this mountain area falls under lesser Himalayas with high topographic and seasonal variation where whole district Rolpâ lies between 28.8° to 28.38°N latitudes and 82.10° to 83.90° E longitudes with an elevation range of 700-3600 m asl (above sea level) (District Forest Office [DFO], 2013). The climate of this region is influenced by monsoon. It is warm-temperate at lower elevations and alpine at higher elevations. The mean annual precipitation recorded was 1569.7 mm (range: 1161.2 mm in 2014 to 2497.1 mm in 2000). Most of the precipitation (1240.8 mm in average) occurs during the monsoon period (June to September). Similarly, the mean annual Maximum temperature (T_{max}) varies from 19.4 to 35.1°C and minimum temperature (T_{min}) varies from 5.3 to 22.9°C.

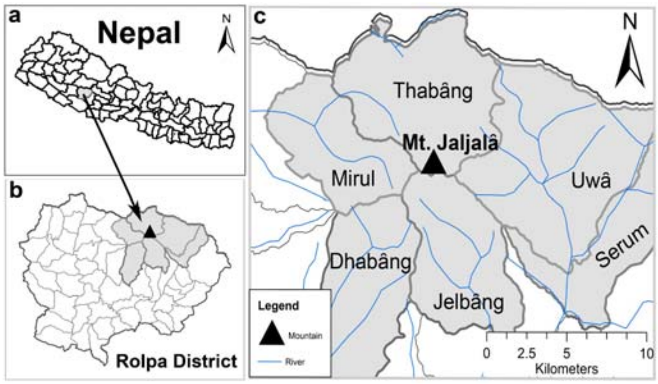

Figure 1: Map of study area showing a) Nepal, b) Rolpa district and c) Mt. Jaljalâ

Plant specimen collection, herbarium preparation and identification

Plant specimens were collected along an elevation gradient of 1600-3580 m asl during three field visits throughout different seasons (spring, summer, rainy and autumn) in 2014 - 2017 in three routes. The study was done along two transects from Thabâng village on the North-West and one transect to the East which was from Jelbâng village. At each 200 m asl elevation gain, a distance of about 50 m right or left from the trail was traversed and all the plants growing in the area were carefully inspected and representative vouchers were collected. All mountain ridges and valleys along five transects were explored covering all aspects from north to south and east to west on mountain range including the highest peak Dharampani at 3580 m asl to the south east and second highest peak Banchare (3400 m asl), Bhâmâtâkurâ, Kâlumtum, and Zerbe (Table 1) for plant specimen collection. Field note was recorded, and plant specimens were tagged. The collected specimens were pressed, dried, mounted and preserved based on standard methods as given by Forman and Bridson (1989). The plant specimens including the endemics were identified with the help of relevant literatures (Grierson & Long, 1983-2001; Polunin & Stainton, 1984; Stainton, 1987 and 1988;

Wu et al., 1994-2008; Noltie, 1994-2000; Press et al., 2000; Pearce & Cribb, 2002; Obha et al., 2008; Rajbhandari & Adhikari 2009; Raskoti 2009; Rajbhandari & Dhugana 2010, 2011; Watson et al., 2011). Some species were identified through expert consultation. All the species were scrutinized at least up to generic level. Finally, species were confirmed by comparing with herbarium specimens deposited at KATH (National Herbarium & Plant Laboratories), TUCH (Tribhuvan University Central Herbarium) and images of specimens deposited at TI (Herbarium of the University of Tokyo, Japan), K (Herbarium, Royal Botanic Garden, Kew, UK) and E (Herbarium, Royal Botanic Garden, Edinburgh, UK). The prepared specimens were deposited in TUCH and KATH.

Each plant species was scored for their local status as abundant, common and rare. The plant that were with less than 10 counts or citations in the study (covered by only our survey) were scored as rare or available in limited number. Those plant species with 10-50 counts/occurrence in the area at the time of the survey were scored as common and those with more than 50 counts were considered as abundantly available (modified from Wangchuk et al., 2013).

For the nomenclature, author citation and distribution of plant specimens, Catalogue of life, 2019 was followed. Species represented by asterisk (*) were identified based on photographs. The distribution range was based on relevant literatures and online floras (Press et al., 2000; Obha et al., 2008; Rajbhandari, et al., 2010,2011, 2012; Shrestha et al., 2017; Flora of Nepal Vol. 3; www.efloras.org). The families are arranged alphabetically in the checklist.

Results and Discussion

The flora of study area was observed to harbor a total of 560 species belonging to 329 genera and 98 families which was primarily based on present collection (Table 3). Regarding life form, the highest species were herbs with 395 species which was followed by shrub (84 spp.) and tree (58 spp.) (Figure 2.)

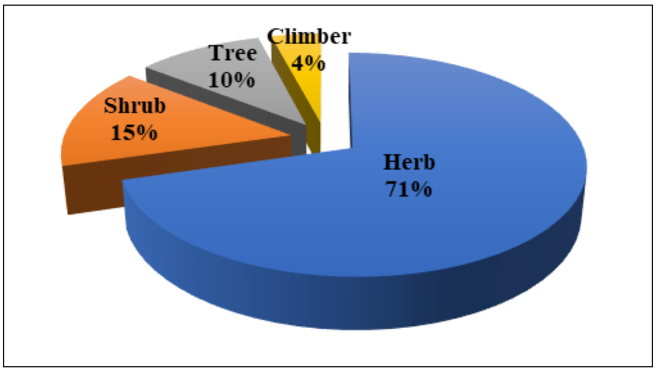

Figure 2: Life form of the plant species recorded in Jaljal Mountain area

Family composition

In the present research, the largest family recorded was Asteraceae (45 species) followed by Rosaceae (41), Orchidaceae (38), Poaceae (36), Lamiaceae and Ranunculaceae (24 species each), and Fabaceae (18) in terms of species. This is similar with previous research works (Hara et al., 1978-1982). The high dominance of family Asteraceae shows that the area is influenced by temperate elements (Pendry & Watson, 2009). In terms of genus also, Asteraceae was found to be highest family (31 genera), and then Poaceae was the second largest (30), followed by Orchidaceae (22), Rosaceae (17), Lamiaceae (16), Fabaceae (13), Ranunculaceae (10) and Urticaceae (9). *Carex* was the largest genus (8 species) followed by *Rubus*, *Impatiens*, *Hypericum* (7 each), *Swertia*, *Herminium*, *Potentilla* and *Clematis* (6 species each).

Out of 560 species, 9 species were reported to be endemic to Nepal, 86 species were Himalayan endemic, 206 species with Eastern Asiatic and 204 had broad distribution (Table 2 and Table 3). This result showed that the study area is highly influenced by Eastern Asiatic elements. Furthermore, 138 species were reported for the first time from West Nepal (Press et al., 2000; Rajbhandari, et al., 2010, 2011, 2012; Bhandari et al., 2016; Shrestha et al., 2017). This result shows that there is vast research gap in western Nepal specifically in mid-hills of West Nepal.

This study revealed 226 species were found abundantly which occurred in more than 50 places, 200 common species which were sited at between 10-50 places and 133 species were found rare which were sited in less than 10 places. Some species were found in only one place with very few (1-3) individuals. For example, *Notholirion macrophyllum* was found at only one place and a single individual was found. This availability status categorized as abundant, common and rare was based on the field observation during field work.

Conclusion

The flowering plants of Jaljalâ and adjoining areas were enumerated. Altogether 560 plant species that includes 200 common, 133 rare and 9 endemic species in Nepal were reported. The presence of high species richness in the area can be linked with the unique landscapes with diverse physiography and climate of Jaljalâ. The Government of Nepal has planned to protect this area as Conservation Area, which is very important to conserve the biological resources and the historical value of the region.

Note

Himalaya = Nepal, India, Bhutan, China (Tibet), and Pakistan

Pan-Himalaya = NE Afganistan, N. Pakistan, N. India, Nepal, Bhutan, N. Myanmar, and SW China (S. Xiang, SE Quasi, SE Qunghai, SE Gansu, W Sinchuan, NW Yunan)

SE Asia = South of China, east of India, West of New Guinea and north of Australia. It two regions Maritime Southeast Asia, comprising Indonesia, East Malaysia, Singapore, Philippines, East Timor, Brunei, and Christmas Island. Mainland Southeast Asia, also known as Indochina, comprising Cambodia, Laos, Myanmar (Burma), Thailand, Vietnam, and West Malaysia

Broad distribution = Beyond above three

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References

Akinyemi, D.S & Oke, S.O. (2014). Floristic composition and structural diversity of Shasha Forest Reserve in Ile-Ife, southwestern Nigeria. *Notulae Scintia Biologicae 6* (4), 433-440.

Basnet, K. (2010). *Feasibility study of the Jaljalâ Area*, Rolpa (for Conservation Area). Kathmandu, Nepal: National Trust for Nature Conservation.

Bhandari, P., Budha-Magar, S., & Shrestha, K. K. (2016). A checklist of flowering plants of Panchase Protected Forest, Kaski district, central Nepal. *Journal Natural History Museum*, *30*, 55–84.

Budha-Magar, S., Bhandari, P., & Ghimire, S. K. (2020). Ethno-medicinal survey of plants used by Magar (Kham) community, Rolpa district, Western Nepal. *Ethnobotany Research and Applications*, *19*. https://doi.org/10.32859/era.19.18.1-29

Catalogue of life. (2019). Indexing the known species of World. Published in www.catalogueoflife.org. (Accessed on 12 December)

Central Bureau of Statistics. (2011). Nepal Census 2011. District profiles (Demography). District

Profile of Rolpa. Kathmandu, Nepal: Author (Demography). Accessed from http://ddcrolpa.gov.np/wp-content/uploads/2016/05/6 शंक्षिप्त-परिचय.pdf on April 16, 2019.

Department of National Parks and Wildlife Conservation. (2019). *Biodiversity Profile of The Api Nampa Conservation Area, Nepal*. Kathmandu, Nepal: Author.

District Forest Office. (2013). *Annual report of Community Forest Development Program*. Rolpa: Author.

Eflora. (2019). *Annotated Checklist of Flowering Plants of Nepal*. http://www.efloras.org. (Accessed on 12 December)

Elliott, A. (2014). *Flora of Nepal Expedition-Baglung, Rukum and Dolpa*. Retrieved from: http://www.srgc.net/forum/index.php?topic=12304.0.

Forman, L. & Bridson, D. (1989). *The Herbarium Handbook*. Kew, UK: *Royal Botanic Gardens*.

Georgieva, N., Pachedjieva, K. & Lyubenova, M. (2013). Sci "Zapanda Stara Planina and Predbalkan"- Floristic studies on xerothermic oak forests. *Bulgarian Journal ofAgricultural Science 19*(2), 218-221.

Grierson, A.J.C. & Long, D.G. (1983-2000). *Flora of Bhutan*. (Vol. 1, Part 1-3; Vol. 2, Part 1-3). Edinburgh, UK: Royal Botanic Garden and Bhutan: Royal Government.

Hara, H & Williams, L.H.J. (1979). *An enumeration of the flowering plants of Nepal*, Vol. 2. London: British Museum (Natural History).

Hara, H., Chater, A.O. & Williams, L.H.J. (1982). *An enumeration of the flowering plants of Nepal* Vol. 3, UK: British Museum (Natural History).

Hara, H., Stearn, W.T. & Williams, L.H.J. (1978). *An enumeration of the flowering plants of Nepal*, Vol.1. London: British Museum (Natural History).

Harris, J.G. & Harris, M.W. (2001). *Plant identification terminology: An illustrated glossary*. (2nd ed). Spring Lake, Utah, USA: Spring Lake Publishing.

Hooker, J.D. (1872-1887). *Flora of British India*. Vol. I-VII. London: Reev and Company.

Noltie, H.J. (1994-2000). *Flora of Bhutan*. Vol. 3, Part 1-2, Edinburgh, UK: Royal Botanic Garden and Bhutan: Royal Government of Bhutan.

Obha, H., Iokawa, Y. & Sharma, L.R. (2008). *Flora of Mustang, Nepal*: Kodansha Scientific Ltd., Tokyo.

Pearce, N.R. & Cribb, P.J. (2002). *Flora of Bhutan*. Vol. 3, Part 3. Edinburgh, UK: Royal Botanic Garden.

Pendry, C. A. & Watson, M.F. (2009). Flora of Malesiana and the Flora of Nepal: floristic links and the potential for collection. *Blumea*, 54, 18-22.

Polunin, O. & Stainton, J.D.A. (1984). *Flowers of the Himalaya*. New Delhi, India: Oxford University Press.

Press, J.R., Shrestha, K.K. & Sutton, D.A. (2000). *Annotated Checklist of the Flowering Plants of Nepal*. London: The Natural History Museum.

Rajbhandari, K. R., & Adhikari, M. K. (2009). *Endemic flowering plants of Nepal. Part 1*. Kathmandu, Nepal: Department of Plant Resources.

Rajbhandari, K. R., & Dhungana, S. K. (2010). *Endemic flowering plants of Nepal. Part 2*. Kathmandu, Nepal: Department of Plant Resources.

Rajbhandari, K. R., & Dhungana, S. K. (2011). *Endemic flowering plants of Nepal. Part 3*. Kathmandu, Nepal: Department of Plant Resources.

Rajbhandari, K.R. & Baral S.R. (2010). *Catalogue of Nepalese Flowering Plants – I (Gymnosperms and Monocotyledons)*. Kathmandu, Nepal: Department of Plant Resources.

Rajbhandari, K.R. (1976). History of botanical explorations in Nepal. *Journal of the Bombay Natural History Society, 73*(1), 468-481.

Rajbhandari, K.R., Bhattarai K.R. & Baral S.R. (2011). *Catalogue of Nepalese Flowering Plants*

– II, Dicotyledons (Ranunculaceae to Dipsacaceae). Kathmandu, Nepal: Department of Plant Resources

Rajbhandari, K.R., Bhattarai K.R. & Baral S.R. (2012). *Catalogue of Nepalese Flowering Plants – III, Dicotyledons (Compositae to Salicaceae).* Kathmandu, Nepal: Department of Plant Resources.

Rajbhandary, T.K., Bista, M.S. & Gurung, V.L. (1994). *Enumeration of the Vascular Plants of West Nepal*. Kathmandu, Nepal: Department of Plant Resources.

Raskoti, B.B. (2009). *The Orchids of Nepal*. Kathmandu, Nepal: Quality Printers.

Sahu, S.C. & Dhal, N.K. (2012). Floristic composition, diversity and status of Threatened medicinal plants in tropical forests of Malyagiri Hill Ranges, Eastern Ghats, India. *Tropical forests*. Retrieved from http://www.intechopen.com/books/tropical-forests/floristic-composition-diversity-and-status-of-threatened-medicinal-plants-in-tropical-forests-of-mal.

Shrestha, K. K., Bhattarai, S., & Bhandari, P. (2018). *Handbook of Flowering Plants of Nepal (Vol. 1 Gymnosperms and Angiosperms: Cycadaceae-Betulaceae)*, Kathmandu, Nepal: Scientific Publishers.

Stainton, J.D.A. (1987). *Concise Flowers of Himalaya*. New Delhi, India: Oxford University Press.

Stainton, J.D.A. (1988). *Flowers of the Himalaya*: A supplement. New Delhi, India: Oxford University Press.

The Plant List (2019). Version 1.1. Published on the Internet; http://www.theplantlist.org/ (accessed on 12 December).

TROPICOS. (2019). Tropicos.org Missouri Botanical Garden. Published on http://www.tropicos.org (accessed on 12 December)

Wangchuk, P., Namgay, K., Gayleg, K., & Dorji, Y. (2016). Medicinal plants of Dagala region in Bhutan: their diversity, distribution, uses and economic potential. *Journal of ethnobiology and ethnomedicine*, *12*(1), 28.

Watson, M. F., Mkiyama, S, Ikeda, H, Pendry, C, Rajbhandari, K R, Shrestha, K. K. (eds) (2011). *Flora of Nepal*, Vol. 3. Edinburgh, UK: The Royal Botanic Garden.

Wu, Z., Raven, P.H. & Hong, D. (1994-2008). *Flora of China*. Vol 4, 8, 11, 13, 14, 16, 18, 19, 20, 22, 24, 25. Beijing, China: Science Press and St. Louis, USA: Missouri Botanical Garden Press.

Table 1: List of study sites in Jaljalâ Mountain and its adjoining areas

List of places	Latitude/longitude	Elevation (m)
Arnipo	28°27.261'/82°43.243'	2914
Bagtare	28°24.003'/82°44.012'	2300
Banchare	28°26.515'/82°44.199'	3370
Bhaisikharka	28°29.016'/82°45.001'	2740
Bhama	28°26.013'/82°43.007'	3175
Bhamatakura	28°26.004'/82°43.009'	3340
Bhamatakura	28°26.012'/82°43.007'	3190
Chalabang	28°28.859'/82°43.459'	2146
Daunne	28°27.992'/82°43.862'	2606
Dhangnai	28°26.515'/82°44.199'	3370
Dhangnai	28°26.007'/82°43.006'	3350
Dharampani	28°25.991'/82°42.150'	3580
Ghoreneta	28°29.012'/82°43.344'	2088
Ghosobang	28°29.016'/82°45.001'	2740
Jaljalâ	28°26.577'/82°43.500'	3145
Jelbang	28°24.015'/82°43.015'	2530
Kalenai khola	28°26.005'/82°44.000'	3166
Kalumtum Chheda	28°25.016'/82°44.012'	3230
Khanbe	28°26.015'/82°43.003'	3500
Kherbang	28°28.514'/82°44.423'	2357
Maalang rii	28°25.988'/82°42.370'	3444
Phuntibang	28°28.511'/82°44.502'	2351
Odarkhola, Phuntibang	28°28.739'/82°45.326'	2587
Rachibang	28°29.201'/82°43.233'	2150
Salap	28°27.011'/82°45.000'	2650
Zerbe, Jaljalâ	28°26.014'/82°42.011'	3410
Tijhibang	28°27.782'/82°43.388'	2494
Tila	28°27.261'/82°43.243'	2914

Table 2: List of endemic species in Jaljalâ mountain area

Family	Latin name (corrected)	Elevation (m)	Distribution Nepal	Distribution elevation
Asteraceae	*Cicerbita nepalensis* Kitam.	3400	WC Nepal	1600-3000m
Balsaminaceae	*Impatiens harae* H. Ohba & S. Akiyama	3444	E Nepal	2600-2700m
Begoniaceae	*Begonia leptoptera* H.Hara	2000	CE Nepal	1500-2600m
Fabaceae	*Crotalaria kanaii* H. Ohashi	2700	WC Nepal	2100-2900m
Lamiaceae	*Isodon dhankutanus* Murata	2200	WCE Nepal	1900-3800m
Lamiaceae	*Salvia transhimalaica* Yonek.; Fl. Mustang [Nepal]: 265 (2008)	2914	WC Nepal	2800m
Papaveraceae	*Meconopsis regia* G. Tayl.	3570	WCE Nepal	3500-4600m
Polygonaceae	*Fagopyrum megacarpum* Hara; J. Jap. Bot., 47(5): 137 (1972)	2500	WC Nepal	2400-3000m
Urticaceae	*Pilea kanaii* H. Hara	3250	WC Nepal	1500m

Table 3: Checklist of flowering plant in and around Jaljalâ Mountain, Rolpa, Western Nepal

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
Acanthaceae	***Strobilanthes foliosus* (Wight) T. Anders.		Herb	Tijhibhang	C	WCE Nepal	2400	*	
Acanthaceae	*Strobilanthes lachenensis* C.B. Clarke	*Angaari*	Herb	Tijhibhang	C	WE Nepal	2300	478	KATH/TUCH
Acanthaceae	†*Strobilanthes pentstemonoides* (Wall. ex Nees) T. Anders		Herb	Chalabang	C	WC Nepal	2300	1017J	KATH
Acanthaceae	***Strobilanthes thomsonii* T. Anderson	*Angaari*	Herb	Arnipo	C	WE Nepal Himalaya	2730	692	KATH/TUCH
Acanthaceae	*Strobilanthes tomentosa* (Nees) J.R.I.Wood		Herb	Rachibang	A	WCE Nepal	2300	873J	KATH
Acoraceae	*Acorus calamus* L.	*Baja*	Herb	Phuntibang	C	WCE Nepal	2500	*	
Adoxaceae	*Viburnum cotinifolium* D. Don	*Huirang*	Shrub	Arnipo	A	WC Nepal	2760	508	KATH/TUCH
Adoxaceae	*Viburnum cylindricum* Buch.-Ham. ex D. Don	*Mununmchorni*	Shrub	Gobang	A	WCE Nepal	2100	*	
Adoxaceae	***Viburnum erubescens* Wall.	*Hyanbur*	Shrub	Chalabang	A	WCE Nepal	2200	839	KATH/TUCH
Adoxaceae	*Viburnum mullaha* Buch.-Ham. ex D. Don	*Batapsai*	Shrub	Bhedakharka	A	WCE Nepal	2760	506	TUCH
Amaranthaceae	*Chenopodium album* L.	*Changya*	Herb	Gobang	C	WC Nepal	2100	*	
Amaranthaceae	***Cyathula capitata Wall. ex* Moq.	*Koora*	Herb	Phuntibang	C	WCE Nepal	2500	542	KATH/TUCH
Amaranthaceae	*Cyathula tomentosa* (Roth) Moq	*Koora*	Herb	Phuntibang	C	WCE Nepal	2500	552	KATH/TUCH
Amaryllidaceae	†*Allium tuberosum* Rottler ex Spreng.	*Haddijor*	Herb	Phuntibang	R	WC Nepal	2500	550	TUCH
Amaryllidaceae	*Allium wallichii* Kunth	*Balainow*	Herb	Jaljalâ, Banchare	C	WCE Nepal	3230-3370	654/993J	KATH/TUCH
Anacardiaceae	*Toxicodendron succedaneum (*L.) Kuntze	*Bhalayo*	Tree	Gobang	C	WCE Nepal	2100	*	
Apiaceae	*Bupleurum hamiltonii* N.P. Balakr.		Herb	Arnipo	A	WCE Nepal	2800	597	TUCH
Apiaceae	*Bupleurum longicaule* Wall. ex DC.		Herb	Jaljalâ	A	WCE Nepal	3240	648/914J	KATH/TUCH
Apiaceae	*Heracleum candicans* var. *obtusifolium* (Wall. ex DC.) F.T. Pu & M.F. Watson	*Tee*	Herb	Bhamatakura	C	WCE Nepal	2400-3370	307/897J/978 J	KATH/TUCH
Apiaceae	†*Heracleum nepalense D.Don*		Herb	Banchare	R	WCE Nepal	3370	994J	KATH
Apiaceae	*Hydrocotyle himalaica* P.K. Mukh.		Herb	Gongkhola	C	WCE Nepal	2200	719	KATH/TUCH
Apiaceae	†*Hymenidium apiolens* (C. B. Cl.) M. G. Pimenov & E. V Kljuykov		Herb	Dharampani	R	WCE Nepal	3448	938J	KATH
Apiaceae	*Ligusticopsis wallichiana (*DC.) Pimenov & Kljuykov	*Surkun*	Herb	Phuntibang	A	WCE Nepal	2500	553/985J/987 J	TUCH
Apiaceae	*Sanicula elata* Buch.-Ham. ex D.Don		Herb	Harpenaa	C	WCE Nepal	2900	606	TUCH
Apiaceae	*Selinum carvifolia* (L.) L.	*Dhanaura*	Herb	Arnipo	A	WC Nepal	2825	602/977J	TUCH
Apocyanaceae	***Ceropegia pubescens* Wall.		Herb	Tijhibhang	R	WCE Nepal	2400	493	KATH/TUCH
Apocyanaceae	†*Marsdenia lucida* Edgew. ex Madden	*Moor rala*	Climber	Tijhibhang	R	WCE Nepal	2550	575	KATH/TUCH
Apocyanaceae	*Periploca calophylla* (Wight) Falc.		Climber		R	WC Nepal		818	KATH/TUCH
Apocyanaceae	*Trachelospermum lucidum* (D. Don) K. Schum.		Climber	Chalabang	R	WCE Nepal	2100	454	KATH/TUCH
Apocyanaceae	*Vincetoxicum hirundinaria* Medik.		Herb	Phuntibang	R	WC Nepal	2500	*	
Aquifoliaceae	*Ilex dipyrena* Wall.	*Syaru*	Tree	Ghosobang	C	WCE Nepal	2740	758	KATH
Araceae	***Arisaema concinnum* Schott		Herb	Bhedakharka	C	WCE Nepal	2600	504	TUCH
Araceae	***Arisaema erubescens* (Wall.) Schott	*Bhinu*	Herb	Tijhibhang	A	WC Nepal	2400	487	TUCH
Araceae	**†*Arisaema griffithii* Schott	*Dhokaya*	Herb	Jaljalâ	A	WCE Nepal	3300	499	TUCH
Araceae	†*Arisaema jacquemontii* Blume		Herb	Tijhibhang	A	WCE Nepal	2400	*	
Araceae	*Arisaema tortuosum* (Wall.) Schott	*Basabhinu*	Herb	Tijhibhang	A	WCE Nepal	2460	503	TUCH
Araceae	*Sauromatum diversifolium* (Wall. ex Schott) Cusimano & Hett.	*Tinchhyo*	Herb	Bhamatakura	A	WCE Nepal	3190	424	KATH/TUCH
Araceae	***Sauromatum venosum* (Dryand. ex Aiton) Kunth		Herb	Gobang	R	WCE Nepal	2150	817	TUCH
Araliaceae	*Eleutherococcus cissifolius* (Griff. ex C.B. Clarke) Nakai		Shrub	Harpenaa	C	WCE Nepal	3000	439	TUCH
Araliaceae	*Hedera nepalensis* K. Koch		Climber	Tijhibhang	C	WCE Nepal	2450	639	KATH/TUCH
Araliaceae	***Panax pseudoginseng* Wall.	*Banaalu*	Herb	Arnipo	R	WCE Nepal	2790	595	TUCH
Aristolochiaceae	***Aristolochia griffithii* Hook. f. &		Climber	Bhedakharka	C	WCE Nepal	2600	477	KATH/TUCH

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
	Thomson ex Duch.								
Asparagaceae	*Agave americana* L.	*Siundi*	Shrub	Jurbang	C	WE Nepal	2100	*	
Asparagaceae	***Asparagus racemosus* Willd.	*Kuurila*	Shrub	Phuntibang	R	WCE Nepal	2500	*	
Asparagaceae	*Maianthemum purpureum* (Wall.) La Frankie	*Jyanbir*	Herb	Dhangnai	A	WCE Nepal	3400	361	KATH/TUCH
Asparagaceae	*Ophiopogon intermedius* D. Don		Herb	Above Jaljalâ pond	A	WCE Nepal	3200	391	KATH/TUCH
Asparagaceae	*Polygonatum cirrhifolium* (Wall.) Royle	Rukan	Herb	Phuntibang	A	WCE Nepal	2500	*	
Asparagaceae	†*Polygonatum singalilense* H.Hara	Rukan	Herb	On the way to Banchare	R	WCE Nepal	3205	1005J	KATH
Asparagaceae	*Polygonatum verticillatum* (L.) All.	Rukan	Herb	Dharampani	R	WCE Nepal	3444	934J	KATH
Asteraceae	***Ageratina adenophora* (Spreng.) R.M. King & H. Rob.		Herb	Thabâng	A	WCE Nepal	2100	*	
Asteraceae	*Ainsliaea latifolia* (D.Don) Sch.Bip.		Herb	Tijibhang	A	WCE Nepal	2530	785	KATH/TUCH
Asteraceae	*Anaphalis busua* (Buch.-Ham.) DC.	*Traawo*	Herb	Jaljalâ	A	WCE Nepal	3240	667/986J	KATH/TUCH
Asteraceae	†*Anaphalis cavei* Chatterjee		Herb	Bhama	R	WCE Nepal	3215	805J	KATH
Asteraceae	***Anaphalis griffithii* Hook. f.	*Trawo*	Herb	Jaljalâ	A	WCE Nepal	3210	631	KATH/TUCH
Asteraceae	*Anaphalis royleana* DC.	*Trawo*	Herb	Right to Jhankrithan	A	WCE Nepal	2500-3200	319/876J	KATH
Asteraceae	*Anaphalis triplinervis* (Sims) C.B.Clarke	*Trawo*	Herb	Phuntibang	A	WCE Nepal	2500	543	KATH/TUCH
Asteraceae	*Artemisia dubia* L. ex B.D.Jacks.	Paati	Herb	Phuntibang	A	WCE Nepal	2500	835	KATH/TUCH
Asteraceae	*Artemisia roxburghiana* Besser	*Gongpaati*	Herb	Right to Jhankrithan	C	WC Nepal	3200-3335	320/956J	KATH/TUCH
Asteraceae	†*Aster albescens* (DC.) Wall. ex Hand.-Mazz.		Shrub	Tila	R	WCE Nepal	2914	909J	KATH
Asteraceae	†*Aster asperulus* Wall. ex Nees	*Gaandowaasa*	Herb	Phuntibang	C	WC Nepal	2500	546	TUCH
Asteraceae	***Aster himalaicus* C.B. Clarke		Herb	Phuntibang	C	WCE Nepal	2500	743	KATH
Asteraceae	**†*Aster tricephalus* C.B. Clarke		Herb	Serbe, Jaljalâ	C	WCE Nepal	3360	663	KATH/TUCH
Asteraceae	Bidens pilosa L.	Kapche kuro	Herb	Gobang	C	WCE Nepal	2100	879J	TUCH
Asteraceae	***Blumea hieraciifolia* Hayata		Herb	Bagtare	A	WCE Nepal	2300	813	KATH/TUCH
Asteraceae	*Carpesium nepalense* Less.		Herb	Bhedakharka	A	WCE Nepal	2770	635	TUCH
Asteraceae	***Cicerbita nepalensis* Kitam.		Herb	Jaljalâ	C	WC Nepal	3400	355	KATH/TUCH
Asteraceae	***Cirsium falconeri* (Hook. f.) Petr.		Herb	Jaljalâ	C	WCE Nepal	3300	666	TUCH
Asteraceae	*Cirsium verutum* (D. Don) Spreng.	*Jhyankal*	Herb	Phuntibang	A	WCE Nepal	2500	810	TUCH
Asteraceae	**Cosmos bipinnatus Cav.	Kagate	Herb	Kherbang	A	C Nepal	2500	858J	KATH
Asteraceae	*Dubyaea hispida* (D. Don) DC.		Herb	Jaljalâ	C	WCE Nepal	3210	627	KATH/TUCH
Asteraceae	†*Duhaldea cappa* (Buch.-Ham. ex D. Don) Pruski & Anderberg		Shrub	Gobang	C	WCE Nepal	2100	885J	
Asteraceae	*Erigeron cuneifolius* DC.		Herb	Dharamshala	C	WCE Nepal	3180	614	KATH/TUCH
Asteraceae	*Erigeron multiradiatus* (Lindl. ex DC.) Benth.		Herb	On the way to Banchare	A	WCE Nepal	3198	1007J	KATH
Asteraceae	*Erigeron sumatrensis* Retz.		Herb	Rachibang	A	WCE Nepal	2300	854J	KATH
Asteraceae	*Galinsoga parviflora* Cav.	Raawande	Herb	Gobang	A	WCE Nepal	2100	*	
Asteraceae	*Himalaiella auriculata* (DC.) Raab-Straube	Nakali kuth	Herb	Bhama	R	WCE Nepal	3175	427	KATH/TUCH
Asteraceae	†*Jacobaea graciliflora* (DC.) Sennikov		Herb	Dharampani	R	WCE Nepal	3444	933J	KATH/TUCH
Asteraceae	*Leibnitzia nepalensis* (Kunze) Kitam.	Jhuula	Herb	Jaljalâ	C	WCE Nepal	3200	625	TUCH
Asteraceae	*Ligularia fischeri* (Ledeb.) Turcz.		Herb	Kalenai khola , Jaljalâ	C	WCE Nepal	3180	679	KATH/TUCH
Asteraceae	*Melanoseris violifolia* (Decne.)		Herb	Jaljalâ	C	WC Nepal	2100-3568	625/875J/982 J	TUCH
Asteraceae	*Myriactis nepalensis* Less.	*Lesse kuro*	Herb	Jaljalâ	C	WCE Nepal	3350	661	TUCH
Asteraceae	*Oreoseris maxima* (D. Don) X. D. Xu & W. Zheng	Ban Jhula	Herb	Rachibang	A	WCE Nepal	2300	859J	KATH
Asteraceae	*Prenanthes brunoniana* Wall. ex DC		Herb	Chalabang	C	WC Nepal	2200	*	
Asteraceae	*Pseudognaphalium affine* (D.Don) Anderb.	*Trawo*	Herb	Phuntibang	A	WCE Nepal	2500	521	KATH/TUCH

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
Asteraceae	*Senecio analogus* DC.	*Binjauri*	Herb	Jaljalâ	A	WCE Nepal	3200	480	KATH/TUCH
Asteraceae	*Senecio* sp.		Herb	Jaljalâ	A	Jaljal	3200	668	TUCH
Asteraceae	*Sigesbeckia orientalis* L.		Herb	Gobang	A	WCE Nepal	2100	869J	KATH
Asteraceae	*Sonchus wightianus* DC.	Dhudya	Herb	Kherbang	A	WCE Nepal	2500	868J	KATH
Asteraceae	***Synotis acuminata* (Wall. ex DC.) C. Jeffrey & Y.L. Chen		Herb	Arnipo	A	WCE Nepal	2770	592	TUCH
Asteraceae	*Synotis alata* C.Jeffrey & Y.L.Chen		Herb	Bhedakharka	A	WCE Nepal	2770	636	TUCH
Asteraceae	***Synotis cappa* (Buch.-Ham. ex D. Don) C. Jeffrey & Y.L. Chen		Shrub	Phuntibang	A	WCE Nepal	2500	548	TUCH
Asteraceae	*Taraxacum parvulum* DC.	*Dhaalmundra*	Herb	Jaljalâ	C	WCE Nepal	3190	426	KATH/TUCH
Asteraceae	*Tragopogon gracilis* D. Don	Sorno	Herb	Phuntibang	C	WCE Nepal	2500	*	
Asteraceae	*Xanthium strumarium* L.		Herb	Gobang	R	WCE Nepal	2100	867J	KATH
Balsaminaceae	†*Impatiens amplexicaulis* Edgew.		Herb	Jaljalâ	C	WCE Nepal	3145	919J	KATH
Balsaminaceae	***Impatiens arguta* Hook. f. & Thomson		Herb	Above Jaljalâ pond	C	WE Nepal	3200-3335	384/955J	TUCH
Balsaminaceae	†*Impatiens bicornuta* Wall.		Herb	Dharampani	R	WCE Nepal	3444	935J	KATH
Balsaminaceae	†*Impatiens falcifer* Hook. f.		Herb	Gongkhola	C	WCE Nepal	2200	562	TUCH
Balsaminaceae	***Impatiens harae* H. Ohba & S. Akiyama		Herb	Dharampani	C	E Nepal	3444	936J	KATH
Balsaminaceae	†*Impatiens scabrida* DC.		Herb	Tijhibhang	C	WC Nepal	2625	577	TUCH
Balsaminaceae	†*Impatiens urticifolia* Wall.	*Banbhango*	Herb	Phuntibang	C	WCE Nepal	2800	502	TUCH
Begoniaceae	†*Begonia dioica* Buch.-Ham. ex D.Don		Herb	Chalabang	R	WCE Nepal	2146	912J	KATH
Begoniaceae	***Begonia leptoptera* H.Hara		Herb	Chalabang	A	CE Nepal		892J	KATH
Begoniaceae	†*Begonia picta* Sm.		Herb		R	WCE Nepal	2000	530	KATH/TUCH
Berberidaceae	*Berberis aristata* DC.	*Katike chyantro*	Shrub	Chalabang	A	WC Nepal	2200	*	
Berberidaceae	Berberis asiatica Roxb. ex DC.	*Jethe chyantro*	Shrub	Chalabang	A	WCE Nepal	2240	1011J	KATH/TUCH
Berberidaceae	*Berberis napaulensis* (DC.) Spreng.	*Madalechyantro*	Shrub	Chalabang	C	WCE Nepal	2200		
Berberidaceae	†*Berberis thomsonia* C.K. Schneid.	*Gong chyantro*	Shrub	Near bojyuthan	A	WCE Nepal	3180	301	KATH/TUCH
Betulaceae	*Alnus nepalensis* D.Don	*Zhar*	Tree	Tijhibhang	A	WCE Nepal	2400	*	
Betulaceae	Betula alnoides Buch.-Ham. ex D.Don		Tree	Chalabang	C	WE Nepal	2072	1014J	KATH
Betulaceae	*Betula utilis* D. Don	*Bhuuja*	Tree	Tila	A	WCE Nepal	3100	*	
Betulaceae	***Corylus ferox* Wall.	*Ghyamonaa*	Tree	Arnipo	R	WCE Nepal	2800	*	
Boraginaceae	*Cynoglossum glochidiatum* Wall. ex Benth.	*Kuura*	Herb	Jaljalâ	C	WC Nepal	3350	325	KATH/TUCH
Boraginaceae	*Cynoglossum lanceolatum* Forssk	Lesse kuro	Herb	Jaljalâ	C	WCE Nepal	3200	*	
Boraginaceae	*Cynoglossum wallichii* G. Don	Lesse kuro	Herb	Banchare, Jaljalâ	C	WC Nepal	3300	687	KATH/TUCH
Boraginaceae	*Hackelia uncinata* (Benth.) C.E.C. Fisch.	*Lesse kuro*	Herb	Arnipo	A	WCE Nepal	3180	302	KATH/TUCH
Boraginaceae	†*Maharanga bicolor* (Wall. ex G. Don) A. DC.		Herb	Tutu (Mirul)	R	WC Nepal		890J	KATH
Boraginaceae	*Maharanga emodi* (Wall.) A. DC.	*Maharangi*	Herb	Bhama	R	WCE Nepal	3046-3398	415/959J/1001J	KATH/TUCH
Brassicaceae	*Arabidopsis himalaica (Edgew.) O. E. Schulz*		Herb	Dharampani	C	WCE Nepal	3570	335	KATH/TUCH
Brassicaceae	*Capsella bursa-pastoris* (L.) Medik.	*Chaangan*	Herb	Bhaisikharka	A	WCE Nepal	2740	768	KATH/TUCH
Buxaceae	*Sarcococca saligna* (D. Don) Müll. Arg.	*Ratawaicharo*	Shrub	Tijhibhang	C	W Nepal	2280	567	KATH/TUCH
Buxaceae	***Sarcococca wallichii* Stapf	Ratawaicharo	Shrub	Chalabang	C	WCE Nepal	2200	*	
Campanulaceae	**†*Campanula nakaoi* Kitam.		Herb	Dhangnai	C	CE Nepal	3335	957J	KATH
Campanulaceae	*Campanula pallida* Wall.		Herb	Phuntibang	A	WCE Nepal	2500	540/855J	KATH/TUCH
Campanulaceae	†*Codonopsis viridis* Wall.	Kaakolong	Climber	Chalabang	R	WCE Nepal	2200	*	
Campanulaceae	***Cyananthus inflatus* Hook. f. & Thomson		Herb	Dharamshala,Jaljalâ	A	WCE Nepal	3155	611	KATH/TUCH
Campanulaceae	*Lobelia heyneana* Schult.		Herb	Bhedakharka	R	WCE Nepal	2600	847	KATH/TUCH
Campanulaceae	*Lobelia pyramidalis* Wall.		Herb	Bhedakharka	C	WC Nepal	2730	644	KATH/TUCH
Cannabaceae	*Cannabis sativa* L.	*Bhaango*	Shrub	Phuntibang	A	WCE Nepal	2500	*	
Caprifoliaceae	*Dipsacus inermis* Wall.	*Taukejhar*	Herb	Phuntibang	C	WC Nepal	2500	537/960J	KATH/TUCH

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
Capprifoliaceae	*Leycesteria formosa* Wall.		Shrub	Harpenaa	C	WCE Nepal	2870	604	KATH/TUCH
Capprifoliaceae	†*Lonicera angustifolia* Wall. ex DC.		Shrub	Dhangnai	C	WCE Nepal	3350	322	KATH/TUCH
Caprifoliaceae	*Lonicera lanceolata* Wall.		Shrub	Khanbe	C	WCE Nepal	3400	*	
Caprifoliaceae	†*Morina longifolia* Wall. ex DC.	*Jhyangkaatu*	Herb	Jaljalâ	A	WCE Nepal	3260	656	KATH/TUCH
Caprifoliaceae	***Valeriana barbulata* Diels		Herb	Dharampani	A	E Nepal	3374	949J	KATH
Caprifoliaceae	*Valeriana hardwickii* Wall.	*Somaya*	Herb	Bhedakharka	A	WCE Nepal	2600	478	KATH/TUCH
Caprifoliaceae	*Valeriana jatamansii* Jones	*Somaya*	Herb	Tijhibhang	C	WCE Nepal	2600	497	KATH/TUCH
Caryophyllaceae	***Brachystemma calycinum* D. Don		Herb	Rachibang	R	CE Nepal	2300	860J	KATH
Caryophyllaceae	*Drymaria cordata* (L.) Willd. ex Schult.	*Nayajungle*	Herb	Jurbang	A	WCE Nepal	2100	*	
Caryophyllaceae	*Gypsophila cerastioides* D. Don		Herb	Khanbe	C	WCE Nepal	3500	326	KATH/TUCH
Caryophyllaceae	†*Silene caespitella* F. N. Williams		Herb	Dhangnai	R	WC Nepal	3398-3497 m	951J/958J	KATH
Caryophyllaceae	***Silene holosteifolia* Bocquet & Chater		Herb	Jaljalâ	R	WC Nepal	3200	630	KATH/TUCH
Caryophyllaceae	*Silene indica* Roxb. ex Otth	*Ombo*	Herb	Jurbang/Dharam pani	A	WCE Nepal	2100-3522	770/948J	TUCH
Caryophyllaceae	†*Silene stracheyi* Edgew.		Herb	Kalenai khola , Jaljalâ	C	WC Nepal	2610-3170	677/1011J	KATH/TUCH
Caryophyllaceae	†*Stellaria latifolia* Edgew. & Hook. f.	*Armaale*	Herb	Jurbang	A	W Nepal	2100	*	
Caryophyllaceae	*Stellaria monosperma* Buch.-Ham. ex D. Don	Mahajari	Herb	Dangza/Daune	R	WCE Nepal	2400-2900 m	895J/984J	KATH
Caryophyllaceae	***Stellaria patens* D. Don		Herb	Phuntibang	A	WC Nepal	2500	749	KATH/TUCH
Celastraceae	†*Euonymus echinatus* Wall.		Shrub	Tijhibhang	C	WCE Nepal	2400	489	KATH/TUCH
Celastraceae	***Euonymus frigidus* Wall.		Shrub	Ghosobang	C	WE Nepal	2740	766	TUCH
Celastraceae	*Euonymus lucidus* D.Don	*Gonammohar*	Tree	Tijhibhang	C	WCE Nepal	2400	485/1008J	KATH/TUCH
Celastraceae	***Parnassia trinervis* Drude		Herb	Pond	R	WC Nepal	3190	362	TUCH
Colchicaceae	*Disporum cantoniense (Lour.) Merr.*	*Rukan*	Herb	Dharampani	C	WCE Nepal	3570	515	TUCH
Commellinaceae	*Commelina paludosa* Blume		Herb	Gobang	A	WCE Nepal	2100	865J	50
Coriariaceae	†*Coriaria nepalensis* Wall.		Shrub	Chalabang	R	WCE Nepal	2240	1013J	51
Cornaceae	*Cornus capitata* Wall.	*Phuli*	Tree	Tijhibhang	C	WC Nepal	2470	510	KATH/TUCH
Crassulaceae	*Rhodiola bupleuroides* (Wall. ex Hook. f. & Thomson) S.H. Fu		Herb	Bhamatakura	C	WCE Nepal	3330	310	KATH/TUCH
Crassulaceae	*Rhodiola chrysanthemifolia* (H. Lév.) S.H. Fu		Herb	Above Jaljalâ pond	C	WC Nepal	3200	387	KATH/TUCH
Crassulaceae	***Rhodiola cretinii* (Raym.-Hamet) H. Ohba		Herb	Chalabang	C	WE Nepal	2100	463	TUCH
Crassulaceae	†*Rhodiola fastigiata* (Hook. f. & Thomson) S.H. Fu		Herb	Dharampani	C	WCE Nepal	3580	351	TUCH
Crassulaceae	*Rhodiola wallichiana* (Hook.) Fu		Herb	Dharampani	R	WCE Nepal	3568	941J	KATH
Crassulaceae	†*Sedum multicaule* Wall.		Herb	Ghoreneta	R	C Nepal	2088	903J	KATH
Cucurbitaceae	**†*Herpetospermum darjeelingense* (C. B. Clarke) H. Schaef. & S. S. Renner		Climber	Phuntibang	R	CE Nepal	2500	878J	KATH
Cucurbitaceae	†*Herpetospermum pedunculosum* (Ser.) C.B. Clarke		Climber	Bhama	R	WCE Nepal	3165	803	TUCH
Cucurbitaceae	*Solena heterophylla* Lour.	*Bidumba*	Climber	Chalabang	C	WCE Nepal	2100	450	KATH
Cyperaceae	*Carex anomoea* Hand.-Mazz.		Herb	Above Jaljalâ pond	C	WE Nepal	3200	321	KATH
Cyperaceae	*Carex atrata* subsp. *pullata* (Boott) Kük.		Herb	Above Jaljalâ pond	A	WCE Nepal	3200	400	TUCH
Cyperaceae	**†*Carex daltonii* Boot		Herb	Above Jaljalâ pond	A	WC Nepal	3200	400	TUCH
Cyperaceae	**†*Carex decora* Boott		Herb	Above Jaljalâ pond	A	WCE Nepal	3200	385	TUCH
Cyperaceae	***Carex myosurus* Nees		Herb	Above Jaljalâ pond	A	WCE Nepal	3200	401	TUCH
Cyperaceae	*Carex nubigena* D. Don ex Tilloch & Taylor		Herb	Above Jaljalâ pond	A	WCE Nepal	3220	407	KATH/TUCH
Cyperaceae	***Carex rochebrunii* Franch. & Sav.		Herb	Above Jaljalâ	A	WCE Nepal	3220	405	KATH/TUCH

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
				pond					
Cyperaceae	*Carex setigera* D. Don		Herb	Above Jaljalâ pond	A	WCE Nepal	3200	395	TUCH
Cyperaceae	*Cyperus cyperioides* (L.) Kuntze	*Tolagantha*	Herb	Gobang	C	WCE Nepal	2100	*	
Cyperaceae	*Eriophorum comosum* (Wall.) Nees	*Syanbhun*	Herb	Phuntibang	A	WCE Nepal	2500	723	KATH/TUCH
Dioscoreaceae	*Dioscorea bulbifera* L.	*Banseka*	Climber	Phuntibang	R	WCE Nepal	2500	523	KATH
Droseraceae	*Drosera peltata* Thunb.		Herb	Chalabang	A	WCE Nepal		893J	KATH
Elaeagnaceae	**†*Elaeagnus infundibularis* Momiy.	*Dhakari*	Tree	Tijhibhang	C	WCE Nepal	2400	483	TUCH
Elaeagnaceae	*Elaeagnus parvifolia* Wall. ex Royle	*Dhakari*	Tree	Chalabang	C	WCE Nepal	2250	776	KATH/TUCH
Ericaceae	*Gaultheria fragrantissima* Wall.	*Chotro*	Shrub	Gobang	A	WCE Nepal	2100	453	KATH/TUCH
Ericaceae	*Gaultheria nummularioides* D.Don	*Kaasai*	Herb	Arnipo	A	WCE Nepal	2720	691	KATH/TUCH
Ericaceae	*Lyonia ovalifolia* (Wall.) Drude	*Sirwan*	Tree	Bhedakharka	A	WCE Nepal	2740	507	KATH/TUCH
Ericaceae	*Lyonia villosa* (Wall. ex C.B. Clarke) Hand.-Mazz.	*Gongsirwan*	Shrub	Dhangnai	A	WCE Nepal	3400	368	KATH/TUCH
Ericaceae	***Monotropa uniflora* L.		Herb	Harpenaa	R	WCE Nepal	3000	433	KATH/TUCH
Ericaceae	***Rhododendron arboreum* Sm.	*Sarwai*	Tree	Bhedakharka	A	WCE Nepal	2730	509	TUCH
Ericaceae	*Rhododendron barbatum* Wall. ex G. Don	Kanwai	Tree	Jaljalâ	A	WCE Nepal	3300	*	
Ericaceae	*Rhododendron campanulatum* D. Don	*Chemala*	Shrub	Jaljalâ	A	WCE Nepal	3160	816	TUCH
Ericaceae	***Rhododendron setosum* D. Don	*Sunpati*	Shrub	Dharampani	R	WCE Nepal	3570	330/946J	TUCH
Euphorbiaceae	*Euphorbia thomsoniana* Boiss.		Herb	Dharampani	C	WCE Nepal	3575	336	KATH/TUCH
Euphorbiaceae	*Ricinus communis* L.	*Arenda*	Shrub	Gobang	C	WCE Nepal	2100	*	
Fabaceae	*Albizia julibrissin* Durazz.	*Baakar*	Tree	Chalabang	C	WCE Nepal	2100	462	KATH/TUCH
Fabaceae	†*Campylotropis speciosa* (Royle ex Schindl.) Schindl.	*Sankhina*	Shrub	Ghoreneta	C	WCE Nepal	2200	708	KATH/TUCH
Fabaceae	***Crotalaria kanaii* H. Ohashi		Herb	Bhedakharka	R	WC Nepal	2700	535	KATH/TUCH
Fabaceae	*Desmodium concinnum* DC.		Shrub	Kharaahaang	C	WCE Nepal	2000	842	TUCH
Fabaceae	*Desmodium elegans* DC.	*Tildala*	Shrub	Chalabang	C	WC Nepal	2200	709	KATH/TUCH
Fabaceae	Desmodium triflorum (L.)DC.		Herb		R	WCE Nepal	2100-2500	864J	KATH
Fabaceae	Erythrina arborescens Roxb.		Tree	Maranthana	R	WCE Nepal	2300	891J	KATH
Fabaceae	*Flemingia strobilifera* (L.) W.T. Aiton	*Bhuisankhina*	Herb	Gobang	C	WCE Nepal	2150	700	KATH/TUCH
Fabaceae	**†*Hedysarum campylocarpon* H. Ohashi	*Sankhina*	Shrub	Phuntibang	C	WC Nepal	2500	518	KATH/TUCH
Fabaceae	*Hedysarum sikkimense* Benth. ex Baker		Shrub	Bhama	C	WE Nepal	3215	420	KATH/TUCH
Fabaceae	*Indigofera bracteata* Graham ex Baker		Shrub	Bhama	C	WCE Nepal	3170	419	TUCH
Fabaceae	Indigofera dosua D.Don		Herb	Rachibang	R	WCE Nepal	2150	1000J	KATH
Fabaceae	*Lespedeza gerardiana* Graham		Shrub	Phuntibang	C	WC Nepal	2500	556	KATH/TUCH
Fabaceae	*Medicago denticulata* Willd.		Shrub	Dharampani	A	WCE Nepal	3520	328	KATH/TUCH
Fabaceae	*Parochetus communis* Buch.-Ham. ex D. Don		Herb	Jaljalâ	A	WCE Nepal	3340	306	TUCH
Fabaceae	*Piptanthus nepalensis* (Hook.) D. Don	*Jelawai*	Shrub	Bhama	A	WCE Nepal	3165	799	KATH/TUCH
Fabaceae	*Vicia angustifolia* L.	*Khosa*	Herb	Ghosobang	A	WCE Nepal	2740	767	KATH/TUCH
Fabaceae	*Vicia hirsuta* (L.) Gray	*Khosa*	Herb	Phuntibang	A	WCE Nepal	2500	746	KATH/TUCH
Fagaceae	†*Quercus floribunda* Lindl. ex A.Camus	Maikuti dalla	Tree	Phuntibang	C	WC Nepal	2500	883J	KATH
Fagaceae	*Quercus lanata* Sm.	*Mising*	Tree	Chalabang	C	WCE Nepal	2200	*	
Fagaceae	***Quercus oblongata* D.Don	Saipa	Tree	Kherbang	A		2500	884J	KATH
Fagaceae	***Quercus mespilifolioides* A. Camus	*Sari*	Tree	Tijhibhang	C	WE Nepal	2600	471	KATH/TUCH
Fagaceae	*Quercus semecarpifolia* Sm.	*Kar*	Tree	Tijhibhang	A	WCE Nepal	2400	*	
Gentianaceae	***Gentiana aristata* Maxim.		Herb	Kherbang	R		2357	1002J	KATH
Gentianaceae	*Gentiana capitata* Buch.-Ham. ex D.Don	*Wosyabada*	Herb	Phuntibang	A	WCE Nepal	2500	815	TUCH
Gentianaceae	***Gentiana micans* C.B. Clarke		Herb	Arnipo	C	WC Nepal	2770	593	KATH/TUCH
Gentianaceae	*Gentiana pedicellata* (D.Don) Wall.		Herb	Tijhibhang	C	WCE Nepal	2500	*	
Gentianaceae	**†*Gentiana robusta* King ex Hook. f.		Herb	Kalumtum Chheda, Jaljalâ	R	WC Nepal	3230	684	TUCH
Gentianaceae	*Halenia elliptica* D. Don	*Damphe*	Herb	Arnipo	A	WCE Nepal	2825	601	KATH/TUCH
Gentianaceae	*Swertia angustifolia* Buch.-Ham. ex D.	*Damphe*	Herb	Ghoreneta	A	WCE Nepal	2200	704	KATH/TUCH

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
	Don								
Gentianaceae	*Swertia bimaculata* (Siebold & Zuccarini) J. D. Hooker & Thomson ex C. B. Clarke	*Damphe*	Herb	Arnipo	A	WE Nepal	2800	598	KATH/TUCH
Gentianaceae	**†*Swertia chirayita* H. Karst.	*Runka*	Herb	Arnipo	R	WCE Nepal	2083-2825	600/901J	TUCH
Gentianaceae	*Swertia ciliata* (D. Don ex G. Don) B.L. Burtt	*Damphe*	Herb	Ghoreneta	C	WCE Nepal	2200	702	KATH/TUCH
Gentianaceae	***Swertia dilatata* C.B. Clarke	*Damphe*	Herb	above Jaljalâ pond	A	WC Nepal	3200	393	KATH/TUCH
Gentianaceae	*Swertia nervosa* (Wall. ex G. Don) C.B. Clarke	*Damphe*	Herb	Arnipo	A	WCE Nepal	2300-2765	588/871J	KATH/TUCH
Geraniaceae	†*Geranium lambertii* Sweet	*Chhapa jhar*	Herb	Dharampani	A	WC Nepal	3550	342	KATH/TUCH
Geraniaceae	*Geranium nepalense* Sweet		Herb	Phuntibang	A	WC Nepal	2500	545	KATH/TUCH
Geraniaceae	**†*Geranium procurrens* Yeo	*Chhapa jhar*	Herb	Phuntibang	A	WCE Nepal	2500	554	KATH/TUCH
Gesneriaceae	**†*Aeschynanthus sikkimensis* (C.B. Clarke) Stapf		Climber	Baagtare	C	WCE Nepal	1740	528	KATH/TUCH
Gesneriaceae	†*Platystemma violoides* Wall.		Herb	Phuliban	C	WCE Nepal	2000-2150	531/906J	KATH/TUCH
Grossulariaceae	*Ribes takare* D. Don	*Rijhyaunsai*	Shrub	Jerbe	R	WCE Nepal	3400	*	
Grossulariaceae	*Ribes alpestre* Wall. ex Decne.		Shrub	Ghosobang	C	WC Nepal	2740	764	TUCH
Grossulariaceae	*Ribes himalense* Royle ex Decne.		Shrub	Phuntibang	R	WCE Nepal	2500	*	
Grossulariaceae	***Ribes laciniatum* Hook. fil. & Thomson		Shrub	Dharampani	R	CE Nepal	3447	944J	KATH
Hydrangiaceae	*Deutzia compacta* Craib		Shrub	Chalabang	C	WCE Nepal	2100	460	KATH/TUCH
Hydrangeaceae	***Hydrangea aspera* Buch.-Ham. ex D. Don		Tree	Dangza	R	C Nepal		899J	KATH
Hypericaceae	*Hypericum choisyanum* Wall. ex N. Robso		Shrub	Bhama	A	WCE Nepal	3170-3320	419/915J	KATH
Hypericaceae	*Hypericum elodeoides* Choisy		Herb	Jaljalâ	A	WCE Nepal	3210	634	KATH/TUCH
Hypericaceae	*Hypericum himalaicum* N. Robson		Herb	Jaljalâ	A	WCE	3145	922J	KATH
Hypericaceae	*Hypericum japonicum* Thunb.		Herb	Nisel	C	WCE Nepal	2700	534	KATH/TUCH
Hypericaceae	***Hypericum monanthemum* Hook. f. & Thomson ex Dyer		Shrub	Bhama	R	WCE Nepal	3215	421	TUCH
Hypericaceae	**†*Hypericum tenuicaule* Hook. fil. & Thoms. ex Dyer		Shrub		R	E Nepal	2400	1000	KATH
Hypericaceae	*Hypericum uralum* Buch.-Ham. ex D.Don		Shrub	Chalabang	C	WCE Nepal	2200	707	TUCH
Iridaceae	***Iris decora* Wall.		Herb	Chalabang	C		2200	*	
Juglandaceae	*Juglans regia* L.	*Khasai*	Tree	Phuntibang	R	WCE Nepal	2500	757	TUCH
Juncaceae	*Juncus allioides* Franch.		Herb	Above Jaljalâ pond	C	WCE Nepal	3220	408	KATH/TUCH
Juncaceae	***Juncus benghalensis* Kunth		Herb	Dharampani	C	WCE Nepal	3550	340	KATH/TUCH
Juncaceae	*Juncus concinnus* D. Don		Herb	Serbe, Jaljalâ	A	WCE Nepal	3357	662	KATH/TUCH
Juncaceae	*Luzula multiflora* (Ehrh.) Lej.		Herb	Above Jaljalâ pond	C	WCE Nepal	3210	374	KATH/TUCH
Lamiaceae	*Ajuga bracteosa* Wall. ex Benth.		Herb	Bhedakharka	R	WCE Nepal	2650	759	KATH/TUCH
Lamiaceae	***Ajuga lobata* D. Don		Herb	Bhedakharka	R	WCE Nepal	2600	*	
Lamiaceae	*Craniotome furcata* (Link) Kuntze		Herb	Phuntibang	C	WCE Nepal	2500	547	KATH/TUCH
Lamiaceae	*Elsholtzia eriostachya* (Benth.) Benth.		Herb	Phuntibang	A	WCE Nepal	2500	555	KATH/TUCH
Lamiaceae	*Elsholtzia fruticosa* (D. Don) Rehder	*Thankra*	Shrub	Phuntibang	A	WCE Nepal	2500	551	KATH/TUCH
Lamiaceae	*Elsholtzia pilosa* (Benth.) Benth.		Herb	Jaljalâ	A	WCE Nepal	3320	659	KATH/TUCH
Lamiaceae	*Hyptis radicans* (Pohl) Harley & J.F.B.Pastore		Herb	Gobang	R	WCE Nepal	2100	853J	KATH
Lamiaceae	*Isodon coetsa* (Buch.-Ham. ex D.Don) Kudô		Herb	Phuntibang	A	WCE Nepal	2500	827	TUCH
Lamiaceae	*Isodon dhankutanus* Murata		Herb	Between Gobang and Rachibang	R	WCE Nepal	2200	862J	KATH
Lamiaceae	*Isodon repens* (Wall. ex Benth.) Murata		Herb	Tijhibhang	A	WCE Nepal	2550-3046	574/1003J	KATH/TUCH
Lamiaceae	*Lamium amplexicaule* L.	Dhakar	Herb		A	WCE Nepal	2700m	*	

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
Lamiaceae	*Leucosceptrum canum* Sm.	*Phusare*	Shrub	Bagtare	C	WCE Nepal	2300	806	TUCH
Lamiaceae	***Mesosphaerum suaveolens* (L.) Kuntze		Herb	Phuntibang	R	CE Nepal	2351	1003J	KATH
Lamiaceae	*Micromeria biflora* (Buch.-Ham. ex D.Don) Benth.		Herb	Chalabang	A	WC Nepal	2100	456	KATH/TUCH
Lamiaceae	*Nepeta lamiopsis* Benth. ex Hook. f.		Herb	Phuntibang	C	WCE Nepal	2500	591	KATH/TUCH
Lamiaceae	*Nepeta laevigata* (D.Don) Hand.-Mazz.		Herb	Dharampani	R	WCE Nepal	3568	983J	KATH
Lamiaceae	*Ocimum basilicum* L.		Herb	Chalabang	C	WCE Nepal	2200	*	
Lamiaceae	***Phlomis cashmeriana* Royle ex Benth.		Herb	Jaljalâ	R		3145	913J	KATH
Lamiaceae	†*Phlomis setigera* Falc. ex Benth.		Herb	Khanbe	C	WCE Nepal	3500	325	KATH/TUCH
Lamiaceae	***Phlomoides macrophylla* (Benth.) Kamelin & Makhm.		Herb	On the way to Banchare	A	CE Nepal	3198	1008J	KATH
Lamiaceae	*Prunella vulgaris* L.		Herb	Nisel	A	WCE Nepal	2700-2930	1010J	KATH
Lamiaceae	***Salvia campanulata* Wall. ex Benth.		Herb	Jaljalâ	R	WCE Nepal	3210	624	KATH/TUCH
Lamiaceae	***Salvia transhimalaica* Yonek.		Herb	Arnipo	A		2914	908J	KATH
Lamiaceae	†*Stachys melissaefolia* Benth.		Herb	Jaljalâ	R	WCE Nepal	3210	629	KATH/TUCH
Lardizabalaceae	***Holboellia latifolia* Wall.	*Banbaalu*	Climber	Chalabang	C	WCE Nepal	2200	711	KATH/TUCH
Lauraceae	*Actinodaphne sikkimensis* Meisn.		Tree	Jelbang	C	WE Nepal	2530	825	TUCH
Lauraceae	*Cinnamomum glanduliferum* (Wall.) Meisn.	*Malegiri*	Tree	Chalabang	R	WC Nepal	2200	715	KATH/TUCH
Lauraceae	***Lindera neesiana* (Wall. ex Nees) Kurz	*Tipa*	Tree	Chalabang	R	WCE Nepal	2310	697	KATH/TUCH
Lauraceae	***Lindera pulcherrima* (Nees) Hook. f.	*Phusare*	Tree	Jelbang	C	WC Nepal	2530	822	KATH/TUCH
Lauraceae	***Litsea doshia* (D. Don) Kosterm.		Tree	Nisel	C	WCE Nepal	2700	722	TUCH
Lauraceae	†*Machilus duthiei* King ex Hook.f.	*Syorga*	Tree	Chalabang	R	WCE Nepal	2310	781	KATH/TUCH
Lauraceae	*Phoebe lanceolata* (Nees) Nees	*Kaaru*	Tree	Chalabang	C	WCE Nepal	2200	710	TUCH
Liliaceae	*Lilium nepalense* D. Don	*Gaa*	Herb	Phuntibang	R	WCE Nepal	2100-2500	905J	KATH
Liliaceae	*Notholirion macrophyllum* (D. Don) Boiss.		Herb	Dhangnai	R	WCE Nepal	3300	369	KATH/TUCH
Linaceae	***Anisadenia saxatilis* Wall.		Herb	Dangza	R	CE Nepal	2300	894J	KATH
Loranthaceae	*Scurrula elata* (Edgew.) Danser	*Jokhare*	Shrub	Bhedakharka	C	WCE Nepal	2730	505	KATH/TUCH
Loranthaceae	*Scurrula parasitica* L.	*Jokhare*	Shrub	Ghoreneta	C	WCE Nepal	2200	713	TUCH
Lythraceae	*Rotala rotundifolia* (Buch.-Ham. ex Roxb.) Koehne		Herb		C	WCE Nepal	2200	*	
Magnoliaceae	***Magnolia campbellii* Hook.f. & Thomson		Tree	Phuntibang	R	WCE Nepal	2590	1004	KATH
Melanthiaceae	*Trillium govanianum* Wall. ex Royle	*Gong satUwâ*	Herb	Bhamatakura	R	WCE Nepal	3300	305	KATH/TUCH
Melanthiaceae	*Paris polyphylla* Sm.	*SatUwâ*	Herb	Phuntibang	R	WCE Nepal	2500	836	TUCH
Meliaceae	*Melia azedarach* L.	*Bakena*	Tree	Jhakibang	R	WE Nepal	2200	*	
Meliaceae	*Toona sinensis* (A. Juss.) M. Roem.	*Tooni*	Tree	Chalabang	R	WC Nepal	2100	459	KATH/TUCH
Menispermaceae	*Stephania glandulifera Miers*	*Badalpate*	Climber	Phuntibang	C	WCE Nepal	2500	519	TUCH
Moraceae	***Ficus neriifolia* Sm.	*Dudeula*	Tree	Bagtare	R	WC Nepal	2300	808	KATH/TUCH
Moraceae	*Ficus religiosa* L.	*Pipal*	Tree	Chalabang	R	WCE Nepal	2250	*	
Moraceae	*Ficus sarmentosa* Buch.-Ham. ex Sm.	*Bedula*	Climber	Chalabang	C	WCE Nepal	2100	458	KATH/TUCH
Moraceae	*Morus alba* L.	*Hoi/Toot*	Tree	Phuntibang	C	WCE Nepal	2500	558	KATH/TUCH
Moraceae	*Morus serrata* Roxb.	*Hoiapa*	Tree	Chalabang	C	WC Nepal	2200	*	
Myrsinaceae	*Eurya acuminata* DC.		Tree	Chalabang	C	WCE Nepal	2297	1010J	KATH
Nartheciaceae	***Aletris pauciflora* (Klotzsch) Hand.-Mazz.		Herb	Dharampani	C	WC Nepal	3570	333	KATH/TUCH
Oleaceae	*Jasminum humile* L.	*Phadulaa*	Shrub	Bhama	A	WC Nepal	3160	416	KATH/TUCH
Oleaceae	*Syringa emodi* Wall. ex G. Don		Tree	Tila	R	WC Nepal	3070	428	KATH/TUCH
Onagraceae	*Circaea alpina* L.		Herb	Jaljalâ	C	WCE Nepal	3240	652	KATH/TUCH
Onagraceae	***Epilobium palustre* L.; Sp. Pl.		Herb	Gobang	A	C Nepal	2100	866J	KATH
Onagraceae	*Epilobium sikkimense* Hausskn.		Herb	Jaljalâ	A	WCE Nepal	3145	918J	KATH
Orchidaceae	*Aphyllorchis* sp.		Herb	Daaune	A		2610	1015	KATH
Orchidaceae	*Bulbophyllum polyrrhizum* Lindl.		Herb	Phuliban	R	WC Nepal	1700	*	
Orchidaceae	***Calanthe plantaginea* Lindl.		Herb	Tijhibhang, Between Lamakhung and	R	WC Nepal	2450-2494	1005J	KATH

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
				Sano daunne					
Orchidaceae	***Calanthe puberula* Lindl.		Herb	Dangza	R	C Nepal	2300	896J	KATH
Orchidaceae	*Cephalanthera longifolia* (L.) Fritsch		Herb	Herpena	R	WC Nepal	3000	482	KATH/TUCH
Orchidaceae	†*Chiloschista viridiflava* Seidenf.		Herb	Baagtare	R	WCE Nepal	1740	819	KATH/TUCH
Orchidaceae	†*Cypripedium himalaicum* Rolfe ex Hemsl.		Herb	Arnipo	R	WCE Nepal	2850	498	KATH
Orchidaceae	*Dactylorhiza hatagirea* (D. Don) Soó	*Paanchaule*	Herb	Dharampani	R	WCE Nepal	3560	*	
Orchidaceae	***Dendrobium heterocarpum* Wall. ex Lindl.		Herb	Baagtare	R	WC Nepal	1740	820	KATH/TUCH
Orchidaceae	***Dendrobium longicornu* Lindl.		Herb	Tijhibhang	R	WCE Nepal	2400	*	
Orchidaceae	*Epipactis gigantea* Douglas ex Hook.		Herb	Harpenaa	R	WC Nepal	3000	443	KATH
Orchidaceae	*Epipactis helleborine* (L.) Crantz		Herb	Phuntibang/Tila	R	WC Nepal	2500-2910	881J/911J	KATH
Orchidaceae	*Goodyera biflora* (Lindl.) Hook. f.		Herb	Chalabang	R	WC Nepal	2200	514	KATH
Orchidaceae	***Goodyera foliosa* (Lindl.) Benth. ex C.B.Clarke		Herb	Between Tijhibang and Bhedakharka	R	E Nepal	2437	907J	KATH
Orchidaceae	*Goodyera repens* (L.) R. Br.		Herb	Tijhibang	R	WCE Nepal	2494	1009J	KATH
Orchidaceae	***Goodyera schlechtendaliana* Rchb. f.		Herb	Jaljalâ	C		3100	837	TUCH
Orchidaceae	*Habenaria intermedia* D.Don		Herb	Phuntibang	R	WC Nepal	2500	882J	KATH
Orchidaceae	***Herminium edgeworthii* (Hook.f. ex Collett) X.H.Jin, Schuit.		Herb	On the way to Dharampani	R		3350	931J	KATH
Orchidaceae	*Herminium josephi* Rchb. f.		Herb	Dhangnai	A	WC Nepal	3145	980J	KATH
Orchidaceae	*Herminium lanceum* (Thunb. ex Sw.) Vuijk		Herb	Jaljalâ	A	WCE Nepal	3145	926J	KATH
Orchidaceae	***Herminium latilabre* (Lindl.) X.H.Jin, Schuit., Raskoti & Lu Q.Huang		Herb	Ghoreneta	R		2072	900J	KATH
Orchidaceae	**†*Herminium macrophyllum* (D. Don) Dandy		Herb	Bhedakharka	C	WCE Nepal	2650-3145	694/924J	TUCH
Orchidaceae	*Herminium monorchis* (L.) R.Br.		Herb	Jaljalâ	A	WC Nepal	3145	928J	KATH
Orchidaceae	*Kingidium taenialis* (Lindl.) P.F. Hunt		Herb	Bagtare	R	WCE Nepal	1740	851	TUCH
Orchidaceae	*Malaxis monophyllos* (L.) Sw.		Herb	Arnipo	C	WCE Nepal	2755-3350	587/927J/998J/999J	KATH
Orchidaceae	*Malaxis muscifera* (Lindl.) Kuntze		Herb	Dhangnai	R	WCE Nepal	3335	954J	KATH
Orchidaceae	*Neottia listeroides* Lindl.		Herb	Daunne	A	WC Nepal	2606	1014J	KATH
Orchidaceae	*Peristylus affinis* (D. Don) Seidenf.		Herb	Jaljalâ	C	WCE Nepal	3130	674	KATH
Orchidaceae	†*Peristylus duthiei* (Hook.f.) Deva & H.B.Naithani		Herb	Jaljalâ	C	WCE Nepal	3145	925J	KATH
Orchidaceae	†*Peristylus fallax* Lindl.		Herb	Jaljalâ	A	WCE Nepal	3145	929J	KATH
Orchidaceae	*Peristylus lacertifer* (Lindl.) J.J. Sm.	*Sirkii*	Herb	Above Dharamshala	C	WC Nepal	3190	615	TUCH
Orchidaceae	*Pinalia graminifolia* (Lindl.) Kuntze		Herb	Chalabang	C	WE Nepal	2250	780	TUCH
Orchidaceae	***Platanthera stenantha* (Hook. f.) Soo		Herb	Dhangnai	R	C Nepal	3335	953J	KATH
Orchidaceae	***Pleione hookeriana* (Lindl.) B.S. Williams	*Ghabeto*	Herb	Banchare	R	WCE Nepal	3215-3400	411/988J	KATH
Orchidaceae	***Pleione humilis* (Sm.) D. Don	*Ghabeto*	Herb	Salap	R	WC Nepal	2650	742	KATH
Orchidaceae	*Ponerorchis chusua* (D. Don) Soo		Herb	On the way to Banchare	R	WCE Nepal	3205	1006J	KATH
Orchidaceae	*Satyrium nepalense* D.Don	*Sirkii*	Herb	Above Dharamshala	A	WCE Nepal	3180-3335	613/979J	KATH
Orchidaceae	*Spiranthes sinensis* (Pers.) Ames		Herb	Nisel	R	WCE Nepal	2100-2750	910J	KATH
Orobanchaceae	***Boschniakia himalaica* Hook. f. & Thomson		Herb	Bhamatakura	R	WCE Nepal	3310	311	KATH/TUCH
Orobanchaceae	*Lindenbergia muraria* (Roxburgh ex D. Don) Brühl		Herb	Jurbang	C	WCE Nepal	2100	705	TUCH
Orobanchaceae	†*Pedicularis bifida* (Buch.-Ham.ex D.Don) Pennell		Herb	Jaljalâ	C	WCE Nepal	2100-3170	532/902J	TUCH
Orobanchaceae	†*Pedicularis gracilis* Wall. ex Benth.		Herb	Jaljalâ	A	WCE Nepal	2300-3000	647/872J	KATH/TUCH

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
Orobanchaceae	†*Pedicularis hoffmeisteri* Klotzsch		Herb	Jaljalâ	A	WCE Nepal	3100	626	KATH/TUCH
Orobanchaceae	**†*Pedicularis megalantha* D. Don		Herb	Banchare	C	CE Nepal	2933	1002J	KATH
Orobanchaceae	†*Pedicularis roylei* Maxim.		Herb	Jaljalâ	C	WCE Nepal	3145	916J	KATH
Oxallidaceae	*Oxalis acetosella* L.		Herb	Tijhibhang	C	WE Nepal	2400	734	TUCH
Papaveraceae	*Corydalis casimiriana* Duthie & Prain ex Prain		Herb	Jaljalâ	A	WCE Nepal	3180	690/992J	KATH/TUCH
Papaveraceae	†*Corydalis chaerophylla* DC.		Herb	Kalenai khola , Jaljalâ	R	WCE Nepal	3170	678	KATH/TUCH
Papaveraceae	*Fumaria indica* Pugsley		Herb		A	WCE Nepal	2200	*	
Papaveraceae	*Meconopsis regia* G. Tayl.	*Bhokte*	Herb	Dharampani	A	WCE Nepal	3570	337/991J	TUCH
Parnasiaceae	*Parnassia nubicola* Wall. ex Royle		Herb	Jaljalâ	C	WCE Nepal	3204	622	KATH/TUCH
Phytolaccaceae	*Phytolacca latbenia* (Moq.) H. Walter	*Jargo*	Herb	Phuntibang	R	WC Nepal	2500	*	
Pinaceae	***Abies densa* Griff.	*Bhum*	Tree	Jaljalâ	A	WE Nepal	3200	*	
Pinaceae	*Abies spectabilis* (D. Don) Spach	*Bhum*	Tree	Jaljalâ	A	WC Nepal	2930-3200	1013J	KATH
Pinaceae	*Pinus roxburghii* Sarg.	*Daang*	Tree	Dahabang	A	WCE Nepal	2000	*	
Pinaceae	*Pinus wallichiana* A.B. Jacks.	*Dhupi*	Tree	Tijhibhang	A	WCE Nepal	2450	*	
Pinaceae	*Tsuga dumosa* (D. Don) Eichler	*Jham*	Tree	Tila	A	WCE Nepal	2930-3070	431/1012J	KATH/TUCH
Plantaginaceae	*Hemiphragma heterophyllum* Wall.	*Bokrosip*	Herb	Chalabang	A	WCE Nepal	2200	512	KATH/TUCH
Plantaginaceae	*Plantago asiatica* subsp. *erosa* (Wall.) Z.Yu Li	*Gando wasa*	Herb	Nisel	A	WCE Nepal	2800	501	KATH/TUCH
Plantaginaceae	*Veronica cana* Wall. ex Benth.		Herb	Tijhibhang	C	WCE Nepal	2600	468	KATH/TUCH
Plantaginaceae	*Wulfenia amherstiana* Benth.	*Deoraliwo*	Herb	Tijhibhang	A	W Nepal	2400	472	KATH/TUCH
Poaceae	***Agrostis gigantea* Roth		Herb	Jaljalâ	A	WC Nepal	3240	650	KATH/TUCH
Poaceae	***Agrostis micrantha Steud.*		Herb	Jaljalâ	C	WCE Nepal	3210	646	KATH/TUCH
Poaceae	*Agrostis pilosula* Trin.		Herb	Jaljalâ	C	WCE Nepal	3250	649	KATH/TUCH
Poaceae	*Aristida tsangpoensis* L. Liou		Herb	Jaljalâ	A	WC Nepal	3210	645	KATH/TUCH
Poaceae	*Arundinella setosa* Trin.		Herb		C	WCE Nepal	2500	863J	KATH
Poaceae	***Arthraxon lancifolius* (Trin.) Hochst.	*Chikapima*	Herb	Phuntibang	A	WCE Nepal	2500	718	KATH/TUCH
Poaceae	***Axonopus affinis* Chase		Herb	Tijhibhang	A	WCE Nepal	2400	717	TUCH
Poaceae	*Bothriochloa bladhii* (Retz.) S.T.Blake		Herb	Kustim	C	WE Nepal	2300	861J	KATH
Poaceae	*Briza media* L.		Herb	Banchare	A	WC Nepal	3372	1000J	KATH
Poaceae	***Bromus himalaicus* Stapf		Herb	Jaljalâ	C	WCE Nepal	3200	669	KATH/TUCH
Poaceae	*Calamagrostis pseudophragmites* (Haller f.) Koeler		Herb	Kalenai khola , Jaljalâ	R	WC Nepal	3200	681	KATH/TUCH
Poaceae	*Cenchrus flaccidus* (Griseb.) Morrone	*Kasabhu*	Herb	Phuntibang	A	WC Nepal	2700	724	KATH/TUCH
Poaceae	*Chrysopogon gryllus* (L.) Trin.	*Syopal*	Herb	Gobang	A	WCE Nepal	2150	728	KATH/TUCH
Poaceae	*Cymbopogon munroi* (C.B. Clarke) Noltie	*Pusai*	Herb	Gobang	C	WCE Nepal	2150	726	KATH/TUCH
Poaceae	*Cynodon dactylon* (L.) Pers.	Dubaa	Herb	Jurbang	A	WCE Nepal	2100	*	
Poaceae	*Dactylis glomerata* L.		Herb	Jaljalâ	C	WC Nepal	3230	655	KATH/TUCH
Poaceae	*Danthonia cumminsii* Hook. f.		Herb	Dhangnai	C	WCE Nepal	3400	359	KATH/TUCH
Poaceae	*Deschampsia caespitosa* (L.) P. Beauv.		Herb	Jaljalâ	A	WCE Nepal	3150	672	KATH
Poaceae	*Eulalia mollis* (Griseb.) Kuntze		Herb	Arnipo	A	WCE Nepal	2700	845	KATH/TUCH
Poaceae	*Eulaliopsis binata* (Retz.) C.E. Hubb.		Herb	Gobang	A	WCE Nepal	2150	*	
Poaceae	*Festuca leptopogon* Stapf		Herb	Above Jaljalâ pond	A	WC Nepal	3200	389	KATH/TUCH
Poaceae	*Festuca pamirica* Tzvelev	*Bukichhi*	Herb	Dharampani	A	WCE Nepal	3550	339	KATH/TUCH
Poaceae	***Helictotrichon junghuhnii* (Buse) Henrard		Herb	Jaljalâ	C	WCE Nepal	3200	834	KATH/TUCH
Poaceae	*Imperata cylindrica* (L.) P. Beauv.	*Siru*	Herb	Jhakibang	C	WC Nepal	2000	*	
Poaceae	*Koeleria cristata* var. *gracilis* DC.		Herb	Phuntibang	C	WC Nepal	2500	752	KATH/TUCH
Poaceae	*Koeleria litvinowii subsp. argentea* (Griseb.) S.M. Phillips & Z.L. Wu		Herb	Above Jaljalâ pond	C	WC Nepal	3220	406	KATH/TUCH
Poaceae	***Oryzopsis lateralis* (Munro ex Regel) Stapf ex Hook.f.		Herb	Jaljalâ	C	WC Nepal	3320	658	KATH/TUCH
Poaceae	***Paspalum thunbergii* Kunth ex Steud.		Herb	Chalabang	A	WCE Nepal	2200	*	

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Poaceae	***Poa annua* L.		Herb	Jaljalâ	A	WCE Nepal	3175	671	KATH/TUCH
Poaceae	***Poa pratensis* L.		Herb	Jaljalâ	A	WC Nepal	3175	670	KATH/TUCH
Poaceae	*Poa stapfiana* Bor		Herb	Near Dharamshala	A	WC Nepal	3170	382(DP)	KATH/TUCH
Poaceae	*Polypogon fugax* Nees ex Steud.		Herb	Gongkhola	A	WC Nepal	2400	771	KATH/TUCH
Poaceae	*Setaria viridis* (L.) P. Beauv.		Herb	Chalabang	A	WC Nepal	2200	*	
Poaceae	†*Tenaxia cumminsii* (Hook.f.) N.P.Barker & H.P.Linder		Herb	Jaljalâ	C	WCE Nepal	3046	921J	KATH
Poaceae	***Thamnocalamus spathiflorus* (Trin.) Munro	*Jsing*	Shrub	Dhangnai	C	WCE Nepal	3400	355	TUCH
Poaceae	***Themeda arundinacea* (Roxb.) A. Camus	*Parlap*	Herb	Gobang	A	WC Nepal	2150	727	TUCH
Polygalaceae	*Polygala sibirica* L.		Herb	Phuntibang	R	WC Nepal	2500 m	*	
Polygonaceae	*Bistorta amplexicaulis* (D. Don) Greene	*Paata apaa*	Herb	Bhama	A	WCE Nepal	3165-3440	417/932J	KATH/TUCH
Polygonaceae	*Bistorta macrophylla* (D. Don) Sojak	*Paatawo*	Herb	Dharampani	A	WCE Nepal	3145-3570	332/917J	KATH/TUCH
Polygonaceae	*Bistorta milletii* Leveille	*Paatawo*	Herb	Dharampani	A	WC Nepal	3570	331	KATH/TUCH
Polygonaceae	*Bistorta officinalis* Delarbre		Herb	Dharampani	A	WCE Nepal	3567	942J	KATH
Polygonaceae	*Fagopyrum dibotrys* (D. Don) H. Hara	*Banbhande*	Herb	Phuntibang	A	WCE Nepal	2500	538/870J	KATH/TUCH
Polygonaceae	*Fagopyrum megacarpum* Hara	Ban bhande	Herb	Kherbang	R	WC Nepal	2500	857J	KATH
Polygonaceae	***Koenigia campanulata* (Hook. fil.) T. M. Schust. & Reveal		Herb	Dharampani	A	CE Nepal	3498	943J	KATH
Polygonaceae	***Koenigia mollis* (D. Don) T. M. Schust. & Reveal	Thotane	Herb	Jaljalâ	A		3300	*	
Polygonaceae	*Persicaria chinensis* (L.) H. Gross	*Ratane*	Herb	Nisel	A	WCE Nepal	2600	496	KATH/TUCH
Polygonaceae	*Persicaria nepalensis* (Meisn.) Miyabe	*Ratane*	Herb	Harpenaa	A	WCE Nepal	3000	437	KATH/TUCH
Polygonaceae	***Persicaria runcinata (Buch.-Ham. ex D. Don) Masam.*		Herb	Jaljalâ	A	WCE Nepal	3170	382(DP)	TUCH
Polygonaceae	***Rheum australe* D. Don	*Chhikum/ Padame/Chulthe*	Herb	Bhamatakura	R	WCE Nepal	3350	308	KATH/TUCH
Polygonaceae	*Rumex acetosa* L.	*Theula*	Herb	Bhamatakura	A	WC Nepal	3190	422	KATH/TUCH
Polygonaceae	*Rumex hastatus* D. Don	*Kapu*	Herb	Jurbang	A	WC Nepal	2100	811	KATH/TUCH
Polygonaceae	*Rumex nepalensis* Spreng.	*Theula*	Herb	Near Dharamshala	A	WCE Nepal	3170	381(CP)	KATH/TUCH
Primulaceae	*Androsace geraniifolia* Watt		Herb	Banchare	R	WCE Nepal	3372	995J	KATH
Primulaceae	*Androsace sarmentosa* Wall.		Herb	Jaljalâ	A	WCE Nepal	3190	362	KATH/TUCH
Primulaceae	***Lysimachia congestiflora* Hemsl.		Herb	Bhedakharka	A	WCE Nepal	2700	643	KATH/TUCH
Primulaceae	*Myrsine africana* L.		Shrub	Chalabang	R	WC Nepal	2100	457	KATH/TUCH
Primulaceae	*Myrsine semiserrata* Wall.		Tree	Tijhibhang	A	WCE Nepal	2450	*	
Primulaceae	*Primula denticulata* Sm.	*Gandawo*	Herb	Phuntibang	A	WCE Nepal	2500	744	TUCH
Primulaceae	†*Primula edgeworthii* Pax	*Gandawo*	Herb	Bhama	C	WC Nepal	3165	793	KATH/TUCH
Primulaceae	*Primula glomerata* Pax	*Gandawo*	Herb	Right to Jhankrithan	C	WCE Nepal	3200	316	KATH
Primulaceae	*Primula primulina* (Spreng.) H. Hara		Herb	Nisel	C	WCE Nepal	2800	*	
Primulaceae	*Primula* sp.	*Gandawo*	Herb	Jhankrithan	A		3200	315	KATH
Ranunculaceae	†*Aconitum spicatum* (Brühl) Stapf	*Niing*	Herb	Jaljalâ	R	WCE Nepal	3330	670	KATH/TUCH
Ranunculaceae	*Actaea cimicifuga* L.		Herb	On the way to Banchare	R	WCE Nepal	3205	1004J	KATH
Ranunculaceae	***Anemonastrum demissum* (Hook. fil. & Thoms.) Holub	*Dhaskim*	Herb	Dharampani	A	WCE Nepal	3580	344	KATH/TUCH
Ranunculaceae	†*Anemonastrum polyanthes* (D. Don) Holub	*Rathabiratha*	Herb	Dharampani	A	WCE Nepal	3570	329	KATH/TUCH
Ranunculaceae	*Caltha scaposa* Hook. f. & Thomson		Herb	Jalja	A	WCE Nepal	3200	828	KATH/TUCH
Ranunculaceae	**†*Clematis acuminata* DC.	*Kaasangal*	Climber	Phuntibang	A	WC Nepal	2500	541	KATH/TUCH
Ranunculaceae	***Clematis buchananiana* DC.		Climber	Chalabang	C	WCE Nepal	2200	706	KATH/TUCH
Ranunculaceae	*Clematis connata* var. *confusa* (Grey-Wilson) W.T. Wang	*Kaasangal*	Climbing shrub	Phuntibang	R	WCE Nepal	2500	557	KATH
Ranunculaceae	*Clematis montana* Buch.-Ham. ex DC.		Climber	Dharampani	C	WCE Nepal	2500-3580	366/851J	KATH/TUCH

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Ranunculaceae	***Clematis terniflora* DC.	*Abijale*	Herb	Jurbang	R	WC Nepal	2150	829	TUCH
Ranunculaceae	*Clematis tortuosa* Wall. ex C.E.C. Fisch.		Climber	Bhamatakura	C	WCE Nepal	3190	641	KATH/TUCH
Ranunculaceae	***Delphinium altissimum* Wall.		Herb	Jsurbang	A	WC Nepal	2150	701	KATH/TUCH
Ranunculaceae	*Delphinium kamaonense* Huth		Herb	Dhabga	A	WC Nepal	2700	830	KATH/TUCH
Ranunculaceae	*Delphinium vestitum* Wall. ex Royle	*Mangramhul*	Herb	Dharamshala, Jaljalâ	A	WCE Nepal	3160	612	KATH/TUCH
Ranunculaceae	†*Eriocapitella rivularis* (Buch. Ham. ex DC.)	Phuntinbang	Herb		A	WCE Nepal	2600	*	
Ranunculaceae	*Eriocapitella rupicola* (Cambess.) Christenh. & Byng		Herb	Dharampani	C	WC Nepal	3520	327	KATH/TUCH
Ranunculaceae	*Eriocapitella vitifolia* (Buch.-Ham. ex DC.) Nakai	*Kaapaso*	Herb	Gongkhola	A	WCE Nepal	2200	560	KATH/TUCH
Ranunculaceae	*Oxygraphis polypetala* Royle ex D. Don		Herb	Near Dharamshala	A	WCE Nepal	3170	380	KATH/TUCH
Ranunculaceae	*Ranunculus brotherusii* Freyn		Herb	Jaljalâ	A	WCE Nepal	3200	477	KATH/TUCH
Ranunculaceae	*Thalictrum chelidonii* DC.	*Dhongare wo*	Herb	Bhedakharka	A	WCE Nepal	2660	579	KATH/TUCH
Ranunculaceae	*Thalictrum cultratum* Wall.	*Dhongare*	Shrub	Bhama	C	WCE Nepal	3190	419	KATH/TUCH
Ranunculaceae	†*Thalictrum punduanum* Wall.	*Dhongare*	Herb	Jaljalâ	A	WC Nepal	3250	633	TUCH
Ranunculaceae	***Thalictrum reniforme* Wall.		Herb	Bhedakharka	C	WCE Nepal	2650	*	
Ranunculaceae	†*Thalictrum rostellatum* Hook. f. & Thomson.		Herb	Dangza	R	WCE Nepal	2200	898J	KATH
Rosaceae	*Agrimonia pilosa* Ledeb.		Herb	Chalabang	A	WCE Nepal	2300	517	KATH/TUCH
Rosaceae	*Argentina festiva* (Soják) Soják	*Banmula*	Herb	Serbe, Jaljalâ	C	WCE Nepal	3410	664	KATH/TUCH
Rosaceae	*Argentina leuconota* (D. Don) Sojak		Herb	Dharampani	A	WCE Nepal	3448	939J	KATH
Rosaceae	*Argentina lineata* (Trevir.) Sojak	*Banmula*	Herb	Chalabang	A	WCE Nepal	2200-3335	904J	KATH
Rosaceae	*Argentina peduncularis* (D. Don) Sojak	*Banmula*	Herb	Dhangnai	C	WCE Nepal	3400	354	KATH
Rosaceae	*Argentina polyphylla* (Wall. ex Lehm.) Soják		Herb	On the way to Dharampani	A	WCE Nepal	3336	930J	KATH
Rosaceae	*Cotoneaster acuminatus* Wall. ex Lindl.		Shrub	Bhamatakura	A	WCE Nepal	3330	309	KATH/TUCH
Rosaceae	*Cotoneaster frigidus* Wall. ex Lindl.	*Kalimongjer*	Tree	Bhamatakura	A	WC Nepal	3285	312	TUCH
Rosaceae	*Cotoneaster microphyllus* Wall. ex Lindl.	*Saapithala*	Shrub	Phuntibang	A	WC Nepal	2500	525	KATH/TUCH
Rosaceae	*Dasiphora fruticosa (*L.) Rydb.		Shrub	Dharampani	R	WE Nepal	3570	334	KATH/TUCH
Rosaceae	*Eriobotrya elliptica* Lindl.	*Mahakhya*	Tree	Tijhibhang	C	WCE Nepal	2150-2450	1001J	KATH
Rosaceae	***Fragaria nilgerrensis* Schltdl. ex J. Gay	*Jhompasai*	Herb	Phuntibang	A	WCE Nepal	2500	745	TUCH
Rosaceae	*Fragaria nubicola* (Hook. f.) Lindl. ex Lacaita	*Jhompasai*	Herb	Near bojyuthan	A	WCE Nepal	3190	302	KATH/TUCH
Rosaceae	†*Geum elatum* Wall. ex G. Don		Herb	Khanbe	A	WCE Nepal	3350-3500	937J/940J	KATH
Rosaceae	**†*Potentilla caliginosa* Soják	*Banmula*	Herb	Dharampani	A	WC Nepal	3540	338	KATH/TUCH
Rosaceae	*Potentilla eriocarpa* Wall. ex Lehm.		Herb	Dharampani	R	WCE Nepal	3568	945J	KATH
Rosaceae	*Potentilla griffithii* Hook. f.	*Banmula*	Herb	Dharampani	A	WCE Nepal	3580	367	KATH/TUCH
Rosaceae	†*Potentilla monanthes* Wall. ex Lehm.		Herb	Phuntibang	C	WCE Nepal	2500	750	KATH/TUCH
Rosaceae	†*Potentilla nepalensis* Hook.		Herb	Phuntibang	A	WC Nepal	2500	525	KATH/TUCH
Rosaceae	*Potentilla sundaica* (Blume) Kuntze	*Banmula*	Herb	Phuntibang	C	WCE Nepal	2500	748	KATH/TUCH
Rosaceae	*Prinsepia utilis* Royle	*Kaikiram*	Shrub	Chalabang	A	WCE Nepal	2200	*	
Rosaceae	*Prunus cerasoides* Buch.-Ham. ex D. Don	Painya	Tree	Gobang	C	WCE Nepal	2150	*	
Rosaceae	*Prunus cornuta* (Wall. ex Royle) Steud.	*Gong rikureli*	Shrub	Jaljalâ	A	WE Nepal	3140	676	KATH/TUCH
Rosaceae	*Prunus napaulensis* (Ser.) Steud.		Tree	Tijhibhang	C	WCE Nepal	2400	*	
Rosaceae	*Pyracantha crenulata* (D. Don) M. Roem.	*Ghangaaru*	Shrub	Chalabang	A	WCE Nepal	2200	*	
Rosaceae	*Pyrus pashia* Buch.-Ham. ex D. Don	*Mihel*	Tree	Phuntibang	C	WCE Nepal	2240-2500	1012J	KATH
Rosaceae	*Rosa brunonii* Lindl.	*Dhankila*	Shrub	Chalabang	A	WCE Nepal	2500	524	KATH/TUCH
Rosaceae	*Rosa macrophylla* Lindl.	*Bhaarmasee*	Shrub	Bhama	A	WCE Nepal	3170	418	KATH/TUCH
Rosaceae	*Rosa sericea* Lindl.	*Melaaju*	Shrub	Above Jaljalâ pond	A	WCE Nepal	3220	403	KATH/TUCH
Rosaceae	***Rubus calycinus* Wall. ex D. Don		Shrub	Bhedakharka	R	WCE Nepal	2600	465	KATH/TUCH

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Rosaceae	*Rubus ellipticus* Sm.	*Angselu*	Shrub	Gobang	C	WCE Nepal	2150	*	
Rosaceae	*Rubus hoffmeisterianus* Kunth & Bouché	*Zoosai*	Shrub	Jaljalâ	A	W Nepal	3150	831	TUCH
Rosaceae	***Rubus mesogaeus* Focke		Shrub	Above Jaljalâ pond	C	WCE Nepal	3200	396	TUCH
Rosaceae	†*Rubus nepalensis* (Hook. f.) Kuntze	*Naample*	Herb	Chalabang	C	WCE Nepal	2200	*	
Rosaceae	*Rubus niveus* Thunb.	*Angselu*	Shrub	Chalabang	C	WCE Nepal	2250	777	KATH/TUCH
Rosaceae	*Rubus paniculatus* Sm.		Shrub	Chalabang	A	WCE Nepal	2250	*	
Rosaceae	*Sibbaldia cuneata* Hornem. ex Kuntze		Herb	Dharampani	A	WC Nepal	3500	*	
Rosaceae	†*Sorbus cuspidata* (Spach) Hedl.	*Poronaa*	Tree	Kalenai khola , Jaljalâ	C	WCE Nepal	3150	585	KATH
Rosaceae	*Sorbus foliolosa* (Wall.) Spach		Tree	Dhangnai	A	WCE Nepal	3350	321	KATH/TUCH
Rosaceae	*Spiraea bella* Sims		Shrub	Pond	A	WCE Nepal	3190	361	KATH/TUCH
Rosaceae	†*Spiraea micrantha* Hook. f.		Shrub	Chalabang	C	WCE Nepal	2100	455	KATH/TUCH
Rubiaceae	*Galium asperifolium* Wall.	*Khasare*	Herb	Phuntibang	A	WCE Nepal	2500-3350	516/989J/996 J	KATH/TUCH
Rubiaceae	*Galium asperuloides* Edgew.	*Khasare*	Herb	Bhamatakura	A	W Nepal	3220	303	KATH/TUCH
Rubiaceae	***Galium elegans* Wall. ex Roxb.	*Khasare*	Herb	Chalabang	C	WCE Nepal	2300	589	KATH/TUCH
Rubiaceae	*Galium spurium* subsp. *africanum* Verdc.	*Khasare*	Herb	Arnipo	C	WC Nepal	2770	590	KATH/TUCH
Rubiaceae	*Galium* sp.	*Khasare*	Herb	Jaljalâ	A		3100	304	KATH
Rubiaceae	†*Leptodermis lanceolata* Wall.		Shrub		A	WCE Nepal	2119	923J	KATH
Rubiaceae	*Leptodermis kumaonensis* R. Parker		Shrub	Tila	A	WCE Nepal	3070	429	TUCH
Rubiaceae	*Neohymenopogon parasiticus* (Wall.) Bennet	*Karkare*	Shrub	Chalabang	C	WCE Nepal	2310	698	TUCH
Rubiaceae	***Rubia manjith* Roxb. ex Fleming	*Khasare*	Herb	Phuntibang	C	WCE Nepal	2500	539	TUCH
Rutaceae	*Boenninghausenia albiflora* (Hook.) Rchb. ex Meisn.		Herb	Bhedakharka	C	WCE Nepal	2650	696	KATH/TUCH
Rutaceae	*Zanthoxylum armatum* DC.	*Tinbur*	Shrub	Gobang	R	WCE Nepal	2150	*	
Rutaceae	***Zanthoxylum* oxyphyllum Edgew.	*Jsya tinbur*	Shrub	Arnipo	R	WCE Nepal	2825	603	KATH/TUCH
Sabiaceae	*Sabia campanulata* Wall. ex Roxb.	*Kalirala*	Climber	Phuntibang	C	WCE Nepal	2500	491	KATH/TUCH
Salicaceae	*Salix babylonica* L.	*Bainsa*	Tree	Jurbang	C	WCE Nepal	2100	*	
Salicaceae	***Salix serpyllum* Andersson		Shrub	Tila	R	WC Nepal	3070	430	KATH/TUCH
Salicaceae	***Salix sikkimensis* Andersson	*Kyang*	Shrub	Above Jaljalâ pond	A	WE Nepal	3210	375	KATH/TUCH
Santalaceae	*Viscum album* L.	*Jokhare*	Shrub	Chalabang	R	WC Nepal	2250	779	KATH/TUCH
Sapindaceae	***Acer caudatum* Wall.	*Rijhaunsai*	Tree	Jaljalâ	C	WCE Nepal	3200	625	TUCH
Sapindaceae	*Acer sterculiaceum* Wall.	*Rijhaunsai*	Tree	Ghosobang	C	WCE Nepal	2740	765	KATH/TUCH
Sapindaceae	*Aesculus indica* (Wall. ex Cambess.) Hook.	*Paangar*	Tree	Tijhibhang	A	WC Nepal	2400	782	TUCH
Saxifragaceae	*Astilbe rivularis* Buch.-Ham. ex D. Don		Herb	Phuntibang	C	WCE Nepal	2100-3400	549/874J/952 J	TUCH
Saxifragaceae	†*Bergenia ciliata* (Haw.) Sternb.	*Haangawo*	Herb	Chalabang	C	WC Nepal	2250-3050	775/1016J	KATH/TUCH
Saxifragaceae	†*Saxifraga andersonii* Engl.		Herb	Below Banchare on the way to Jaljalâ from Daunne	R	WCE Nepal	3072	1006J	KATH
Saxifragaceae	*Saxifraga aristulata* Hook. f. & Thomson		Herb	Jaljalâ	C	WCE Nepal	3200	621	KATH/TUCH
Saxifragaceae	*Saxifraga parnassifolia* D. Don		Herb	Maghekharka	A	WCE Nepal	2500	533	KATH/TUCH
Saxifragaceae	*Saxifraga strigosa* Wall. ex Ser.		Herb	Tijhibhang	A	WCE Nepal	2380	570	KATH/TUCH
Schisandraceae	*Schisandra grandiflora* (Wall.) Hook.f. & Thomson	*Rhinghul*	Shrub	Jaljalâ	C	WCE Nepal	3100	628	TUCH
Scrophulariaceae	*Buddleja crispa* Benth.		Shrub	Jelbang	R	WC Nepal	2530	824	KATH/TUCH
Schrophulariaceae	*Scrophularia pauciflora* Benth.		Herb	Dharampani	C	WCE Nepal	3580	364	KATH/TUCH
Scrophulariaceae	*Verbascum thapsus* L.	*Yume*	Herb	Phuntibang	C	WCE Nepal	2500-3370	527/997J	TUCH
Scrophulariaceae	†*Veronica deltigera* Wall. ex Benth.		Herb	Jaljalâ	A	WCE Nepal	3145	920J	KATH
Simaroubaceae	*Brucea javanica* (L.) Merr.	*Bhakimbla*	Tree	Tijhibhang	R	WCE Nepal	2280	566	KATH/TUCH
Smilacaceae	*Smilax aspera* L.	*Dangruu*	Climber	Chalabang	C	WCE Nepal	2200	714	KATH/TUCH
Smilacaceae	*Smilax menispermoidea* A.DC.	*Dangruu*	Herb	Dhangnai	C	WCE Nepal	3400	353	TUCH

Family	Latin name	Local name	Life form	Locality	Local distribution	Distribution Nepal	Elevation (m)	Collection number	Herbaria
Smilacaceae	***Smilax ovalifolia* Roxb. ex D. Don.		Climber	On the way to Banchare	R	CE Nepal	3205	990J	KATH
Solanaceae	*Datura stramonium* L.		Herb	Phuntibang	R	WCE Nepal	2500	877J	KATH
Taxaceae	***Taxus contorta* Griff.	*Jhamchhetri*	Tree	Above Jaljalâ pond	R	WC Nepal	3200	394	TUCH
Theaceae	*Eurya acuminata* DC.	*Makya*	Tree	Gongkhola	C	WCE Nepal	2250	563	TUCH
Thymelaeaceae	***Daphne bholua* Buch.-Ham. ex D. Don	*Ratawaicharo*	Shrub	Tila	C	WCE Nepal	3070	788	KATH/TUCH
Thymelaeaceae	*Wikstroemia canescens* Wall. ex Meisn.		Herb	Harpenaa	R	WC Nepal	3000	610	TUCH
Urticaceae	***Boehmeria virgata* (G. Forst.) Guill. subsp. macrophylla (Hornem.) Friis & Wilmot-Dear		Herb	Tijhibhang	C	WCE Nepal	2400	*	
Urticaceae	*Boehmeria* sp.		Herb	Phuntibang	A		2500	852	TUCH
Urticaceae	*Debregeasia longifolia* (Burm. f.) Wedd.	*Tushaara*	Shrub	Jurbang	R	WCE Nepal	2100	*	
Urticaceae	*Elatostema monandrum* (Buch.-Ham. ex D.Don) H. Hara		Herb	Tijhibhang	C	WCE Nepal	2400	494	KATH
Urticaceae	*Elatostema obtusum* Wedd.		Herb	Above Jaljalâ pond	C	WCE Nepal	3200	399	KATH/TUCH
Urticaceae	*Girardinia diversifolia* (Link) Friis	*PUwâ*	Herb	Phuntibang	C	WCE Nepal	2500	880J	KATH
Urticaceae	*Gonostegia hirta* (Blume ex Hassk.) Miq.	*Barmitana*	Herb	Phuntibang	A	WCE Nepal	2500	*	
Urticaceae	*Lecanthus peduncularis* (Wall. ex Royle) Wedd		Herb	Jaljalâ	A	WCE Nepal	3170	*	
Urticaceae	*Oreocnide frutescens* (Thunb.) Miq.	*Sargil*	Herb	Jurbang	A	WCE Nepal	2100	*	
Urticaceae	***Pilea anisophylla* (Hook. f.) Wedd.		Herb	Bhedakharka	A	WCE Nepal	2600	470	KATH/TUCH
Urticaceae	***Pilea approximata* C.B. Clarke		Herb	Gongkhola	A	WE Nepal	2400	720	TUCH
Urticaceae	***Pilea kanaii* H. Hara		Herb	Jaljalâ	R	WC Nepal	3250	390	KATH/TUCH
Urticaceae	†*Pilea racemosa* (Royle) Tuyama		Herb	Dharampani	R	WCE Nepal	3522	947J	KATH
Urticaceae	*Pilea symmeria* Wedd.		Herb	Above Jaljalâ pond	C	WCE Nepal	3200	390	KATH
Urticaceae	*Urtica dioica* L.	*Nganti*	Herb	Phuntibang	A	WC Nepal	2500	*	
Urticaceae	***Urtica mairei* H. Lév.		Herb	Dhangnai	R	CE Nepal	3335	961J	KATH
Violaceae	*Viola biflora* L.		Herb	Near Dharamshala	A	WCE Nepal	3170	379	KATH/TUCH
Violaceae	*Viola betonicifolia* Sm.		Herb	Bhedakharka	A	WC Nepal	2700	792	KATH/TUCH
Violaceae	*Viola canescens* Wall.		Herb	Bhedakharka	A	WC Nepal	2600	478	KATH/TUCH
Violaceae	***Viola hookeriana* Kunth		Herb	Phuntibang	A	WCE Nepal	2500	755	KATH/TUCH
Violaceae	*Viola pilosa* Blume		Herb	Gongkhola	A	WCE Nepal	2330	783	KATH/TUCH
Vitaceae	†*Ampelocissus rugosa* (Wall.) Planch.	*Jiprang*	Climber	Chalabang	R	WCE Nepal	2250	773	KATH/TUCH
Vitaceae	*Parthenocissus semicordata* (Wall.) Planch.	*Jiprang*	Climber	Gobang	C	WC Nepal	2100	452	KATH/TUCH
Xanthorrhoeaceae	*Dianella ensifolia* (L.) DC.	`	Herb	Chalabang	R	WCE Nepal	2100	461	KATH/TUCH
Zingiberaceae	***Cautleya spicata* (Sm.) Baker	*Tunti*	Herb	Tijhibhang	C	WCE Nepal	2400	*	
Zingiberaceae	**†*Curcuma angustifolia* Roxb.	*Kachur*	Herb	Phuntibang	C	WC Nepal	2500	*	
Zingiberaceae	***Hedychium coronarium* J. Koenig	*Tunti*	Herb	Jhakibang	C	WC Nepal	2100	*	
Zingiberaceae	*Roscoea alpina* Royle		Herb	Above Jaljalâ pond	A	WCE Nepal	3220	404	KATH/TUCH
Zingiberaceae	*Roscoea purpurea* Sm.	*Sugu*	Herb	Dharampani	A	WCE Nepal	3550	341	TUCH

Note: A= Abundant, C= Common, R= Rare; † represents species endemic to Himalaya and ** represents species first recorded from West Nepal

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Floristic Diversity of Vascular Plants in Sikles Region of Annapurna Conservation Area, Nepal

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Abstract

Scientific investigation of floristic diversity is an essential prerequisite for conservation, management and sustainable utilization. The present study was conducted to explore the floristic diversity and life forms in Sikles region of Annapurna Conservation Area. Repeated field surveys with vegetation sampling and herbarium collection were done to find out floristic composition of the area. The study documented a total of 295 vascular plant species belonging to 238 genera and 107 families, including 25 species of fern and fern allies, 5 species of gymnosperms and 265 species of angiosperms. Herbs were dominant life form with 192 species followed by trees with 50 species whereas shrubs and climbers were 35 and 18 respectively. Asteraceae and Rosaceae (18 species each), Poaceae (17 species), Orchidaceae (16 species), Ranunculaceae (9 species) and Asparagaceae (8 species) were found to be dominant families in the region. *Impatiens* was the largest genera with 5 species followed by *Rubus* (4 species). *Begonia*, *Berberis*, *Swertia* had 3 species each. The life form classification shows the dominance of phanerophytes (29.27 %), therophytes (24.46 %) and chamaephytes (17.37 %) in the region. The rich flora of different taxonomic categories with both Eastern and Western Himalayan elements reflects the floristic importance of the region.

Keywords: Conservation, Eastern Himalaya, Flora, Life forms, Protected area

Introduction

Biodiversity is the heritage of millions of years of evolution. The enormous variety of life on earth is the result of complex interactions among all living organisms including microscopic species (Dirzo & Raven, 2003). Himalayan region, with long altitudinal gradients and climatic complexities, is considered as the biodiversity hotspot with rich vegetation, community and floral diversity (Sharma et al., 2014). The diversity of native flora is an important component of terrestrial ecosystems that has a primary role in protecting the environmental stability of a region (Lohbeck et al., 2016). Biodiversity is important for our survival as it provides us with various ecosystem services and goods (Chaudhary, 1998).

Human activities are continuously changing the world's terrestrial, freshwater and marine ecosystems and these changes are resulting in the loss of many species (Chapin et al., 2000), which calls for biodiversity conservation. The first and foremost step in this direction is to measure biodiversity occurring in various regions of the earth periodically. A measure of number of species present (species richness) at a given site, in a given area or country and, ultimately, in the whole world, is still the most straightforward and, in many ways, the most useful measure of biodiversity (Shaheen et al., 2012).

Floristic study refers to the documentation of all plants species in a given geographical region (Simpson, 2006). Floristic study is necessary to understand the present diversity status and conservation of forest biodiversity. It has been realized that the study of local or regional flora is of much more significance than those covering big areas because explorations can be carried out intensively in small areas. Understanding species diversity and distribution patterns is important to evaluate the complexity and resources of these forests. Floristic studies include species lists, life-form spectra, geographical distribution, and identification of threatened species that are useful for evaluating ecological issues such as biodiversity, growth capacity, conservation and regulation (Ali et al., 2018). Thus, floristic studies could provide valuable data which could be used as reference for future studies. The results of such floristic works mostly come in the form of floras (Palmer et al., 1995) which may be local, regional or national. According to Takhtajan (1986), Nepal lies in transitional zone between Eastern and Western Himalayan flora; therefore, due to the topographic and climatic variations high diversity of vascular plants can be seen in different parts of the countries.

Raunkiaer (1934) proposed the term "Biological Spectrum" to express both the life form distribution in a flora and the phytoclimate under which the prevailing life forms evolved. Life form study is thus an important part of vegetation description, ranking next to floristic composition. The basic life form categories include phanerophytes, chamaephytes, hemicryptophytes, cryptophytes and therophytes (Raunkiaer, 1934). Life forms depend on genetic as well as environmental factors because the environmental factors can affect the formation of different critical forms of plants (Shah et al., 2013). Accordingly, in different communities and different regions, plants can have different life forms. The biological spectrum is helpful in comparing geographically far and wide separated plant communities, and is used as an indicator of prevailing environment.

The aim of this study was to explore the floristic composition and plant diversity as well as to find out biological life form spectrum of plants in Sikles region, the southern part of Annapurna Conservation Area, which will help to outline further conservation measures to protect the diversity of the area.

Materials and Methods

Study site

The study was carried out in the Sikles area of Madi Rural Municipality located in the southern part of Annapurna Conservation Area, Kaski District, Gandaki Province. Diverse forest patterns are found in nearby Parche and Sikles villages providing communities with basic services. The forest floor is uneven and elevation ranges from 2000 to 3300 m asl. Geographically, it is located within the coordinate range of 28°28'N-28°47'N latitude and 84°00'E-84°42'E longitude (Figure 1).

The climate of the study area ranges from upper subtropical to lower alpine mostly covering temperate region. The climate is influenced by monsoon with temperate climate in lower elevation while subalpine in higher elevation. Records of Department of Hydrology and Meteorology for the last 8 years (2010-2017) shows maximum annual temperature of 21.9°C and minimum of 12.2°C. The average annual precipitation is 575 mm with maximum mean precipitation of 1020.25 mm in July (Department of Hydrology & Metrology [DHM], 2018).

According to the altitudinal zone and climatic variations *Alnus* forests, mixed forests, broad leaved forests, evergreen forests and bushes and grasslands are seen in the area. Lower area of dense canopy forest and higher alpine meadow area creates suitable habitats for different ungulate species of animals.

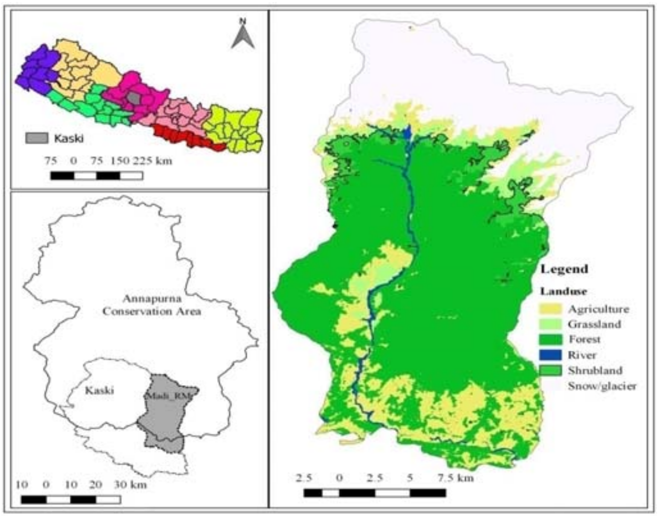

Figure 1: Land use classification map of the study area

Research approval

Prior to undertaking the research, the objectives were discussed with the management authorities of Annapurna Conservation Area. The permission for field work was taken from the Department of National Parks and Wildlife Conservation

(DNPWC) and Annapurna Conservation Area Project (ACAP).

Field survey

The field study was carried out from June 2018 to January 2019 within the elevation range of 2000-4000 m covering different habitat types and vegetation zones. Data on floristic composition of the area were collected by stratified random sampling (Behera et al., 2005; Panthi et al., 2007). The study area within altitudinal range of 2000-4000 m was divided into six sampling sites characterizing different altitudes, forest types and habitats. The sites were Raising Danda, Mouja-Prolu, Thasa Kharka, Chyomi, Tinje and Kori. Altogether 60 plots (10 m × 10 m) were studied in six sampling sites (10 in each). Sampling plots within the sites were selected using reference site such as walking trail. The distance between two plots in each sampling sites was approximately 100 m.

Raising Danda site was located within the altitudinal range of 2000-2300 m. The vegetation was mostly dominated by *Alnus nepalensis* and the community composed of *Daphniphyllum himalayense*, *Viburnum erubescens* and *Brucea javanica*. The altitudinal range of Mouja-Prolu was 2300 2600 m. The mixed forest type was seen in this range with the species of *Symplocos ramosissima*, *Eurya acuminate*, *Lyonia ovalifolia* and *Elaeagnus parvifolia*. Tasha Kharka area was located in the altitudinal range of 2600-2800 m. Species such as *Ilex dipyrena*, *Hydrangea heteromalla* and *Acer cappadocicum* were found with *Rhododendron arboreum*. Chyomi area, lying within the altitudinal range of 2800-3200 m, constituted mainly of Oak-*Rhododendron* forest. The dominant tree species in this site were *Rhododendron arboreum* and *Quercus semecarpifolia*. Tinje area, located within 3200-3600 m altitudinal range, was mostly covered by shrubby vegetation of *Rosa sericea* and *Berberis concinna* with *Rhododendron campanulatum* Kori area, with altitudes from 3700 m to 4000 m, is mostly presenting grasses with scattered *Rhododendron campanulatum*. This area is very sloppy with large rocks cliffs.

Sample collection and identification

Voucher specimens of all vascular plants, either in flowering or fruiting stage, were collected to prepare herbarium specimens (Rajbhandari & Rajbhandary, 2015). All vouchers were taken to the laboratory for identification with the help of detailed field data collected during the field trips. The herbarium specimens were deposited in Tribhuvan University Central Herbarium (TUCH).

Identifications of voucher specimens were carried out by following standard literatures (Grierson & Long, 1983-2001; Polunin & Stainton, 1984; Stainton, 1988; Wu & Raven, 1994; Press et al., 2000; Fraser-Jenkins, 2015; Rajbhandari & Rai, 2017), expert consultation and visit to the National Herbarium and Plant Laboratories (KATH) and Tribhuvan University Central Herbarium (TUCH). Nomenclature follows the Catalogue of Life (Roskov et al., 2019) and Plants of the World Online (POWO, 2019). The plants were classified into different life form following Raunkiaer (1934) (Table 5).

Results and Discussion

Floristic composition

The floristic composition of Sikles region comprised of 295 species belonging to 107 families and 238 genera (Tables 1, 2, 3 and 4). In terms of species, 206 were dicots, 59 monocots, 25 ferns and 5 Gymnosperms (Figure 2). Asteraceae and Rosaceae were dominant families with 18 species each followed by Poaceae (17 species), Orchidaceae (16 species), Ranunculaceae (9 species) and Asparagaceae (8 species). Polygonaceae, Fabaceae, and Lamiaceae (7 species each) were among other families with most species (Figure 3). The present findings are similar to those of Chalise et al. (2019) in Gyasumbdo valley Manang, a territory within Annapurna Conservation Area, where dicots were dominant with Asteraceae as dominant family. However, results differ in monocots, where Orchidaceae was dominant family in Manang, whereas Poaceae was dominant family in the present study. Major parts of the present study area were located in higher altitude mostly covered by the grasses and with scattered *Rhododendron campanulatum*.

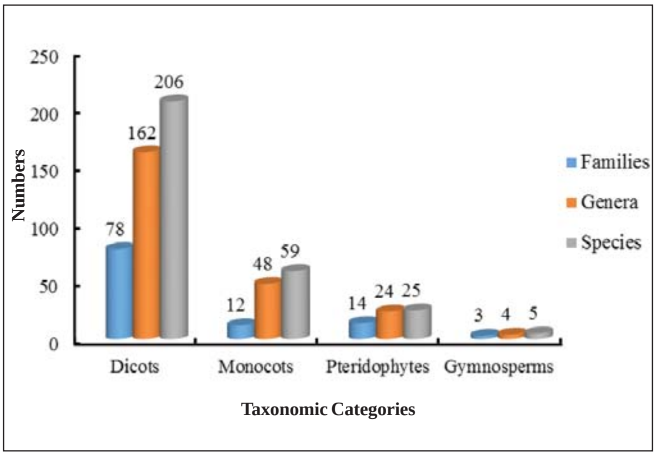

Figure 2: Number of families, genera and species of plant groups

Among 14 families of Pteridophytes, Pteridaceae (3 genera, 5 species) was found to be the largest family, while in case of Gymnosperms, Pinaceae (2 genera, 2 species) was found to be the largest family. Similar results were also reported by Chalise et al. (2019).

Floristic study revealed that dicots (206 species) were the most diverse group followed by monocots (59 species) in terms of species composition. *Impatiens* was the largest genus with 5 species followed by 4 species of *Rubus*. *Begonia*, *Berberis* and *Swertia* had 3 species each. Based on plant habits, 192 (65%) species were herbs, 50 (17%) species were trees and 35 (12%) species were shrubs while climbers included 18 (6%) species (Figure 4).

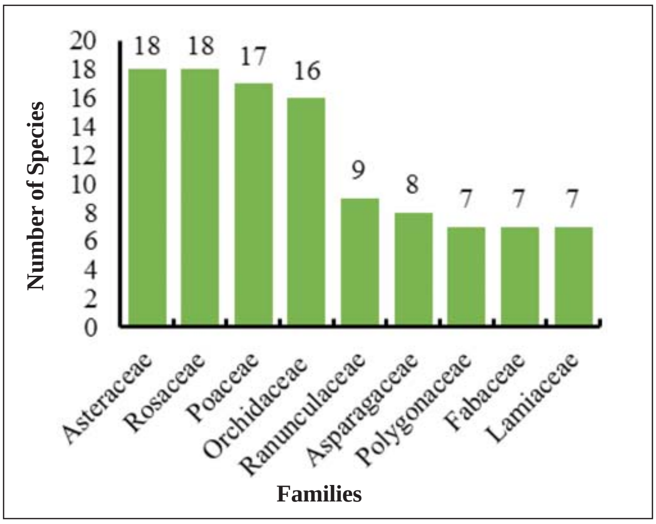

Figure 3: Dominant families in study area

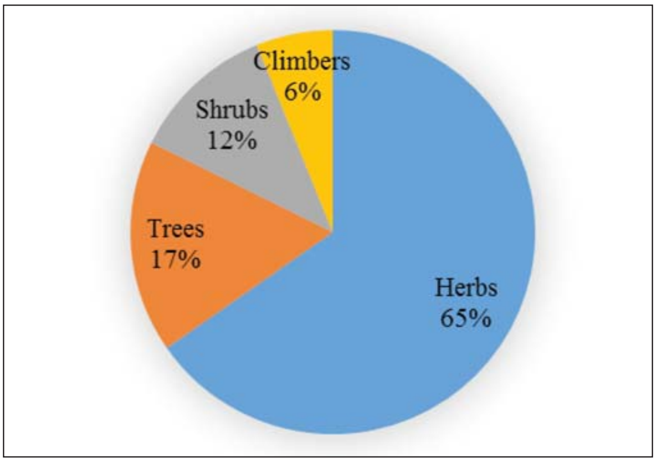

Figure 4: Habits of plants

The study area lies near to Kali Gandaki River which separates the Eastern and Western Himalayan floristic regions. The summer rainfall is high in this area. Therefore, the area is rich with the assemblage of both Eastern and Western floristic elements with some other unique species such as *Alsophila spinulosa*, *Dolomiaea macrocephala*, *Meconopsis regia* and *Hymenidium benthamii*. In the context of the floral diversity, the vegetation is mostly dominated by the Eastern Himalayan elements such as species of *Aconitum*, *Berberis*, *Calanthe*, *Cicerbita*, *Corydalis*, *Potentilla*, *Rubus*, *Saxifraga*, *Delphinium*, and *Impatiens* with other some Western Himalayan elements such as species of *Abies*, *Quercus* (Takhtajan, 1986; Welk, 2015; Chalise et al., 2019). The differing flora of the East Himalaya and the West Himalaya merge in Central Nepal.

Some potentially high value medicinal plants such as *Aconitum gammiei*, *Bergenia ciliata*, *Dactylorhiza hatagirea*, *Paris polyphylla*, *Picrorhiza scrophulariiflora*, *Rheum australe* and *Swertia chirayta* were also recorded. Among the documented species, 17 species had been included in one of the categories of Conservation Assessment and Management Plan (CAMP) (Bhattarai et al., 2002). Two species (*Taxus wallichiana* and *Nardostachys jatamansi* had been included in International Union for Nature Conservation (IUCN) red list categories (IUCN, 2019). Five species had been included in the *Convention on International Trade in Endangered Species* of Wild Fauna and Flora (CITES) list (UNEP-WPMC, 2018). Species such as *Dactylorhiza hatagirea*, *Picrorhiza scrophulariiflora*, *Swertia chirayta* and *Rubia manjith* which had been included in government list of medicinal plants prioritized for research and development (Gurung & Pyakurel, 2017), were also documented during this study (Table 6).

Life form classes

A total of 283 species were classified based on life forms in different categories (species which identified up to species level). The life form classes showed that phanerophytes (82 species, 29.07%), therophytes (69 species, 25%) and chamaephytes (49 species, 17.37%) were the most abundant life forms. They were followed by cryptophytes (46 species, 16.25%) and hemicryptophytes (37 species, 13.12%) (Figure 5). *Cuscuta reflexa* was the only parasitic species. The life form is an important physiognomic attribute that has been widely used in vegetation studies (Khan et al., 2013). It indicates micro- and macroclimate as well as human disturbance of a particular area. The life form of plant species reflects the adaptation of plants to the climatic conditions (Shah et al., 2013).

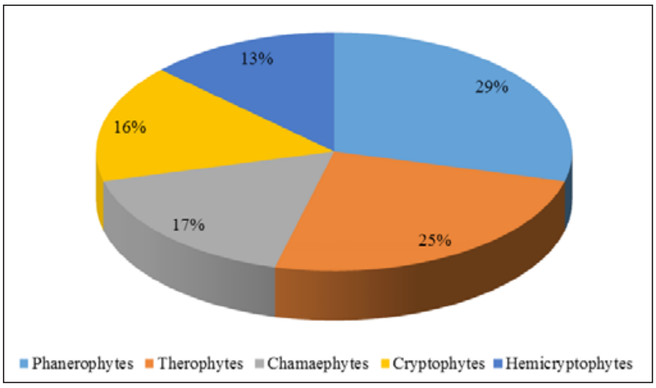

Figure 5: Life-form classes of the flora

The domination of phanerophytes and therophytes over other life forms observed during this study seems to be a response to the topographic divergence, human being and creature disturbance. It also indicates the temperate type (warm and moist) of climate in the study area. The result is contrasting from the study of Joshi et al. (2015) in Nyeshang valley of Manang district within Annapurna Conservation Area where chamaephytes were dominant followed by hemicryptophytes and phenerophytes. Several studies had shown that phanerophytes and therophytes dominance over other life forms might be due to harsh climate and the anthropogenic pressure such as fuel wood collection, grazing, forest fire, lopping and felling of the trees (Khan et al., 2013). Chamaephytes and hemicryptophytes are considered indicators of unfavorable environment and highly vulnerable to any environmental change (Joshi et al., 2015).

Floristic investigations along with life form classification provide reliable information about the nomenclature, distribution, ecology and utility of various plant species. It has been realized that intensive rather than extensive floristic studies of different geographical region are necessary for the proper documentation, conservation plans and sustainable utilization of plant resources (Ali et al., 2018). Due to recurrent forest fires, indiscriminate exploitation of forest resources, destruction of forest areas for construction and introduction of invasive exotic species, several native species are under pressure and may face threat of extinction in future. The description and identification of plants in an area is very important because it shows distinct species in an area and their occurrence in the growing season. Such assessment also helps to identify the ecological vulnerability of the area and to suggest conservation priority (Uprety et al., 2011). It also helps in finding new species of the area and their adjustment in local climatic condition (Ali, 2008).

Conclusion

This study provides fundamental information about the flora of the Sikles region of Annapurna Conservation Area by means of a thorough botanical inventory. Asteraceae and Poaceae were found to be the largest families of dicots and monocots respectively. Likewise, Pinaceae was found to be the largest family of Gymnosperms and Pteridaceae was found to be the largest family of Pteridophytes. The dominance of phanerophytic and therophytic life forms showed that the area was under heavy biotic pressure. These findings could have special significance for further ecological research and for recommendations of proper guidance for the management, reclamation, and development of the area and other similar regions. Describing the floristic composition of a habitat is valuable for continuation of ecological research, management and conservation of plants. Presence of numerous species of *Aconitum*, *Berberis*, *Delphinium*, *Impatiens*, *Potentilla* and *Saxifraga* represents the dominance of Eastern Himalayan floristic elements.

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References

Ali, H., Muhammad, Z., Khan, W.M., Jelani, G., Majeed, A., & Rehmanullah. (2018). Floristic inventory and ecological attributes of plant resources of Hazar Nao hills, district Malakand Pakistan. *Pakistan Journal of Weed Science Research*, *24*(3), 241-255.

Ali, S.I. (2008). The significance of flora with special reference to Pakistan. *Pakistan Journal of Botany*, *40*(3), 967-971.

Behera, M.D., Kushwaha, S.P.S., & Roy, P.S. (2005). Rapid assessment of biological richness in a part of Eastern Himalaya: an integrated three-tier approach. *Forest Ecology and Management*, *207*, 363-384.

Bhattarai, N., Tandon, V., & Ved, D.K. (2002). Highlights and outcomes of the Conservation Assessment and Management Planning (CAMP) Workshop, Pokhara, Nepal. *In:* N. Bhattarai & M. Karki (Eds.), *Sharing local and national experience in conservation of medicinal and aromatic plants in South Asia* (pp. 46-53). New Delhi, India: IDRC.

Chalise, P., Paneru, Y.R., & Ghimire, S.K. (2019). Floristic diversity of vascular plants in Gyasumbdo Valley, Lower Manang, Central Nepal. *Journal of Plant Resources*, *17*(1), 42-57.

Chapin, F.S., Zavaleta, E.S., Eviner, V.T., Naylor, R.L., Vitousek, P.M., Reynolds, H.L., Hooper, D.U., Lavorel, S., Sala, O.E., Hobbie, S.E., Mack, M.C., & Diaz, S. (2000). Consequences of changing biodiversity. *Nature*, *405*(6783), 234-242.

Chaudhary, R.P. (1998). *Biodiversity in Nepal: Status and Conservation*. Bangkok, Thailand: Tec Press Books.

Department of Hydrology and Meteorology. (2018). *Records of Meteorological data 2010-2017*. Kathmandu, Nepal: Author.

Dizro, R., & Raven, P.H. (2003). Global state of biodiversity and loss. *Annual Review of Environment and Resources*, *28*, 137–167.

Fraser-Jenkins, C.R., Kandel, D.R., & Pariyar, S. (2015). *Ferns and fern-allies of Nepal* (Vol. 1). Kathmandu, Nepal: Department of Plant Resources.

Grierson, A.J.C., & Long, D.G. (1983-2000). *Flora of Bhutan* (Vols 1-3). Edinburgh, Scotland: Royal Botanic Garden.

Gurung, K., & Pyakurel, D. (2017). *Identification manual of commercial medicinal and aromatic plants of Nepal*. Kathmandu, Nepal: Nepal Herbs and Herbal Products Association (NEHHPA).

International Union for Nature Conservation. (2019). *The IUCN red List of Threatened Species* (Version 2019-2): Author. Retrieved October 1, 2019, from https://www.iucnredlist.org.

Joshi, L.R., Ghimire, S.K., Salick, J., & Konchar, K.M. (2015). Distribution of vascular plants in a subalpine-nival gradient of Central Himalaya: current patterns and predictions for future warming climate. *Botanica Orientalis*, *9*, 27–39.

Khan, S.M., Page, S., Ahmad, H., Zahidullah. Shaheen, M.A., & Harper, D. (2013). Phytoclimatic gradient of vegetation and habitat specificity in the high elevation Western Himalayas. *Pakistan Journal of Botany*, *45*, 223-230.

Lohbeck, M., Bongers, F., Ramos, M.M., & Poorter, L. (2016). The importance of biodiversity and dominance for multiple ecosystem functions in a human-modified tropical landscape. *Ecology*, *97*(10), 2772–2779.

Palmer, M.W., Wade, G.L., & Neal, P. (1995). Standard for the Writing of Floras. *Bioscience*, *45*(5), 339-345.

Panthi, M.P., Chaudhary, R.P., & Vetas, O.R. (2007). Plant species richness and composition in a trans-Himalayan inner valley of Manang District, Central Nepal. *Himalayan Journal of Sciences*, *4*(6), 31-37.

Polunin, O., & Stainton, J.D.A. (1984). *Flowers of the Himalaya*. New Delhi, India: Oxford University Press.

Plants of the World Online. (2019). Retrieved on 15-28 November 2019 from http://www.plantsoftheworldonline.org.

Press, J.R., Shrestha, K.K., & Sutton, D.A. (2000). *Annotated checklist of the flowering plants of Nepal*. London, UK: The Natural History Museum.

Rajbhandari, K.R., & Rai, S.K. (2017). *A handbook of the flowering plants of Nepal* (Vol. 1). Kathmandu, Nepal: Department of Plant Resources.

Rajbhandari, K.R., & Rajbhandary, S. (2015). Herbarium Preparation and Storage Technique. *In:* M. Siwakoti & S. Rajbhandary (Eds.), *Taxonomic tools and flora writing* (pp. 70-80). Kathmandu, Nepal: Department of Plant Resources and TU Central Department of Botany.

Raunkiaer, C. (1934). *The life form of plants and statistical plant geography*. London, UK: Oxford University Press.

Roskov, Y., Abucay, L., Orrell, T., Nicolson, D., Bailly, N., Kirk, P.M., Bourgoin, T., DeWalt, R.E., Decock, W., DeWever, A., Nieukerken, E.V., Zarucchi, J., & Penev, L. (2019). *Species 2000 & ITIS catalogue of life*. Retrieved November 15-28, 2019, from www.catalogueoflife.org/col.

Shah, M., Hussain, F., Shah, S.N., Ahmad I., & Wasila, H. (2013). Life-form and floristic characteristics along altitudinal gradient of humid temperate forests located in remote area of Pakistan. *Global Journal of Biodiversity Science and Management*, *3*(2), 276- 281.

Shaheen, H., Ullah, Z., Khan, S.M., & Harper, D.M. (2012). Species composition and community structure of western Himalayan moist temperate forests in Kashmir. *Forest Ecology and Management*, *278*, 138–145.

Sharma, P.J.C., Rana, U.D., Randhawa, S.S., & Kumar, R. (2014). Floristic diversity and distribution pattern of plant communities along altitudinal gradient in Sangla Valley, Northwest Himalaya. *The Scientific World Journal*, *2014*, 264878. https://doi.org/10.1155/2014/264878.

Simpson, M.G. (2006). *Plant Systematics*. Massachusetts, USA: Elsevier Academy Press.

Stainton, A. (1988). *Flowers of the Himalaya: A supplement*. New Delhi, India: Oxford University Press.

Takhtajan, A. (1986). *Floristic Regions of the World*; English translation. Berkeley, California, USA: University of California Press.

UNEP-WCMC. (2018). Checklist of CITES species website. Retrieved on September 28, 2019, from http://www.checklist.cites.org.

Uprety, Y., Poudel, R.C., Asselin, H., & Boon, E. (2011). Plant biodiversity and ethnobotany inside the projected impact area of the Upper Seti Hydropower Project, Western Nepal. *Environment, Development and Sustainability*, *13*(3), 463-492.

Welk, E. (2015). Phytogeography of the Nepalese flora and its floristic links to neighbouring regions. *In:* G. Miehe, C. Pendry, & R.P. Chaudhary (Eds.), *Nepal: An introduction to the natural history, ecology and human environment of the Himalayas: A companion volume to the Flora of Nepal* (pp. 140-144). U.K: Royal Botanic Garden Edinburgh.

Wu, Z.Y., & Raven, P.H. (1994). *Flora of China*. Beijing, China: Science Press, and St. Louis, USA: Missouri Botanical Garden Press.

Table 1: Dicotyledons

S.N.	Family	Scientific Name	Collection Number	Life Forms	Habits
1	Acanthaceae	*Achyranthus aspera* L.	KSD391	TH	H
2		*Asystasia macrocarpa* Wall. ex Nees		TH	H
3		*Strobilanthes lachenensis* C. B. Cl.	KSD387	TH	H
4	Acoraceae	*Acorus calamus* L.		CR	H
5	Actinidiaceae	*Saurauia napaulensis* DC.	KSD19	PH	T
6	Adoxaceae	*Viburnum erubescens* Wall.	KSD112	PH	T
7		*Viburnum mullaha* Buch.-Ham. ex D. Don	KSD105	PH	T
8	Amaranthaceae	*Amaranthus spinosus* L.	KSD392	TH	H
9		*Chenopodium album* L.	KSD399	TH	H
10	Anacardiaceae	*Choerospondias axillaris* (Roxb.) B.L. Burtt & A.W. Hill		PH	T
11		*Rhus succedanea* L.	KSD89	PH	T
12	Apiaceae	*Centella asiatica* (L.) Urb.	KSD43	CH	H
13		*Hymenidium benthamii* (Wall. ex DC.) M.G. Pimenov & E. V. Kljuykov	KSD28	CH	H
14		*Cortia depressa* (D. Don) C. Norman	KSD587	HE	H
15		*Selenium* sp.	KSD598		H
16	Apocyanaceae	*Ceropegia pubescens* Wall.		CH	C
17	Aquifoliaceae	*Ilex dipyrena* Wall.	KSD30	PH	T
18	Araliaceae	*Trevesia palmata* (Roxb. ex Lindl.) Vis.	KSD	PH	T
19		*Hedera nepalensis* K. Koch	KSD103	HE	C
20	Asteraceae	*Ageratina adenophora* (Spreng.) R. King & H. Rob.		CH	H
21		*Anaphalis busua* (Buch.-Ham. ex D. Don) DC.	KSD808	CH	H
22		*Anaphalis contorta* (D. Don) Hook. fil.	KSD71	CH	H
23		*Anaphalis triplinervis* (Sims) C. B. Cl.	KSD806	CH	H
24		*Bidens pilosa* L.		TH	H
25		*Cirsium verutum* (D. Don) Spreng.	KSD803	TH	H
26		*Crassocephalum crepidioides* (Benth.) S. Moore.		TH	H
27		*Cremanthodium reniforme* (Wall. ex DC.) Benth.	KSD809	CH	H
28		*Duhalde acappa* (Buch.-Ham. ex D. Don) Pruski & Anderberg	KSD815	PH	S
29		*Senecio graciliflorus* (Wall.) DC.	KSD854	TH	H
30		*Ligularia fischeri* (Ledeb.) Turcz.	KSD824	TH	H
31		*Cicerbita macrorhiza* (Royle) Beauv.	KSD855	TH	H
32		*Pseudognaphalium affine* (D. Don) A. Andeb.	KSD863	TH	H
33		*Dolomiaea macrocephala* Royle		CH	H
34		*Taraxacum officinale* (L.) Weber.	KSD63	TH	H
35		*Ageratum conyzoides* L.		TH	H
36		*Leontopodium stracheyi* (Hook. f.) C. B. Cl. ex Hemsl.	KSD856	CH	H
37		*Artemisia indica* Willd.	KSD05	HE	H
38	Balsaminaceae	*Impatiens edgeworthii* Hook. fil.	KSD781	CH	H
39		*Impatiens racemosa* DC.	KSD782	CH	H
40		*Impatiens* sp.	KSD783		H
41		*Impatiens sulcata* Wall.	KSD784	CH	H
42		*Impatiens urticifolia* Wall.	KSD785	CH	H
43	Begoniaceae	*Begonia dioica* Buch.-Ham. ex D. Don	KSD97	TH	H
44		*Begonia palmata* D. Don	KSD99	TH	H
45		*Begonia picta* Sm.	KSD98	TH	H
46	Berberidaceae	*Berberis aristata* DC.	KSD06	PH	S
47		*Berberis concinna* Hook. fil.	KSD791	PH	S
48		*Berberis napaulensis* (DC.) Spreng.		PH	S
49	Betulaceae	*Alnus nepalensis* D. Don.	KSD07	PH	T

S.N.	Family	Scientific Name	Collection Number	Life Forms	Habits
50		*Betula utilis* D. Don		PH	T
51	Boraginaceae	*Cynoglossum zeylanicum* (Vahl) Thunb. ex Lehm.	KSD45	TH	H
52		*Cynoglossum* sp.	KSD223		H
53		*Maharanga emodi* (Wall.) A. DC.	KSD107	TH	H
54		*Maharanga bicolor* (Wall.ex G. Don) A. DC.	KSD793	TH	H
55	Campanulaceae	*Lobelia pyramidalis* Wall.	KSD46	TH	H
56	Cannabaceae	*Cannabis sativa* L.	KSD47	CH	S
57	Caprifoliaceae	*Nardostachys jatamansi* (D. Don) DC.		CR	H
58		*Dipsacus inermis* Wall.	KSD786	TH	H
59	Caryophyllaceae	*Stellaria* sp.	KSD788	TH	H
60		*Drymaria cordata* (Blume) J.A. Duke	KSD792	CR	H
61	Celastraceae	*Parnassia nubicola* Wall. ex Royle	KSD797	HE	H
62	Convolvulaceae	*Cuscuta reflexa* Roxb.		P	C
63	Crassulaceae	*Rhodiola bupleuroides* (Wall. ex Hook. fil. &Thoms.) Fu	KSD767	CH	S
64	Cucurbitaceae	*Herpetospermum pedunculosum* (Ser.) C.B. Clarke	KSD675	TH	C
65		*Solena amplexicaulis* (Lam.) Gandhi	KSD48	TH	C
66		*Trichosanthes tricuspidata* Lour.	KSD776	TH	C
67	Daphniphyllaceae	*Daphniphyllum himalayense* (Benth.) Mull. Arg.	KSD772	PH	T
68	Elaeagnaceae	*Elaeagnus infundibularis* Momiy.	KSD11	PH	T
69	Ericaceae	*Lyonia ovalifolia* (Wall.) Drude	KSD13	PH	T
70		*Rhododendron arboreum* Sm.	KSD100	PH	T
71		*Rhododendron campanulatum* D. Don	KSD103	PH	T
72		*Vaccinium nummularia* Hook. Fil & b Thoms. ex C.B. Cl.	KSD101	HE	H
73	Euphorbiaceae	*Euphorbia royleana* Boiss.	KSD768	PH	S
74		*Macaranga denticulata* (Blume) Mull. Arg	KSD762	PH	T
75	Fabaceae	*Parochetus communis* D. Don		TH	H
76		*Erythrina arborescens* Roxb.	KSD761	PH	T
77		*Desmodium elegans* DC.	KSD751	TH	S
78		*Desmodium* sp.	KSD752		S
79		*Crotalaria* sp.	KSD755		H
80		*Piptanthus nepalensis* (Hook.) D.Don	KSD102	PH	S
81		*Indigofera heterantha* Wall. ex Brandis	KSD82	PH	S
82	Fagaceae	*Castanopsis indica* (Roxb. ex Lindl.) A.DC.		PH	T
83		*Quercus galuca* Thunb.	KSD31	PH	T
84		*Quercus semecarpifolia* Sm.	KSD735	PH	T
85	Gentianaceae	*Gentiana depressa* D. Don	KSD388	HE	H
86		*Halenia elliptica* D. Don	KSD70	TH	H
87		*Swertia angustifolia* Buch.-Ham. ex D. Don	KSD51	TH	H
88		*Swertia chirayta* (Roxb.) Karst.	KSD50	HE	H
89		*Swertia paniculata* Wall.	KSD78	TH	H
90		*Gentiana capitata* Buch.-Ham. ex D. Don	KSD65	HE	H
91	Geraniaceae	*Geranium* sp.	KSD731		H
92		*Geranium wallichianum* D. Don ex Sweet	KSD732	CH	H
93	Gesneriaceae	*Aeschynanthus hookeri* C.B. Clarke		TH	H
94		*Chirita pumila* D. Don	KSD736	TH	H
95		*Didymocarpus primulifolius* D. Don	KSD115	TH	H
96	Grossulariaceae	*Ribes himalense* Royle ex Decne.	KSD91	PH	T
97	Hydrangeaceae	*Hydrangea febrifuga* (Lour.) Y. De Smet & Granados	KSD113	TH	S
98		*Hydrangea heteromalla* D. Don	KSD84	PH	T
99	Hypericaceae	*Hypericum elodeoides* Choisy	KSD676	TH	H
100		*Hypericum japonicum* Thunb.	KSD373	TH	H
101		*Hypericum cordifolium* Choisy	KSD273	PH	S

S.N.	Family	Scientific Name	Collection Number	Life Forms	Habits
102	Juglandaceae	*Juglans regia* L.		PH	T
103		*Engelhardia spicata* Lesch. ex Bl.	KSD731	PH	T
104	Lamiaceae	*Coolebrokea oppositifolia* Sm.	KSD36	PH	S
105		*Leucosceptrum canum* Sm.	KSD32	PH	T
106		*Phlomoides bracteosa* (Royle ex Benth.) Kamelin & Makhm.	KSD724	TH	H
107		*Stachys melissifolia* Benth.	KSD721	TH	H
108		*Elsholtzia blanda* (Benth.) Benth.	KSD66	TH	H
109		*Elsholtzia fruticosa* (D. Don) Rehder	KSD83	PH	S
110		*Colquhounia coccinea* Wall.	KSD805	PH	S
111	Lardizabalaceae	*Stauntonia angustifolia* (Wall.) Christenh.	KSD04	TH	C
112	Lauraceae	*Lindera neesiana* (Wall. ex Nees) Kruz	KSD33	PH	T
113		*Lindera* sp.		PH	T
114		*Cinnamomum tamala* (Buch.-Ham.) Th. G. G. Nees	KSD55	PH	T
115	Linaceae	*Reinwardtia indica* Dumort.	KSD56	PH	S
116	Magnoliaceae	*Magnolia champaca* (L.) Baill. ex Pierre	KSD14	PH	T
117		*Magnolia* sp.	KSD15	PH	T
118	Mazaceae	*Mazus surculosus* D. Don	KSD569	TH	H
119	Melanthiaceae	*Paris polyphylla* Sm.	KSD57	CR	H
120	Melastomataceae	*Osbeckia stellata* Buch.-Ham. ex D. Don	KSD442	PH	H
121	Meliaceae	*Toona ciliata* M. Roem.	KSD802	PH	T
122	Moraceae	*Ficus auriculata* Lour.	KSD87	PH	T
123		*Ficus religiosa* L.		PH	T
124		*Ficus neriifolia* Sm.	KSD719	PH	T
125		*Ficus semicordata* Buch.-Ham. ex Sm.	KSD86	PH	T
126	Myricaceae	*Myrica esculenta* Buch.-Ham. ex D. Don	KSD714	PH	T
127	Oleaceae	*Chrysojasminum fruticans* (L.) Banfi	KSD657	PH	S
128	Onagraceae	*Epilobium brevifolium* D. Don	KSD716	TH	H
129	Orobanchaceae	*Pedicularis siphonantha* D. Don	KSD59	CH	H
130	Oxalidaceae	*Oxalis corniculata* L.	KSD707	CR	H
131	Papaveraceae	*Corydalis juncea* Wall.	KSD706	CH	H
132		*Corydalis* sp.	KSD708	CH	H
133		*Meconopsis paniculatus* (D. Don) Prain	KSD705	CH	H
134		*Meconopsis regia* G. Tayl.		HE	H
135	Pentaphylacaceae	*Eurya acuminata* DC.	KSD35	PH	T
136	Phyllanthaceae	*Phyllanthus parvifolius* Buch.-Ham. ex D. Don	KSD08	PH	S
137	Phytolaccaceae	*Phytolacca acinosa* Roxb.	KSD60	PH	H
138	Piperaceae	*Piper mullesua* Buch.-Ham. ex D. Don	KSD42	TH	C
139	Plantaginaceae	*Plantago major* L.	KSD701	TH	H
140		*Hemipharagma heterophyllum* Wall.	KSD106	CR	H
141		*Picrorhiza scrophulariiflora* (Pennell) D.Y. Hong	KSD61	CR	H
142	Polygonaceae	*Bistorta macrophylla* (D. Don) Sojak	KSD73	CR	H
143		*Fagopyrum acutatum* (Lehm.) Mansf. ex K. Hammer	KSD101	TH	H
144		*Koenigia polystachya* (Wall. ex Meisn.) T.M.Schust. & Reveal	KSD710	CH	H
145		*Rheum acuminatum* Hook. fil. & Thoms.	KSD771	CH	H
146		*Rumex nepalensis* Spreng.	KSD62	CH	H
147		*Rheum australe* D. Don		CH	H
148		*Koenigia mollis* (D. Don) T.M.Schust. & Reveal	KSD76	CH	H
149	Primulaceae	*Maesa chisia* Buch.-Ham. ex D. Don	KSD34	PH	S
150		*Primula denticulata* Sm.	KSD381	TH	H
151	Ranunculaceae	*Anemone rivularis* Buch.-Ham. ex DC.	KSD364	CH	H
152		*Clematis buchananiana* DC.	KSD371	CH	C
153		*Thalictrum reniforme* Wall.	KSD359	CH	H

S.N.	Family	Scientific Name	Collection Number	Life Forms	Habits
154		*Aconitum spicatum* (Brühl) Stapf	KSD365	TH	H
155		*Aconitum gammiei* Stapf	KSD366	CR	H
156		*Thalictrum foliolosum* DC.	KSD119	CR	H
157		*Delphinium vestitum* Wall.	KSD120	CR	H
158		*Delphinium* sp.	KSD116	CR	H
159		*Ranunculus sceleratus* L.	KSD351	TH	H
160	Rosaceae	*Argentina lineata* (Trevir.) Soják	KSD79	HE	H
161		*Neillia thyrsiflora* D. Don.	KSD94	PH	S
162		*Potentilla* sp.	KSD356		H
163		*Potentilla* sp.	KSD354		H
164		*Prinsepia utilis* Royle	KSD37	PH	S
165		*Pyracantha crenulata* (Roxb. ex D. Don) M.Roemer	KSD26	PH	S
166		*Rubus biflorus* Buch.-Ham. ex Sm.	KSD92	PH	S
167		*Rubus ellipticus* Sm.	KSD87	PH	S
168		*Rubus nepalensis* (Hook. fil.) Kuntze	KSD86	CH	H
169		*Rubus rosifolius* Sm.	KSD88	PH	S
170		*Spiraea bella* Sims.	KSD96	PH	S
171		*Potentilla indica* (Andr.) Wolf	KSD345	HE	H
172		*Fragaria nubicola* Lindl.	KSD344	HE	H
173		*Rosa sericea* Lindl.	KSD336	PH	S
174		*Cotoneaster rotundifolius* Wall. ex Lindley	KSD95	CH	S
175		*Prunus cerasoides* D. Don	KSD16	PH	T
176	Rubiaceae	*Galium elegans* Wall. Ex Roxb.	KSD110	CH	C
177		*Neohymenopogon parasiticus* (Wall.) Bennet	KSD104	PH	S
178		*Rubia manjith* Roxb.	KSD09	TH	C
179	Rutaceae	*Boenninghausenia albiflora* (Hook.) Rchb. ex Meisn.	KSD123	TH	H
180		*Zanthoxylum armatum* DC.	KSD311	PH	T
181		*Zanthoxylum* sp.	KSD315	PH	S
182	Santalaceae	*Pyrularia edulis* (Wall.) A. DC.	KSD317	PH	T
183	Sapindaceae	*Acer cappadocicum* Gled.	KSD38	PH	T
184		*Acer* sp.	KSD328	PH	T
185	Saururaceae	*Houttuynia cordata* Thunb.	KSD122	CR	H
186	Saxifragaceae	*Astilbe rivularis* Buch.-Ham. ex D. Don	KSD121	CH	H
187		*Saxifraga brachypoda* D. Don	KSD318	CH	H
188		*Saxifraga parnassifolia* D. Don	KSD321	CH	H
189		*Bergenia ciliata* (Haw.) Sternb.	KSD324	CR	H
190	Scrophulariaceae	*Buddleja paniculata* Wall.	KSD308	PH	S
191	Simaroubaceae	*Brucea javanica* (L.) Merr.	KSD305	PH	T
192	Solanaceae	*Solanum aculeatissimum* Jacq.	KSD304	TH	H
193		*Solanum nigrum* L.	KSD307	TH	H
194		*Nicotiana tabacum* L.	KSD10	PH	S
195	Symplocaceae	*Symplocos ramosissima* Wall.	KSD17	PH	T
196	Theaceae	*Schima wallichii* Choisy	KSD801	PH	T
197	Thymelaeaceae	*Daphne papyracea* Wall. ex Steud.	KSD03	PH	S
198	Urticaceae	*Boehmeria platyphylla* D. Don	KSD306	CH	H
199		*Elatostema monandrum* (D. Don) H. Hara	KSD303	HE	H
200		*Girardinia diversifolia* (Link) Friis	KSD01	HE	H
201		*Urtica dioca* L.	KSD02	HE	H
202		*Debregeasia salicifolia* (D. Don.) R.	KSD302	PH	S
203		*Pouzolzia sanguinea* (Blume) Merr.	KSD75	HE	H
204	Violaceae	*Viola biflora* L.		HE	H
205		*Viola pilosa* Bl.	KSD64	HE	H
206	Vitaceae	*Tetrastigma serrulatum* (Roxb.) Planch.	KSD63	CH	C

Table 2: Monocotyledons

S.N.	Family	Scientific Name	Collection Number	Life Froms	Habits
1	Amryllidaceae	*Allium wallichii* Kunth	KSD97	CR	H
2	Araceae	*Arisaema costatum* (Wall.) Mart.		CR	H
3		*Arisaema nepenthoides* (Wall.) Mart. ex Schott	KSD80	CR	H
4	Asparagaceae	*Asparagus filicinus* Buch.-Ham. ex D. Don	KSD205	CR	C
5		*Asparagus racemosus* Willd.	KSD20	CR	C
6		*Chlorophytum nepalense* (Lindl.) Baker	KSD114	TH	H
7		*Ophiopogon clarkei* Hook.f.	KSD109	TH	H
8		*Polygonatum cirrhifolium* (Wall.) Royle	KSD21	TH	H
9		*Polygonatum punctatum* Royle ex Kunth	KSD93	TH	H
10		*Maianthemum purpureum* (Wall.) LaFrankie	KSD85	TH	H
11		*Agave cantala* (Haw.) Roxb. ex Salm-Dyck		PH	S
12	Commelinaceae	*Commelina benghalensis* L.	KSD206	CR	H
13	Cyperaceae	*Carex filicina* Nees	KSD208	HE	H
14		*Carex gammiei* (C.B.Clarke) S.R.Zhang & O. Yano	KSD592	HE	H
15		*Carex vesiculosa* Boott	KSD408	HE	H
16		*Cyperus brevifolius (*Rottb.) Hassk.	KSD209	HE	H
17	Dioscoreaceae	*Dioscorea deltoidea* Wall. ex Griseb.	KSD403	CR	C
18		*Dioscorea bulbifera* L.	KSD22	CR	C
19	Juncaceae	*Juncus himalensis* Klotzsch	KSD521	CR	H
20		*Juncus thomsonii* Buch.	KSD215	CR	H
21		*Luzula multiflora* (Retz.) Lej.	KSD211	CR	H
22	Liliaceae	*Cardiocrinum giganteum* (Wall.) Makino		TH	H
23		*Fritillaria cirrhosa* D. Don		CR	H
24	Orchidaceae	*Calanthe tricarinata* Lindl.	KSD216	CR	H
25		*Coelogyne corymbosa* Lindl.	KSD578	CR	H
26		*Coelogyne cristata* Lindl.	KSD77	CR	H
27		*Cymbidium iridioides* D. Don	KSD225	CR	H
28		*Dendrobium porphyrochilum* Lindl.	KSD245	CR	H
29		*Eria coronaria* (Lindl.) Rchb.f.	KSD228	HE	H
30		*Malaxis muscifera* (Lindl.) Kuntze	KSD556	CR	H
31		*Neottia pinetorum* (Lindl.) Szlach.	KSD249	CR	H
32		*Oberonia* sp.			H
33		*Oberonia falcata* King & Pantl.	KSD280	TH	H
34		*Oreorchis micrantha* Lindl.	KSD289	CR	H
35		*Platanthera* sp.	KSD298		H
36		*Pleione humilis* (Sm.) D. Don	KSD584	CR	H
37		*Satyrium nepalense* D. Don	KSD218	CR	H
38		*Dactylorhiza hatagirea* (D. Don) Soó	KSD58	CR	H
39		*Dendrobium amoenum* Wall. ex Lindl.	KSD219	CR	H
40	Poaceae	*Agrostis micrantha* Steud.	KSD231	TH	H
41		*Bromus himalaicus* Stapf	KSD232	TH	H
42		*Chrysopogon gryllus* (L.) Trin.	KSD510	HE	H
43		*Cyrtococcum patens* var. *latifolium* (Honda) Ohwi	KSD234	TH	H
44		*Festuca* sp.	KSD235		H
45		*Isachne albens* Trin.	KSD236	TH	H
46		*Poa infirma* Kunth		TH	H
47		*Setaria plicata* (Lam.) T. Cooke	KSD237	TH	H
48		*Themeda arundinacea* (Roxb.) A.Camus	KSD244	TH	H
49		*Trisetum spicatum* (L.) K.Richt.	KSD555	TH	H
50		*Cynodon dactylon* (L.) Pers.		HE	H
51		*Dendrocalamus hamiltonii* Nees & Arn. Munro		CR	H
52		*Thysanolaena latifolia* (Roxb.) Kuntze		CH	H

S.N.	Family	Scientific Name	Collection Number	Life Froms	Habits
53		*Thamnocalamus spathiflorus* (Trin.) Munro		CH	H
54		*Himalayacalamus brevinodus* Stapleton		CH	H
55		*Eulaliopsis binata* (Retz.) C.E. Hubb.	KSD247	HE	H
56		*Imperata cylindrica* (L.) P. Beauv.	KSD248	HE	H
57	Smilacaceae	*Smilax aspera* L.	KSD27	CR	C
58		*Smilax menispermoidea* A. DC.	KSD241	CR	C
59	Zingiberaceae	*Hedychium spicatum* Sm.	KSD238	CR	H

Table 3: Gymnosperms

S.N.	Family	Scientific Name	Collection Number	Life Forms	Habits
1	Taxaceae	*Taxus wallichiana* Zucc.	KSD18	PH	T
2	Pinaceae	*Pinus wallichiana* A.B. Jacks	KSD202	PH	T
3		*Abies* sp.		PH	T
4	Cupressaceae	*Juniperus squamata* Buch.-Ham. ex D. Don	KSD29	PH	T
5		*Juniperus recurva* Buch.-Ham. ex D. Don	KSD206	PH	T

Table 4: Pteridophytes

S.N.	Family	Scientific Name	Collection Number	Life Forms	Habits
1	Athyriaceae	*Diplazium esculentum* (Retz.) Sw.	KSD610	CH	H
2	Cyatheaceae	*Alsophila spinulosa* (Wall. ex Hook.) R. M. Tryon		PH	T
3	Dennstaedtiaceae	*Pteridium revolutum* (Bl.) Nakai	KSD612	HE	H
4		*Dennstaedtia appendiculata* (Wall. ex Hook.) J. Sm.	KSD614	CH	H
5	Dryopteridaceae	*Dryopteris barbigera* (Hook.) O. Kuntze	KSD553	HE	H
6		*Polystichum* sp.	KSD604	HE	H
7	Equisetaceae	*Equisetum arvense* L.	KSD615	CR	H
8	Gleicheniaceae	*Dicranopteris linearis* (Brum.fil.) Underw.	KSD609	HE	H
9		*Diplopterygium giganteum* (Wall. ex Hook.) Nakai	KSD607	HE	H
10	Lycopodiaceae	*Palhinhaea cernua* (L.) Carv. Vasc. & Franco	KSD619	CH	C
11		*Phlegmariurus pulcherrimus* (Hook. & Grev.) Löve & Löve	KSD620	CH	H
12		*Lycopodium clavatum* L.	KSD558	CH	H
13	Nephrolepidaceae	*Nephrolepis cordifolia* (L.) Presl	KSD625	CR	H
14	Oleandraceae	*Oleandra wallichii* (Hook.) Presl	KSD623	CR	H
15	Ophioglossaceae	*Ophioglossum reticulatum* L.		TH	H
16		*Japanobotrychum lanuginosum* (Wall. ex Hook. & Grev.) M. Nishida ex Tagawa	KSD621	CR	H
17	Polypodiaceae	*Pyrrosia flocculosa* (D. Don) Ching	KSD635	CH	H
18		*Lepisorus nudus* (Hooker) Ching	KSD634	CH	H
19	Pteridaceae	*Haplopteris taeniophylla* (Copel.) E. H. Crane	KSD630	CH	H
20		*Onychium siliculosum* (Desv.) C. Chr.	KSD631	CH	H
21		*Pteris aspericaulis* Wall. ex Ag.	KSD632	HE	H
22		*Aleuritopteris rufa* (D.Don) Ching	KSD627	HE	H
23		*Pteris* sp.	KSD626	HE	H
24	Selaginellaceae	*Selaginella* sp.	KSD628	HE	H
25	Tectariaceae	*Tectaria coadunata* (J. Smith) C. Christensen		HE	H

Note: Life Froms: PH-Phanerophytes, CH-Chamaephytes, HE-Hemicryptophytes, TH-Therophytes, CR-Cryptophytes, P-Parasite
Habits: C-Climber, H-Herb, S-Shrub, T-Tree

Table 5: Raunkier classification description

S.N.	Life Form	Description
1	**Chamaephytes**	Species with perenating buds born on aerial parts but close to the ground (no more than 25 cm above the soil surface).
2	**Cryptophytes (Geophytes)**	Plant species with buds or shoot apices which survive the unfavorable period below ground or water (species with rhizome, bulb or tuber).
3	**Hemicryptophytes**	All above ground parts of the plant die back in unfavorable conditions and buds are born at ground surface.
4	**Phanerophytes**	Woody species with perenating buds emerging from aerial parts.
5	**Therophytes**	Plant species survive unfavorable condition as seeds (annuals).

Table 6: Prioritized species by different organizations

S.N.	Scientific Name	CAMP	CITES	IUCN	GON/Government Activity
1	*Taxus wallichiana* Zucc.	Endangered	II	Endangered	2
2	*Dolomiaea macrocephala* Royle	Near Threatened			
3	*Maharanga emodi* (Wall.) A. DC.	Data Deficit			
4	*Maharanga bicolor* (Wall.ex G. Don) A. DC.	Data Deficit			
5	*Nardostachys jatamansi* (D. Don) DC.	Vulnerable	II	Critically Endangered	2/ Medicinal Plants for research and development
6	*Swertia angustifolia* Buch.-Ham. ex D. Don	Endangered			
7	*Swertia chirayta* (Roxb.) Karst.	Vulnerable			Medicinal Plants for research and development
8	*Paris polyphylla* Sm	Vulnerable			Medicinal Plants for research and development
9	*Picrorhiza scrophulariiflora* (Pennell) D.Y. Hong	Vulnerable			3/ Medicinal Plants for research and development
10	*Rheum australe* D. Don	Vulnerable			Medicinal Plants for research and development
11	*Aconitum spicatum* (Brühl) Stapf	Vulnerable			Medicinal Plants for research and development
12	*Rubia manjith* Roxb.	Vulnerable			Medicinal Plants for research and development
13	*Arisaema costatum* (Wall.) Mart	Least Concern			
14	*Asparagus racemosus* Willd.	Vulnerable			Medicinal Plants for research and development
15	*Fritillaria cirrhosa* D. Don	Vulnerable			
16	*Dactylorhiza hatagirea* (D. Don) Soó	Endangered	II		1/ Medicinal Plants for research and development
17	*Dioscorea deltoidea* Wall. ex Griseb.	Endangered	II		
18	*Alsophila spinulosa* (Wall. ex Hook.) R. M. Tryon		II		
19	*Zanthoxylum armatum* DC				Medicinal Plants for research and development
20	*Juglans regia* L. (Barks)				1

Note: GoN: Government of Nepal 1= Complete ban: Ban for collection, use, trade, transportation and export, 2= Ban raw export: Banned for export outside the country without processing, 3= Conditional harvesting: The wild harvest and sale allowed only after the taxonomic identification and confirmation of the species as *Picrorhiza scrophulariiflora* Pennell by DPR, and then the final approval of DFO after its inventory and identification of its total natural and harvestable stock in the wild; IUCN: International Union for Nature Conservation; CAMP: Conservation Assessment and Management Plan; CITES: Convention on International Trade in *Endangered Species* of Wild Fauna and Flora

Floristic Diversity of Vascular Plants in Daman and Adjoining Areas, Makawanpur District, Central Nepal

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Abstract

A systematic investigation of floristic diversity of vascular plants from Daman and the adjoining areas of Makawanpur district, Central Nepal was carried out. The study aimed at identifying the diversity of vascular plants, thus included angiosperms, gymnosperms as well as pteridophytes. Altogether, 136 vascular plant species were documented including 121 angiosperms (98 dicots and 23 monocots), 4 gymnosperms and 11 species of ferns and fern allies. Rosaceae, with 5 genera and 8 species, was found to be the largest family. *Rubus*, with 4 species, was found to the largest genus followed by *Thalictrum*, *Anaphalis* and *Hypericum* with 3 species each. Among dicots, Rosaceae was found to be the dominant family, whereas Orchidaceae was the dominant family among monocots. Similarly, Pteridaceae was the dominant family of fern and fern allies. Hence, Daman and the adjoining areas of Makawanpur district were observed to be rich in terms of biodiversity and were concluded to serve as important harbors of threatened and protected plant species as well as high value medicinal and aromatic plants.

Keywords: Biodiversity, Orchidaceae, Pteridaceae, Rosaceae, *Rubus*.

Introduction

Biodiversity includes diversity within species and between species, and ecosystems (Chaudhary, 1998). It is not evenly distributed everywhere, rather it varies greatly across the globe as well as within different geographical regions. The Convention on Biological Diversity (1992) defines documentation of the biodiversity as one of the most prioritized tasks by the world. Biodiversity documentation is possible through extensive botanical exploration and floristic studies (Chalise et al., 2018). Floristic study refers to the documentation of all plants species in a given geographical region (Simpson, 2006). Such studies help in botanical enumeration, updating nomenclature changes, adding herbarium specimens in the existing herbaria and comparison of close or distantly related plants (Chalise et al., 2018). The results of such floristic studies mostly come in the form of floras (Palmer et al., 1995) which may be local, regional, country-wise and so on, or they may be in the form of checklists (Chalise et al., 2018).

Nepal is a mountainous country in the Central Himalayas, which occupies about one-third of the entire length of the Himalayan mountain range and exhibits a unique assemblage of different habitats and a great biodiversity within a small geographical area (Paudel et al., 2011). Nepal lies in a transitional zone between Eastern and Western Himalayan flora (Takhtajan, 1984), thus, is gifted by over 7000 species (Paudel et al., 2011) of vascular plants among which majority are flowering plants. Regarding the history of botanical exploration, Buchanan-Hamilton visited Nepal, especially Central Nepal, from 1802-1803 and collected 433 plant specimens from Makawanpur to Kathmandu along the route. After the establishment of Department of Medicinal Plants in 1960, systematic plant collections were started from Makawanpur by M. S. Bista in collaboration with Japanese botanist H. Kanai during April, 1969 (Joshi, 2014).

Geographically, Makawanpur district lies in the Churia and Mid-Hills of Nepal, and serves as an important hot spot of plant diversity. Joshi (2014) reported 695 angiospermic plant species belonging to 472 genera and 124 families from Makawanpur district while Chapagain et al. (2016) reported a total of 1,045 angiospermic plants belonging to 677 genera and 161 families and 23 gymnosperms belonging to 16 genera and 10 families. Previous studies have indicated towards the occurrence of rich plant biodiversity in Makawanpur district. Thus, extensive botanical explorations should be carried out in order to document as well as update the biodiversity profile of the area. This study was carried out to document the floristic diversity of vascular plants from Daman and the adjoining areas of Makawanpur district.

Materials and Methods

Study site

Makawanpur district lies in Narayani Zone of Nepal and covers an area of 2,426 km² and with a population of 420,477 (CBS, 2012) and is located about 34 km south of the Capital City- Kathmandu. It was the traditional route connecting Mechi-Mahakali with Kathmandu till Prithvi Highway substituted its role. Out of the total area of this district, 25.15% is cultivable, 2.03% is covered with meadows and bushes, 6.83% is occupied by rivers and their shores, 59.145% is filled with forest, 0.66% is an industrial area while 6.19% is occupied by Conservation Area. It is made up of 75% hilly area and 25% flat lands. The major hill ranges include Chandragiri Hill, Mahabharat Hill and Churia hill ranges. The map of study area is given in Figure 1.

The study area consists mainly of three types of forests i.e Laurel- Oak forest, Pine forest and Mixed broad-leaved forest. Laurel- Oak forest occupies majority of the forest in the study area. In lower parts, Oak forest is associated with *Pinus roxburghi* in some places and with *Juglans regia* in the other places. Common shrubs and small trees are *Sarcococca hookeriana*, *Pieris formosa*, *Lindera pulcherrima*, etc, but in the upper parts, oak trees are associated with *Pinus wallichiana*, *Pyrus* sp. etc. with undergrowth of shrubs and small trees. The pine forest, composed exclusively of *Pinus roxburghii*, occupies the southern slopes of the hill mainly at lower altitude. Here only small trees of *Myrica esculenta*, *Schima wallichii* were noted forming the second layer. The common shrubs are *Berberis asiatica*, *Daphne papyracea*, *Rubus ellipticus*, etc. The broad-leaved forests occur in the northern slopes of the ridge and consists main component along with *Quercus glauca*. Very few trees of *Rhododendron arboreum* are found in the forest.

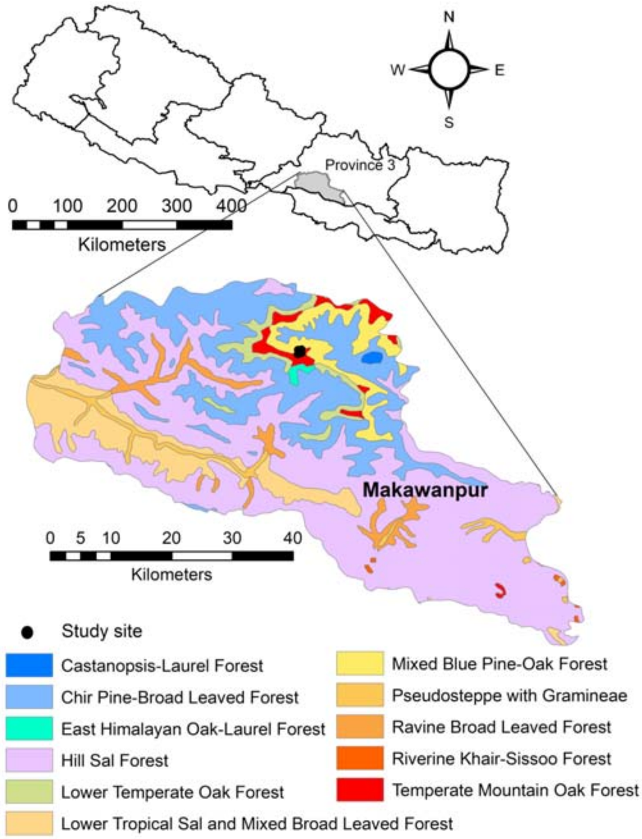

Figure1: Map of Makawanpur district showing different vegetation types in and around the study site.

Methods

Voucher specimens of majority of the vascular plant species, in either flowering or fruiting stage, were collected from the study area during August-September, 2019, especially along the trail from which herbarium specimens were prepared. Identification of those voucher specimens was carried out by following relevant literatures (Grierson & Long, 1983-2001; Polunin & Stainton, 1984; Stainton, 1988; Zheng-Yi et al., 1996-2003; Press et al., 2000; Ohba et al., 2008; Fraser-Jenkins et al., 2015, Chapagain et al., 2016). Identifications were also done by comparing with herbarium specimens at National Herbarium & Plant Laboratories (KATH) and high resolution herbarium images of Herbarium of the University of Tokyo, Japan (TI) and Herbarium, Royal Botanical Garden, Kew, UK (K). Nomenclature follows the Catalogue of Life (Roskov et al., 2019). The herbarium specimens prepared were deposited at KATH.

Results and Discussion

The present study documented a total of 136 vascular plant species including 121 angiosperms, 4 gymnosperms and 11 species of ferns and fern allies. Altogether, 98 dicotyledonous plants belonging to 85 genera of 49 families were recorded during this study. Similarly, 23 monocotyledonous plants belonging to 23 genera of 11 families, 4 species of gymnosperms belonging to 4 genera of 4 different families and 11 species of fern and fern allies belonging to 10 genera of 4 different families were recorded during this study (Figure 2).

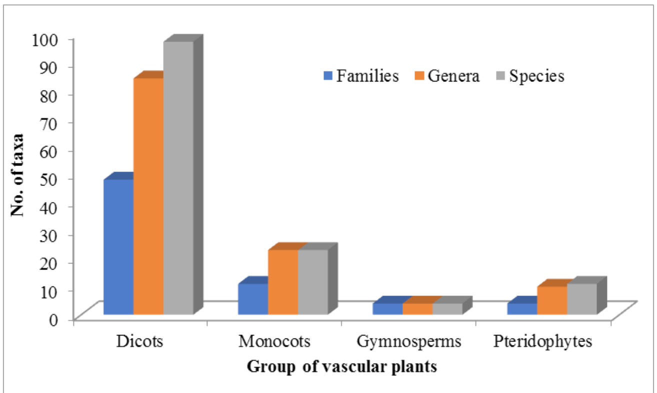

Figure 2: Variation in number of vascular plants in the study area as per the number of families, genera and species.

Rosaceae was the dominant family with eight species belonging to 5 different genera (Figure 3). It was followed by Fabaceae (7 species), Asteraceae (6 species), Ericaceae, Ranunculaceae and Orchidaceae (5 species each), Lamiaceae, Caprifoliaceae, Pteridaceae and Polypodiaceae (4 species each) and so on (Figure 3). However, Joshi (2014) reported, Fabaceae as the largest family of angiosperms in the study area which was followed by Asteraceae.

Comparing the status of dicots as well as monocots, Rosaceae was the dominant family of dicots, whereas Orchidaceae was the dominant family of monocots. Joshi (2014), also reported Orchidaceae as the dominant family of monocots from the study area. Similarly, Pteridaceae was the dominant family of fern and fern allies. *Rubus* was the dominant genus with 4 different species, which was followed by *Thalictrum*, *Anaphalis* and *Hypericum* with 3 species each and by *Geranium*, *Begonia*, *Impatiens*, *Cyathula* and *Lepisorus* with two species each.

Some of the protected plant species such as *Asparagus racemosus* Willd. (Vulnerable: CAMP), *Bergenia ciliata* (Haw.) Sternb (Rare: CAMP), *Juglans regia* L. (Protected), Orchids viz; *Bulbophyllum* sp., *Calanthe* sp., *Goodyera fusca* (Lindl.) Hook. f, *Satyrium nepalense* D.Don, *Spiranthes sinensis* (Pers.) Ames. (CITES II), *Ceropegia pubescens* Wall. (CITES listed plant), *Rubia manjith* Roxb. (Vulnerable: CAMP), *Swertia chirayita* (Roxb.) H. Karst. (Vulnerable: CAMP), etc. were also recorded during the present study (Shrestha & Shrestha, 2012; Joshi et al., 2017) Sharma et al., 2017; Dhakal & Saud, 2018). Together, some of the high valued medicinal plants such as *Allium wallichii* Kunth., *Asparagus racemosus* Willd., *Swertia chirayita* (Roxb.) H. Karst., *Bergenia ciliate* (Haw.) Sternb, *Astilbe rivularis* Buch.-Ham. ex D. Don, *Valeriana hardwickei*, *Schizotechium monospermum*, *Stauntonia latifolia*, *Phyllanthus parvifolius*, *Chlorophytum nepalense*, *Ophioglossum petiolatum*, *Gaultheria fragrantissima*, etc. were also reported from this study. Joshi (2014) also reported the occurrence of many medicinal and aromatic plants from the study area which, if understood and utilized properly, can provide economic benefit to the local people.

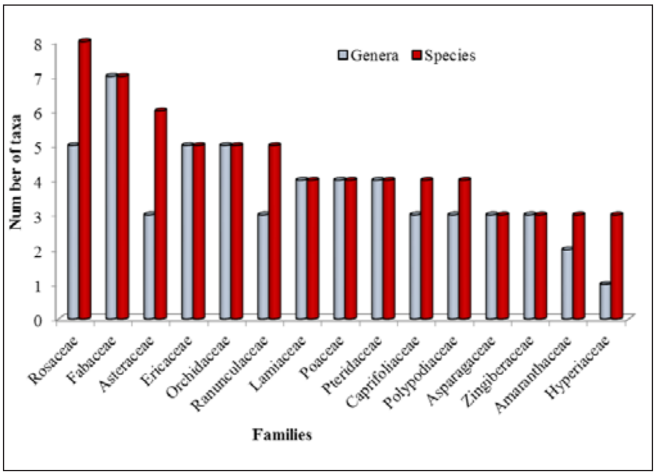

Figure 3: Dominant families of vascular plants in the study area.

Table 1 : List of Angiosperms (Dicots) collected from Daman and adjoining areas of Makawanpur district, Central Nepal

S. N.	Scientific Names	Family	Collection date	Collection No.
1.	*Achyranthes aspera* L.	Amaranthaceae	-	-
2.	*Aeschynanthus hookeri* C.B. Clarke	Gesneriaceae	-	-
3.	*Ageratina adenophora* (Spreng.) R. King and H. Rob.	Asteraceae	-	-
4.	*Ainsliaea latifolia* (D. Don) Sch. Bip.	Asteraceae	-	-
5.	*Alnus nepalensis* D.Don	Betulaceae	-	-
6.	*Anaphalis contorta* (D. Don) Hook.f.	Asteraceae	-	-
7.	*Anaphalis margaritacea* (L.) Benth. and Hook.f	Asteraceae	2/09/2019	D0063
8.	*Anaphalis triplinervis* Sims. ex C.B. Clarke.	Asteraceae	2/09/2019	D0057
9.	*Astilbe rivularis* Buch.-Ham. ex D.Don	Saxifragaceae	1/09/2019	D0044
10.	*Begonia dioica* Buch.-Ham. ex D.Don	Begoniaceae	-	-
11.	*Begonia picta* Sm.	Begoniaceae	-	-
12.	*Berberis asiatica* Roxb. ex DC.	Berberidaceae	-	-
13.	*Bergenia ciliata* (Haw.) Sternb.	Saxifragaceae	-	-
14.	*Bistorta amplexicaulis* (D. Don) Greene.	Polygonaceae	1/09/2019	D0032
15.	*Bupleurum longicaule* Wall.	Apiaceae	1/09/2019	D0043
16.	*Campanula pallida* Wall.	Campanulaceae	1/09/2019	D0028
17.	*Ceropegia pubescens* Wall.	Apocynaceae	1/09/2019	D0040
18.	*Chrysojasminum humile* (L.) Banfi	Oleaceae	-	-
19.	*Clematis buchananiana* DC.	Ranunculaceae	-	-
20.	*Clinopodiumum brosum* (M. Bieb.) K. Koch	Lamiaceae	1/09/2019	D0033
21.	*Crassula* sp.	Crassulaceae	30/08/2019	D0018
22.	*Crotolaria albida* B.Heyne ex Roth	Fabaceae	1/09/2019	D0027
23.	*Cyathula capitata* Moq.	Amaranthaceae	-	-
24.	*Cyathula tomentosa* (Roth) Moq.	Amaranthaceae	-	-
25.	*Cynoglossum zeylanicum* (Sw. ex Lehm.) Thunb. ex Brand	Boraginaceae	30/08/2019	D0017
26.	*Daphne papyracea* Wall. ex G.Don	Thymelaeaceae	-	-
27.	*Dipsacu sinermis* Wall.	Caprifoliaceae	1/09/2019	D0037
28.	*Drosera peltata* Thunb.	Droseraceae	29/08/2019	D0003
29.	*Elsholtzia blanda* (Benth.) Benth.	Lamiaceae	-	-
30.	*Epilobium wallichianum* Hausskn.	Onagraceae	-	-
31.	*Eriocapitella vitifolia* (Buch.-Ham.ex DC.) Nakai	Ranunculaceae	30/08/2019	D0013
32.	*Eurya acuminata* DC.	Pentaphyllaceae	3/09/2019	D0075
33.	*Galinsoga parviflora* Cav.	Asteraceae	-	-
34.	*Galium hirtiflorum* Req. ex DC.	Rubiaceae	3/09/2019	D0067
35.	*Gaultheria fragrantissima* Wall.	Ericaceae	2/09/2019	D0054
36.	*Gentiana capitata* Buch.-Ham. ex D.Don	Gentianaceae	-	-
37.	*Geranium donianum* Sweet	Geraniaceae	30/08/2019	D0015
38.	*Geranium nepalense* Sweet	Geraniaceae	-	-
39.	*Grona heterocarpa* (L.) H.Ohashi and K.Ohashi	Fabaceae	-	-
40.	*Hedera nepalensis* K.Koch	Araliaceae	-	-
41.	*Hemiphragma heterophyllum* Wall.	Plantaginaceae	-	-
42.	*Henckelia pumila* (D.Don) A.Dietr.	Gesneriaceae	-	-
43.	*Hydrangea febrifuga* (Lour.) Y.De Smet and Granados	Hydrangeaceae	-	-
44.	*Hypericum elodeoides* Choisy.	Hypericaceae	30/08/2019	D0012
45.	*Hypericum* sp.	Hyperiaceae	1/09/2019	D0029

S. N.	Scientific Names	Family	Collection date	Collection No.
46.	*Hypericum uralum* Buch.-Ham. ex D. Don	Hypericaceae	2/09/2019	D0061
47.	*Impatiens racemosa* DC.	Balsaminaceae	3/09/2019	D0070
48.	*Impatiens scabrida* DC.	Balsaminaceae	31/08/2019	D0024
49.	*Indigofera cassioides* Rottler ex DC.	Fabaceae	1/09/2019	D0025
50.	*Juglans regia* L.	Juglandaceae	-	-
51.	*Koenigia mollis* (D.Don) T.M.Schust. and Reveal	Polygonaceae	1/09/2019	D0038
52.	*Lecanthus peduncularis* (Royle) Wedd.	Urticaceae	31/08/2019	D0023
53.	*Lindera pulcherrima* (Nees) Benth. ex Hook.f	Lauraceae	-	-
54.	*Litsea* sp.	Lauraceae	31/08/2019	D0021
55.	*Lonicera japonica* Thunb.	Caprifoliaceae	1/09/2019	D0034
56.	*Lyonia ovalifolia* (Wall.) Drude.	Ericaceae	3/09/2019	D0066
57.	*Maesa chisia* D.Don	Primulaceae	-	-
58.	*Magnolia lanuginosa* (Wall.) Figlar and Noot.	Magnoliaceae	29/08/2019	D0001
59.	*Myrica esculenta* Buch.-Ham. ex D.Don	Myricaceae	-	-
60.	*Neohymenopogon parasiticus* (Wall.) Bennet	Rubiaceae	3/09/2019	D0069
61.	*Oxalis corniculata* L.	Oxalidaceae	-	-
62.	*Parochetus communis* Buch.-Ham. ex D.Don	Fabaceae	-	-
63.	*Pedicularis gracilis* Wall. ex Benth.	Orobranchaceae	3/09/2019	D0071
64.	*Phlomoides* sp.	Lamiaceae	2/09/2019	D0062
65.	*Phyllanthus parvifolius* Buch.-Ham. ex D. Don	Euphorbiaceae	1/09/2019	D0039
66.	*Pieris formosa* (Wall.) D. Don	Ericaceae	3/09/2019	D0065
67.	*Pilea* sp.	Urticaceae	1/09/2019	D0042
68.	*Piptanthus nepalensis* (Hook.) Sweet	Fabaceae	-	-
69.	*Potentilla fulgens* Wall. ex Sims.	Rosaceae	2/09/2019	D0056
70.	*Prunella vulgaris* L.	Lamiaceae	30/08/2019	D0016
71.	*Prunus napaulensis* (Ser.) Steud.	Rosaceae	3/09/2019	D0078
72.	*Pteracanthus* sp.	Acanthaceae	1/09/2019	D0035
73.	*Pyrus pashia* Buch.-Ham. ex D.Don	Rosaceae	3/09/2019	D0064
74.	*Quercus semecarpifolia* Sm.	Fagaceae	29/08/2019	D0009
75.	*Rhododendron arboretum* Sm.	Ericaceae	-	-
76.	*Rubia manjith* Roxb.	Rubiaceae	-	-
77.	*Rubus acuminatus* Sm.	Rosaceae	-	-
78.	*Rubus ellipticus* Sm.	Rosaceae	-	-
79.	*Rubus nepalensis* (Hook.f.) Kuntze.	Rosaceae	1/09/2019	D0047
80.	*Rubus* sp.	Rosaceae	2/09/2019	D0053
81.	*Schizotechium monospermum* (Buch.-Ham. ex D.Don) Pusalkar and S.K.Srivast.	Caryophyllaceae	-	-
82.	*Scurrula elata* (Edgew.) Danser	Loranthaceae	-	-
83.	*Spergula arvensis L.*	Caryophyllaceae	29/08/2019	D0006
84.	*Spiraea bella* Sims.	Rosaceae	31/08/2019	D0022
85.	*Stauntonia latifolia* (Wall.) R.Br. ex Wall.	Lardizabalaceae	-	-
86.	*Strobilanthes sp.*	Acanthaceae	-	-
87.	*Swertia chirayita* (Roxb.) H.Karst.	Gentianaceae	29/08/2019	D0005
88.	*Tetrastigma serrulatum* (Roxb.) Planch	Vitaceae	-	-
89.	*Thalictrum chelidonii* DC.	Ranunculaceae	1/09/2019	D0046
90.	*Thalictrum cultratum* Wall.	Ranunculaceae	29/08/2019	D0007
91.	*Thalictrum foliolosum* DC.	Ranunculaceae	-	-

S. N.	Scientific Names	Family	Collection date	Collection No.
92.	*Trifolium repens* L.	Fabaceae	-	-
93.	*Vaccinium nummularia* Hook.f. and Thomson ex C.B. Clarke	Ericaceae	30/08/2019	D0014
94.	*Valeriana hardwickei* Wall.	Caprifoliaceae	-	-
95.	*Viburnum cylindricum* Buch.-Ham. ex D.Don	Caprifoliaceae	3/09/2019	D0068
96.	*Vigna munro* (L.) Hepper.	Fabaceae	31/08/2019	D0020
97.	*Viola biflora* L.	Violaceae	-	-
98.	*Wikstroemia canescens* Meisn.	Thymelaeaceae	2/09/2019	D0058

Table 2 : List of Angiosperms (Monocots)

S. N.	Scientific Names	Family	Collection date	Collection No.
1.	*Allium wallichii* Kunth.	Amaryllidaceae	2/09/2019	D0060
2.	*Arundinella nepalensis* Trin.	Poaceae	-	-
3.	*Asparagus racemosus* Willd.	Asparagaceae	-	-
4.	*Bulbophyllum* sp.	Orchidaceae	-	-
5.	*Calanthe* sp.	Orchidaceae	1/09/2019	D0026
6.	*Cardiocrinum giganteum* (Wall.) Makino	Liliaceae	3/09/2019	D0074
7.	*Carex cruciata* Wahlenb.	Cyperaceae	-	-
8.	*Cautleya spicata* (Sm.) Baker	Zingiberaceae	2/09/2019	D0052
9.	*Chlorophytum nepalense* (Lindl.) Baker	Asparagaceae	-	-
10.	*Cyanotis cristata* (L.) D.Don	Commelinaceae	3/09/2019	D0073
11.	*Elymus* sp.	Poaceae	1/09/2019	D0049
12.	*Globba clarkei* Baker	Zingiberaceae	-	-
13.	*Goodyera fusca* (Lindl.) Hook.f	Orchidaceae	-	-
14.	*Iris* sp.	Iridaceae	1/09/2019	D0036
15.	*Juncus thomsonii* Buchenau.	Juncaceae	1/09/2019	D0030
16.	*Lilium nepalense* D. Don	Liliaceae	29/08/2019	D0004
17.	*Miscanthus nepalensis* (Trin.) Hack.	Poaceae	2/09/2019	D0055
18.	*Roscoea purpurea* Sm.	Zingiberaceae	29/08/2019	D0002
19.	*Satyrium nepalense* D.Don	Orchidaceae	29/08/2019	D0010
20.	*Setaria* sp.	Poaceae	-	-
21.	*Smilax elegans* Wall. ex Kunth.	Smilaceae	1/09/2019	D0048
22.	*Spiranthes sinensis* (Pers.) Ames.	Orchidaceae	30/08/2019	D0011
23.	*Theropogon pallidus* (Wall. ex Kunth) Maxim.	Asparagaceae	-	-

Table 3 : List of Gymnosperms.

S. N.	Scientific Names	Family	Collection date	Collection No.
1.	*Pinus wallichiana* A. B. Jacks	Pinaceae	2/9/2019	D0059
2.	*Thuja orientalis* L.	Cupressaceae	-	-
3.	*Ginkgo biloba* L.	Ginkgoaceae	-	-
4.	*Araucaria bidwilli* Hook.	Araucariaceae	-	-

Table 4 : List of Pteridophytes.

S. N.	Scientific Names	Family	Collection date	Collection No.
1.	*Adiantum tibeticum* Ching and Y.X.Lin	Pteridaceae	-	-
2.	*Aglaomorpha mollis* (Bedd.) Hovenkamp and S.Linds.	Polypodiaceae	-	-
3.	*Asplenium* sp.	Aspleniaceae	1/09/2019	D0045
4.	*Coniogramme* sp.	Pteridaceae	3/09/2019	D0072
5.	*Dryopteris filix-mas* (L.) Schott	Polypodiaceae	-	-
6.	*Japanobotrychum lanuginosum* (Wall. ex Hook. and Grev.) M. Nishida ex Tagawa	Ophioglossaceae	1/09/2019	D0041
7.	*Lepisorus* sp.	Polypodiaceae	29/08/2019	D0008
8.	*Lepisorus* sp.	Polypodiaceae	31/08/2019	D0019
9.	*Onychium cryptogrammoides* Christ.	Pteridaceae	-	-
10.	*Ophioglossum petiolatum* Hook.	Ophioglossaceae	-	-
11.	*Pteridium* sp.	Pteridaceae	1/09/2019	D0051

Conclusion

Heterogenous landscape as well as the climatic variation within short spatial scale make Makawanpur district, especially Daman and the adjoining areas, rich in biodiversity. These divine places not only serve as an important harbor of threatened and protected plant species, but also provide shelter to several high valued medicinal and aromatic plants. Therefore, they have always been the key attraction to the national as well as international researchers and tourists.

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References

Central Bureau of statistics. (2012). *National Population and Housing Census 2011*. National report. Kathmandu, Nepal : Author.

Central Department of Botany. (1992) *Convention on Biological Diversity*. Kathmandu, Nepal: Author. Retrived from https://www.cbd.int/intro.

Chapagain, N.H., Pandit, R.K., & Tamang, R. (2016). *Flowering Plants of Makawanpur.* Makwanpur, Nepal: District Plant Resources Office.

Chalise, P., Paneru, Y.R., & Ghimire, S. K. (2018). Floristic diversity of vascular plants in Gyasumbdo valley, lower Manang, Central Nepal. *Journal of Plant Resources*, *17*(1), 42- 57.

Chaudhary, R.P. (1998). *Biodiversity in Nepal: Status and Conservation*. Bangkok, Thailand: Tecpress Books.

Dhakal, K.S., & Saud, D.S. (2018). CITES listed Plants of Nepal. *In:* GoN, *An Introduction to CITES Implementation in Nepal* (pp. 32- 54). Kathmandu, Nepal: Department of Forests.

Fraser-Jenkins, C.R., Kandel, D.R., & Pariyar S. (2015). *Ferns and Fern-allies of Nepal* (Vol - 1). Kathmandu, Nepal: National Herbarium and Plant Laboratories.

Grierson, A.J.C., & Long, D.G. (1983-2000). *Flora of Bhutan* (Vol. 1, Part 1-3; Vol. 2, Part 1-3). Edinburgh, UK: Royal Botanic Garden; Royal Government of Bhutan.

Joshi, N. (2014). *Utilization Pattern and Conservation Status of Plant Resources of Makawanpur District, Central Nepal*. (PhD thesis). Central Department of Botany, Tribhuvan University, Nepal.

Joshi, N., Dhakal, K.S.,& Saud, D. S. (2017). *Checklist of CITES Listed Flora of Nepal*. Kathmandu, Nepal: Department of Plant Resources.

Obha, H., Iokawa, Y., & Sharma, L.R. (2008). *Flora of Mustang,* Nepal. Kodansha Scientific Ltd., Tokyo.

Palmer, M.W., Wade, G.L.,& Neal, P. (1995). Standard for the Writing of Floras. *Bioscience*, *45*(5), 339-345.

Paudel, P.K., Bhattarai, B.P., & Kindlmann, P. (2011). An Overview of Biodiversity in Nepal. *In:* P. Kindlmann (Ed.), *Himalayan Biodiversity in the Changing World* (pp. 1-40). Dordrect, Netherlands: Springer.

Polunin, O., & Stainton, A. (1984). *Flowers of the Himalaya*. New Delhi, India: Oxford University Press.

Press, J.R., Shrestha K.K., & Sutton D.A. (2000). *Annotated Checklist of the Flowering Plants of Nepal*. London, UK: The Natural History Museum.

Roskov Y., Ower G., Orrell T., Nicolson D., Bailly N., Kirk P.M., Bourgoin T., DeWalt R.E., Decock W., Nieukerken E. van, Zarucchi J., & Penev L. (Eds.). (2019). *Species 2000 & ITIS Catalogue of Life, 2019 Annual Checklist*. Retrieved from www.catalogueoflife.org/annual-checklist/2019.

Sharma, U., Malla, K., & Uprety, R. (2017). Conservation and management efforts of medicinal and aromatic plants in Nepal. *BankoJanakari*, *14*(2), 3-11.

Shrestha, N., & Shrestha, K.K. (2012). Vulnerability assessment of high-valued medicinal Plants in Langtang National Park, Central Nepal. *Biodiversity*, *13*(1), 24-36.

Simpson, M.G. (2006). *Plant Systematics*. Bulington, VT: Elsevier Academy Press.

Stainton, A. (1988). *Flowers of the Himalaya: A supplement*. New Delhi, India: Oxford University Press.

Takhtajan, A. (1984). *Floristic Regions of the World; English translation*. Berkeley, CA: University of California Press.

Zheng-Yi, W, Raven, P.H., & Deyuan, H. (Eds.). (1996-2003). *Flora of China*, all volumes. Beijing, China: Science Press and St. Louis, USA: Missouri Botanical Garden Press.

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Floristic Diversity in a Community Managed Forest of Kanchanpur District, Western Nepal

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Abstract

The present study describes the floristic diversity of Janahit Mahakali community forest in Kanchanpur district, Western Nepal. The forest comprises an area of 198.93 hectares. Altogether, 148 plant species belonging to 123 genera under 59 families were recorded from 72 different sampling plots. Among the recorded species, the herbaceous species were quite higher than shrubs and trees. There were total 117 dicots, 23 monocots and 8 pteridophytes recorded. The study area was found to be dominated by Fabaceae with 13 genera and 19 species followed by Lamiaceae with 8 genera and 9 species. The frequency distribution of *Shorearobusta* and *Terminaliaalata* was found highest among all recorded species. Similarly, *Mazus pumilis* and *Phyllodium pulchellum* were among the most dominant herb and shrub.

Keywords: Dominant, Frequency distribution, Herbaceous, Janahit Mahakali, Plant composition

Introduction

Nepal is well known for its rich plant biodiversity in terms of its size. The country occupies about 0.1 percent of the global area but possesses over three percent of the world's known flora (Ministry of Forest and Soil Conservation [MoFSC], 2014). There are 35 forest types, 75 vegetation units and 118 ecosystems ((MoFSC, 2014).Forest covers a total of 5.96 million hectares which is 40.36% of total area of the country (Department of Forest Research and Survey [DFRS], 2010–2014). Tarai (low land) physiographic region of Nepal occupies 13.7% of the total land area of the country. Out of total area of forest 6.90% lies in Tarai (DFRS, 2010–2014).Tropical forests consists the most diverse plant communities on earth (Givnish, 1999; Anitha et al., 2010).Such forest is suitable habitat for trees, shrubs, herbs, climbers, ferns (Pathak & Baniya, 2017). Species diversity in the tropics varies from place to place. The biological resources of the Tarai are mostly dominated by Sal trees (*Shorea robusta*), tropical deciduous riverine forest and tropical evergreen forest. Sal forest is an identity of the lowland Tarai, associated with *Terminalia alata, Syzgium cumini, Adina cardifolia, Lagerstroemia parviflora*. These ecosystems are of international importance in terms of the number of globally threatened floral species found in them as well as their diversity (DFRS, 2010–2014).

Floristic study is a systematic botanical survey using vegetation plots which provide information on the patterns of plant diversity (Stohlgren et al., 1997). It is a substantial basis for syntaxonomical and phytoecological investigations required for taking conservation measure (Georgievia, 2013). Floristic analyses are very useful for identifying spatial patterns in plant diversity and composition (Slik et al., 2003). Knowledge of plant composition and diversity supports understanding of forest ecosystem dynamics and the utility of forest resources (Hartshorn, 1990). The rapid loss in floristic diversity and changing pattern of vegetation due to various biotic and abiotic factors have necessitated the qualitative and quantitative assessment of vegetation (Sharma et al., 2014). So the study of floristic diversity should attain importance as knowledge on floral diversity of a particular area can reflect the total resources, their use and conservation status which is very helpful for making conservation strategies and policies (Bhatta & Chaudhary, 2009).

Since, the botanical exploration in Nepal done by F. Buchanan Hamilton in 1802–1803 the major floristic enumerations have been focusing in Central and Eastern part of Nepal. Little attention has been given to the floristic study of Western Nepal (Shrestha et al., 2006). Compared to high altitudes, the flora of plains is still under explored (Chaudhary, 1998; Sah et al., 2002). The flora in far-western tarai is even very less surveyed because of its remoteness, relatively hot and dry climate (Sah et al., 2002). Few studies earlier conducted are limited to the documentation of ethno-medicinal knowledge and plant uses (Dhami 2008; Joshi & Singh, 2010; Pant &Yadav, 2013). Floristic study only focusing on the plant composition and diversity in the Western Nepal is still lacking. It has raised the fear that many species may go to be locally extinct prior to being reported as the component of Nepalese flora (Sah et al., 2002). Therefore, the present study has been undertaken to assess the plant diversity in one of the community forest of Kanchanpur district representing western Nepal.

Materials and Methods

Study area

The study was conducted in southwestern lowlands of Nepal, Kanchanpur district (28°32' – 29°8' N and 80°32' – 80°33' E) of Mahakali zone of Western Nepal which has the total area of 1,610 square kilometer (1,61,741 ha.). Topographically, the district is divided into three regions: Churia hills, Bhabar range and Tarai plain. The elevation ranges from 160 m to 1,528 m above sea level. The average annual rainfall of the district is 1,575 mm. The average maximum and minimum temperature is 43°C and 24°C during summer and 19°C and 2°C during winter (Joshi & Singh, 2010; Pant & Yadav, 2013). Janahit Mahakali community forest which extends up to the foothills of Siwalik in Krishnapur municipality with an area of 198.93 hectare was explored for its floral composition and diversity (Figure 1).

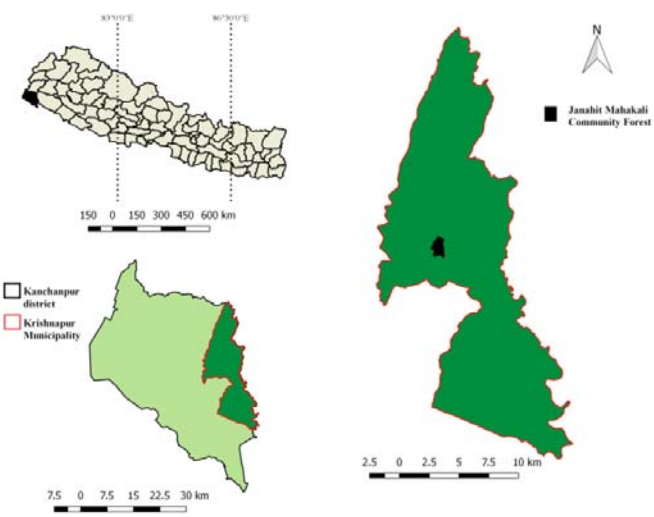

Figure 1: Map of the study area (Source: Department of Survey, GoN)

Data collection

The data collection was conducted during the month of November, 2018. The entire community forest was divided into six vertical transect lines considering the total forest area,each 1km long and 300 m apart for recording almost every plant occurring in the forest (Timilsina & Heinen, 2008; Webb & Sah, 2003). These transects covered whole forest area.On each transect, there were 6 sampling points, each at every 200m distance increment along the transect and each with paired quadrats spaced horizontally about 100 m apart (Swamy et al., 2000; Shrestha et al., 2007; Timilsina & Heinen, 2008). There were 12 plots of 10 m × 10 m size in each transect. A total of 72 sampling plots were employed for recording species presence data throughout the whole study area.

The species presence data were recorded and plant specimens were collected for theherbarium preparation following standard technique (Bridson & Forman, 1998). Plant species were identified as much as possible during field study and those which could not be identified in the field were later checked for identification with the help of relevant standard taxonomic literatures (Press et al., 2000; Rajbhandari et al., 2016), expertise consultation and tallying the specimens at National herbarium and Plant Laboratories, Godavari (KATH). The nomenclature and author citation of each species was validated by using different taxonomic literatures (Rajbhandari & Rai, 2017; Rajbhandari & Rai, 2019) and the online plant database, Catalogue of life (Roskov et al., 2019). All collected species were classified as dicot, monocot and Pteridophytes. Besides that, the dominant families and their broad life-form categories: trees, shrubs and herbs were also categorized. In addition, frequency of each individual species was calculated in order to find out their distribution in the study area (Singh et al., 2014).

$$\text{Frequency} = \frac{\text{Number of plots in which species occured}}{\text{Total number of plots studied}} \times 100\%$$

Results and Discussion

A total of 148 species were recorded within the 72 plots in the study area. Altogether, there were 59 families, 123 genera and 148 species (Figure 2). Among them, five foremost dominant familieswere Fabaceae (13 genera and 19 species) followed by Lamiaceae (8 genera and 9 species), Poaceae (7 genera and 8 species), Malvaceae (6 genera and 8 species) and Asteraceae (6 genera and 7 species) (Figure 3). Other families were Rubiaceae (6 genera and species each), Apocyanaceae (5 genera and species each), Acanthaceae (3 genera and 5 species) and Moraceae (2 genera and 5 species). Families with lesser number of species were represented by Amaranthaceae, Euphorbiaceae, Lythraceae, Anacardiaceae, Arecaceae and Asparagaceae (Table 1).

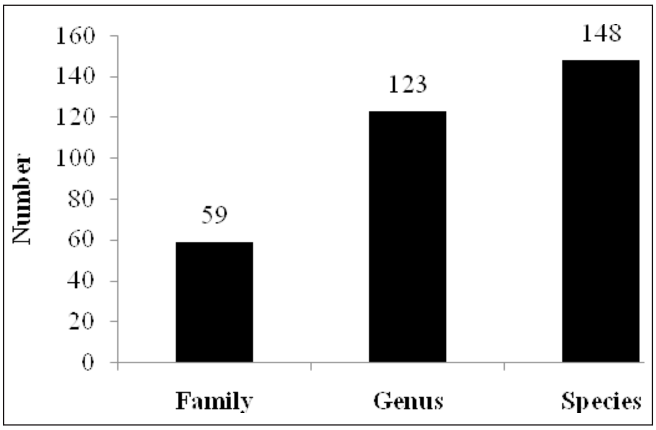

Figure 2: Distribution of families, genera and species recorded in the study plots

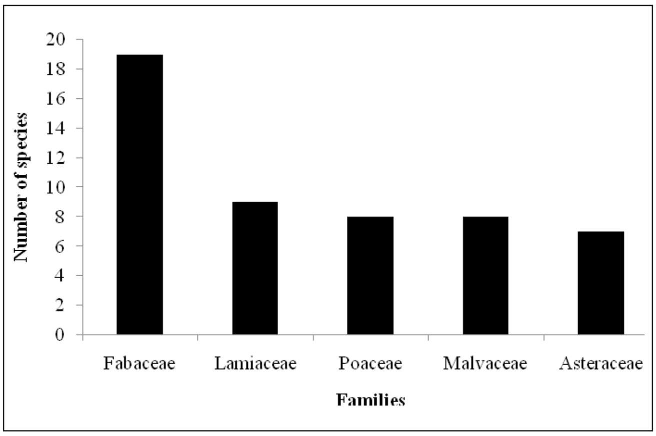

Figure 3: Families with highest species number

Based on the habit, dicots were dominating the study area (117 species), which was followed by the monocots (23 species) and Pteridophytes (8 species). Similarly, the life form categories showed the well representation of herbaceous flora (70 species) and less representation of shrubs (24 species), trees (36 species) and climber species (18 species).

Among the recorded species, *Shorea robusta* was found as the most frequently observed tree species (86.11% frequency), followed by *Terminalia alata* (73.61% frequency), *Lagerstroemia parviflora* (72.22% frequency), *Schleichera oleosa* (68.05% frequency) and *Litsea monopetala* (66.66% frequency). Similarly, *Phyllodium pulchellum* (65.27%) and *Woodfordia fruticosa* (63.88%) were dominant among shrubs, and *Mazus pumilis* (69.44%), *Achyranthes aspera* (62.5 %) were most dominant herbaceous species. In addition, *Milletia extensa* (73.61%), *Spatholobus parviflorus* (70.83%) and *Bauhinia vahlii* (61.11%) were frequently occurring climber species. Furthermore, *Lygodium flexuosum* (66.66% frequency) was the mostdominant Pteridophyte species. Among monocots, *Cynodon dactylon* (62.5%) was found as most dominant species (Table 2).

The present study suggested that the study area was rich in terms of flora comprising 148 plant species belonging to 59 different families. The study revealed that Fabaceae was the dominant family with 19 species which agreed the findings of Pathak & Baniya (2017) and Anbarashan et al. (2011). Dhami (2008) also revealed Fabaceae as most dominant family used by traditional healers for different medicinal purposes. This similarity may suggest that tropical lowland provides the suitable habitat for plants belonging to family Fabaceae which supports their dominancy. Further, herb was most dominant life form followed by tree and shrub. Similar kind of dominance was observed in a community managed tropical *Shorea robusta* forest in Nawalparasi, Nepal (Pathak & Baniya, 2017). The dicots were with highest number of species followed by monocots and pteridophytes which supported the dominance of dicots in the study area which is similar as observed by Vediya & Kharadi (2011). The study

Table 1: List of families with number of genera and species from 72 different sampling plots

S. N.	Families	Number of genera	Number of species
1	Fabaceae	13	19
2	Lamiaceae	8	9
3	Poaceae	7	8
4	Malvaceae	6	8
5	Asteraceae	6	7
6	Rubiaceae	6	6
7	Apocyanaceae	5	5
8	Orchidaceae	4	4
9	Convolvulaceae	3	4
10	Acanthaceae	3	5
11	Moraceae	2	5
12	Amarathaceae, Lythraceae and Rutaceae	3 each (9)	3 each (9)
13	Euphorbiaceae and Vitaceae	2 each (4)	3 each (6)
14	Menispermaceae, Ophioglossaceae, Primulaceae, Pteridaceae,	2 each (8)	2 each (8)
15	Combretaceae, Commelinaceae, Cyperaceae, Phyllanthaceae, Solanaceae and Zingiberaceae	1 each (6)	2 each (12)
16	Anacardiaceae, Arecaceae, Asparagaceae, Athyriaceae, Bixaceae, Costaceae, DioscoreaceaeDipterocarpaceae, Dryopteridaceae, Gentianaceae, Hypoxidaceae, Icacinaceae, Lauraceae, Lecythidaceae, Linderniaceae, Lygodiaceae, Mazaceae, Meliaceae, Myrtaceae, Nyctaginaceae, Onagraceae, Papaveraceae, Phrymaceae, Piperaceae, Plantaginaceae, Polygonaceae, Polygalaceae, Ranunculaceae, Rhamnaceae, Sapindaceae, Smilacaceae, Thelypteridaceae and Urticaceae.	1 each (33)	1 each (33)

also showed the highest frequency distribution of *Shorea robusta* which further indicated that *Shorea robusta* is the main component of vegetation composition of the lowland *Tarai*, as revealed by Bhadra et al. (2010) associated with *Terminalia alata*, *Lagerstroemia parviflora*, *Schleichera oleosa*, *Litsea monopetala*.

Conclusion

Thepresent work has documented the floristic composition and diversity of Janahit Mahakali Community Forest of Kanchanpur district, Western Nepal and helped to explore the flora and species diversity patterns of that particular area. The study had documented 148 plant species from that area with the dominancy of family Fabaceae and species like *Shorea robusta, Phyllodium pulchellum, Mazus pumilis*. The area was well represented by the herbaceous flora as compared to the shrub and tree species. The studyconcluded that more floristic studies are required to document the overall flora and patterns of species composition of Kanchanpur district as well as to deeply understand the availability of endemic and other endangered plant species in the Western lowlands of Nepal.

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References

Anbarashan, M., Parthasarthy, N., & Padmavathy, A. (2011). Ethno-floristic survey in sacred groves, Pudukottai district, Tamil Nadu-India. *Journal of Medicinal Plants Research*, *5*(3), 439-443.

Anitha, K., Joseph, S., Chandran, R. J., Ramasamy, E. V., & Prasad, S. N. (2010). Tree species diversity and community composition in a human-dominated tropical forest of Western Ghats biodiversity hotspot, India. *Ecological Complexity, 7*(2),217-224.

Bhadra, A. K., Dhal, N. K., Rout, N. C., & Reddy, V. R. (2010). Phytosociology of the tree community of Gandhamardan hill ranges. *Indian Forester*, 610-620.

Bhatta, K. P., & Chaudhary, R. P. (2009). Species diversity and distribution pattern of grassland and cultivated land species in upper Manang, Nepal Trans-Himalayas. *Scientific World, 7*(7), 76-79.

Bridson, D., & Forman, L., (1998). *The Herbarium Handbook* (pp. 333). United Kingdom: Royal Botanic Gardens, Kew.

Chaudhary, R. P. (1998). *Biodiversity in Nepal: status and conservation.* Saharanpur, India: S. Devi.

Department of Forest Research and Survey. (2015). *State of Nepal's Forests*. Forest Resource Assessment (FRA) Nepal, (DFRS). Kathmandu, Nepal: Author

Dhami, N. (2008). Ethnomedicinal uses of plants is Western Terai of Nepal: A case study of Dekhatbhuli VDC of Kanchanpur district. *Medicinal plants in Nepal: an anthology of contemporary research*, 165-177.

Georgieva, N., Pachedjieva, K., & Lyubenova, M. (2013). SCI "Zapadna Stara Planina and Predbalkan"–floristic studies on xerothermic oak forests. *Bulgarian Journal of Agricultural Science*, *19*(2), 218-221.

Givnish, T. J. (1999). On the causes of gradients in tropical tree diversity. *Journal of Ecology, 87*, 193-210.

Hartshorn, G. S. (1990). An overview of neotropical forest dynamics. *Four neotropical rainforests*, 585-599.

Joshi, N. R., & Singh, V. 2010. Non-timber forest products (NTFPs) used by Tharu tribe of Kanchanpur district of far-western Nepal. *New York Science Journal*, *3*(11), 111-119.

Ministry of Forest and Soil Conservation. (2014). *Nepal National Biodiversity Strategy & Action plan 2014-2020*. Kathmandu, Nepal: Author

Pant, G., & Yadav, R. K. P. (2013). Plant resource and utilization: a case study in Kanchanpur district, far-western Tarai, Nepal. *Ecoprint: An International Journal of Ecology, 20*, 89-95

Pathak, R. P., & Baniya, C. B. (2017). Species diversity and tree carbon stock pattern in a community-managed tropical shorea forest in Nawalparasi, Nepal. *International Journal of Ecology and Environmental Sciences, 42*(5), 3-17.

Press, J. R., Shrestha, K. K., & Sutton, D. A. (2000). *Annotated checklist of the Flowering Plants of Nepal*. London, UK: The Natural History Museum.

Rajbhandari, K. R., Thapa Magar, M. S., Kandel, D. R., & Khanal, C., (2016). *Plant Resources of Kailali, West Nepal.* 118p. Nepal: District Plant Resources Office, Kailali (pp. 118).

Rajbhandari, K. R., & Rai, S. K. (2017). *A Handbook of the Flowering Plants of Nepal*(pp. 656). Vol. 1. Kathmandu, Nepal: Department of Plant resources.

Rajbhandari, K. R., & Rai, S. K. (2019). *A Handbook of the Flowering Plants of Nepal* (pp. 527). Vol. 2. Kathmandu, Nepal: Department of Plant resources.

Roskov, Y., Ower, G., Orrell, T., Nicolson, D., Bailly, N., Kirk, P.M., Bourgoin, T., DeWalt, R. E., Decock, W., Nieukerken, E. van, Zarucchi J., & Penev L., eds. (2019). Species 2000 & ITIS Catalogue of Life, 2019 Annual Checklist. Digital resource at www.catalogueoflife.org/annual-checklist/2019. Species 2000: Naturalis, Leiden, the Netherlands. ISSN 2405-884X.

Sah, J. P., Singh, R. L., & Bhatta, N. (2002). Floristic diversity and use of plants in Ghodaghodi lake area, Nepal. *Journal of Natural History Museum, 21*(1-4), 243-66.

Sharma, P., Rana, J. C., Devi, U., Randhawa, S. S., & Kumar, R. (2014). Floristic diversity and distribution pattern of plant communities along altitudinal gradient in Sangla Valley, Northwest Himalaya. *The Scientific World Journal*, 1-12.

Shrestha, B. B., Ghimire, B., Lekhak, H. D., &Jha, P. K., (2007). Regeneration of Treeline Birch (*Betulautilis*D. Don) Forest in a Trans-Himalayan Dry Valley in Central Nepal. *Mountain Research and Development*, *27*, 259-267.

Shrestha, M. R., Rokaya, M. B., & Ghimire, S. K. (2006). A checklist of Trans-Himalayan dicot flora of Dolpo and its surrounding region in Northwest Nepal. *Scientific World*, *4*(4), 84-95.

Singh, J. S., Singh, S. P., & Gupta, S. R. (2014). *Ecology, Environmental Science & Conservation*. S. Chand Publishing.

Slik, J. W. F., Poulsen, A. D., Ashton, P. S., Cannon, C. H., Eichhorn, K. A. O., Kartawinata, K., & Payne, J. (2003). A floristic analysis of the lowland Dipterocarp forests of Borneo. *Journal of Biogeography*, *30*(10), 1517-1531.

Stohlgren, T. J., Chong, G. W., & Schell, L. D. (1997). Rapid assessment of plant diversity patterns: a methodology for landscapes. *Environmental Monitoring and Assessment*, *48*(1), 25-43.

Swamy, P. S., Sundarapandian, S. M., Chandrasekar, P., & Chandrasekaran, S. (2000). Plant species diversity and tree population structure of a humid tropical forest in Tamil Nadu, India. *Biodiversity & Conservation*, *9*(12), 1643-1669.

Timilsina, N., & Heinen, J. T. (2008). Forest structure under different management regimes in the western lowlands of Nepal. *Journal of Sustainable Forestry*, *26*(2), 112-131

Vediya, S. D., & Kharadi, H. S. (2011). Floristic diversity of Isari zone, Megharj range forest District Sabarkantha, Gujarat, India. *International Journal of Pharmacy & Life Sciences*, *2*(9), 1033-1034.

Webb, E. L., & Sah, R. N. (2003). Structure and diversity of natural and managed sal (Shorearobusta Gaertn. f.) forest in the Terai of Nepal. *Forest Ecology and Management*, *176*(1-3), 337-353.

Photoplate: Photographs of some plant species of the study area.

Solanum erianthum D. Don

Flemengia macrophylla (Willd.) Merr.

Zingiber chrysanthum Roscoe

Crotolaria albida Roth

Poranopsis paniculata (Roxb.) Roberty

Acilepis squarrosa D. Don

Table 2: List of recorded plant species with their family, habit, life form and frequency

S. N.	Plant species	Family	Habit	Life form	Frequency (%)
1	*Abrus precatorius* L.	Fabaceae	Climber	Dicot	40.27
2	*Achyranthes aspera* L.	Amaranthaceae	Herb	Dicot	62.5
3	*Acilepis squarrosa* D. Don	Asteraceae	Herb	Dicot	13.88
4	*Acmella calva* (DC.) R.K.Jansen	Asteraceae	Herb	Dicot	29.16
5	*Acmella paniculata* (Wall. ex DC.) R.K. Jansen	Asteraceae	Herb	Dicot	23.61
6	*Adiantum incisum* Forsk.	Pteridaceae	Herb	Pteridophyte	11.11
7	*Adina cordifolia (*Roxb.) Benth. & Hook. f. ex B.D. Jacks.	Rubiaceae	Tree	Dicot	29.166
8	*Aegle marmelos* (L.) Correa	Rutaceae	Tree	Dicot	20.83
9	*Aerva lanata* (L.) Juss.	Amaranthaceae	Herb	Dicot	51.38
10	*Ageratum houstonianum* Mill.	Asteraceae	Herb	Dicot	18.05
11	*Alstonia scholaris* (L.) R. Br.	Apocynaceae	Tree	Dicot	22.22
12	*Alternanthera sessilis* (L.) R.Br. ex DC.	Amaranthaceae	Herb	Dicot	30.55
13	*Ammannia auriculata* Willd.	Lythraceae	Herb	Dicot	23.61
14	*Ampelocissus latifolia* (Roxb.) Planch.	Vitaceae	Climber	Dicot	11.11
15	*Ampelopteris prolifera* (Retz.) Copel.	Thelypteridaceae	Herb	Pteridophyte	27.77
16	*Anisomeles indica* (L.) Kuntze	Lamiaceae	Herb	Dicot	23.61
17	*Ardisia elliptica* Thunb.	Primulaceae	Tree	Dicot	4.16
18	*Argemone maxicana* L.	Papaveraceae	Herb	Dicot	11.11
19	*Artocarpus lacucha* Buch.-Ham. ex D. Don	Moraceaea	Tree	Dicot	9.72
20	*Asparagus racemosus* Willd.	Asparagaceae	Climber	Dicot	27.77
21	*Barleria cristata* L.	Acanthaceae	Herb	Dicot	33.33
22	*Bauhinia vahlii* Wight and Arn.	Fabaceae	Woody climber	Dicot	61.11
23	*Bixa orellana* L.	Bixaceae	Tree	Dicot	11.11
24	*Boehmeria virgata* (G.Forst.) Guill.var. *canescens* (Wedd.) Friis & Wilmot-Dear	Urticaceae	Shrub	Dicot	8.33
25	*Boerhaavia diffusa* L.	Nyctaginaceae	Herb	Dicot	31.94
26	*Bombax ceiba* L.	Malvaceae	Tree	Dicot	5.55
27	*Calamus tenuis* Roxb.	Arecaceae	Tree	Monocot	12.5
28	*Calotropis gigantea* (L.) W. T. Aiton	Apocynaceae	Shrub	Dicot	13.88
29	*Canscora alata* (Roth) Wall.	Gentianaceae	Herb	Dicot	45.83
30	*Capillipedium assimile* (Steud.) A.Camus	Poaceae	Herb	Monocot	18.05
31	*Careya arborea* Roxb.	Lecythidaceae	Tree	Dicot	12.5
32	*Cassia fistula* L.	Fabaceae	Tree	Dicot	20.83
33	*Catunaregam spinosa* (Thunb.) Tirveng.	Rubiaceae	Shrub	Dicot	59.72
34	*Ceratopteris thalictroides* (L.) Brongn.	Pteridaceae	Herb	Pteridophyte	41.66
35	*Clausena heptaphylla* (Roxb.) Wight & Arn.	Rutaceae	Tree	Dicot	50
36	*Clematis zeylanica* Poir.	Ranunculaceae	Climber	Dicot	22.22
37	*Clerodendrum indicum* (L.) Kuntze	Lamiaceae	Shrub	Dicot	23.61
38	*Clerodendrum infortunatum* L.	Lamiaceae	Shrub	Dicot	34.72
39	*Colebrookea oppositifolia* Sm.	Lamiaceae	Shrub	Dicot	22.22
40	*Corchorus aestuans* L.	Malvaceae	Herb	Dicot	51.38
41	*Crotalaria albida* Roth	Fabaceae	Shrub	Dicot	50
42	*Crotolaria prostrata* Rottb. ex Willd.	Fabaceae	Herb	Dicot	34.72
43	*Curculigo orchioides* Gaertn.	Hypoxidaceae	Herb	Monocot	23.61

S. N.	Plant species	Family	Habit	Life form	Frequency (%)
44	*Cyanotis axillaris* (L.) D.Don ex Sweet	Commelinaceae	Herb	Monocot	29.16
45	*Cyanotis cristata* (L.) D.Don	Commelinaceae	Herb	Monocot	37.5
46	*Cynodon dactylon* (L.) Pers.	Poaceae	Herb	Monocot	62.5
47	*Cynodon radiatus* Roth	Poaceae	Herb	Monocot	43.05
48	*Cyperus mindorensis* (Steud.) Huygh	Cyperaceae	Herb	Monocot	30.55
49	*Cyperus rotundus* L.	Cyperaceae	Herb	Monocot	13.88
50	*Cyrtococcum patens* (L.) A. Camus var. *latifolium* (Honda) Ohwi	Poaceae	Herb	Monocot	37.5
51	*Dalbergia latifolia* Roxb.	Fabaceae	Tree	Dicot	4.16
52	*Desmodium gangeticum* (L.) DC.	Fabaceae	Shrub	Dicot	55.55
53	*Desmodium oojeinense* (Roxb.)H.Ohashi	Fabaceae	Tree	Dicot	40.27
54	*Desmodium triflorum* (L.)DC.	Fabaceae	Herb	Dicot	30.55
55	*Digitaria ciliaris* (Retz.) Koeler	Poaceae	Herb	Monocot	23.61
56	*Dioscorea belophylla* (Prain) Voigt ex Haines	Dioscoreaceae	Climber	Monocot	29.16
57	*Diplazium esculentum* (Retz.) Sw.	Athyriaceae	Herb	Pteridophyte	11.11
58	*Dryopteris cochleata* (D. Don) C. Chr.	Dryopteridaceae	Herb	Pteridophyte	9.72
59	*Eclipta prostrata* (L.) L.	Asteraceae	Herb	Dicot	38.88
60	*Elephantopus scaber* L.	Asteraceae	Herb	Dicot	30.55
61	*Emilia sonchifolia* (L.) DC. ex Wight	Asteraceae	Herb	Dicot	8.33
62	*Erythranthe tenella* (Bunge) G. L. Nesom	Phrymaceae	Herb	Dicot	36.11
63	*Eulalia mollis* (Griseb.) Kuntze	Poaceae	Herb	Monocot	31.94
64	*Euphorbia hirta* L.	Euphorbiaceae	Herb	Dicot	27.77
65	*Evolvulus nummularius* (L.) L.	Convolvulaceae	Herb	Dicot	50
66	*Ficus benghalensis* L.	Moraceae	Tree	Dicot	12.5
67	*Ficus palmata* Forssk.	Moraceae	Tree	Dicot	8.33
68	*Ficus religiosa* L.	Moraceae	Tree	Dicot	5.55
69	*Ficus rumphii* Blume	Moraceae	Tree	Dicot	31.94
70	*Flemingia macrophylla* (Willd.) Merr.	Fabaceae	Shrub	Dicot	33.33
71	*Flemingia strobilifera* (L.) W.T. Aiton	Fabaceae	Shrub	Dicot	50
72	*Grewia eriocarpa* Juss.	Malvaceae	Tree	Dicot	9.72
73	*Habenaria stenopetala* Lindl.	Orchidaceae	Herb	Monocot	1.38
74	*Hellenia speciosa* (J.Koenig) S.R. Dutta	Costaceae	Shrub	Monocot	12.5
75	*Helminthostachys zeylanica* (L.) Hook.	Ophioglossaceae	Herb	Pteridophyte	30.55
76	*Hemigraphis hirta* (Vahl) T.Anderson.	Acanthaceae	Herb	Dicot	27.77
77	*Holarrhena pubescens* Wall. ex G. Don	Apocynaceae	Tree	Dicot	50
78	*Hymenodictyon orixense* (Roxb.) Mabb.	Rubiaceae	Tree	Dicot	20.83
79	*Ichnocarpus frutescens* (L.) W. T.Aiton	Apocynaceae	Woody climber	Dicot	52.77
80	*Imperata cylindrica* (L.) P. Raeusch.	Poaceae	Herb	Monocot	37.5
81	*Ipomoea muricata* (L.) Jacq.	Convolvulaceae	Climber	Dicot	18.05
82	*Ipomoea quamoclit* L.	Convolvulaceae	Climber	Dicot	43.05
83	*Justicia diffusa* Willd.	Acanthaceae	Herb	Dicot	31.94
84	*Justicia gendarussa* Burm. f.	Acanthaceae	Herb	Dicot	5.55
85	*Justicia pectinata* L.	Acanthaceae	Herb	Dicot	23.61
86	*Knoxia sumatrensis* (Retz.) DC.	Rubiaceae	Herb	Dicot	56.94
87	*Lagerstroemia parviflora* Roxb.	Lythraceae	Tree	Dicot	72.22
88	*Leea aequata* L.	Vitaceae	Shrub	Dicot	22.22

S. N.	Plant species	Family	Habit	Life form	Frequency (%)
89	*Leea asiatica* (L.) Ridsdale	Vitaceae	Shrub	Dicot	31.94
90	*Leucas cephalotes* (Roth) Spreng.	Lamiaceae	Herb	Dicot	16.66
91	*Leucosceptrum canum* Sm.	Lamiaceae	Tree	Dicot	33.33
92	*Litsea monopetala* (Roxb.) Pers.	Lauraceae	Tree	Dicot	66.66
93	*Ludwigia hyssopifolia* (G. Don) Exell	Onagraceae	Herb	Dicot	45.83
94	*Luisia tristis* (G.Forst.) Hook.f.	Orchidaceae	Herb	Monocot	18.05
95	*Lygodium flexuosum* (L.) Sw.	Lygodiaceae	Climber	Pteridophyte	66.66
96	*Lysimachia arvensis* (L.) U. Manns&Anderb.	Primulaceae	Herb	Dicot	16.66
97	*Mallotus nudiflorus* (L.) Kulju&Welzen	Euphorbiaceae	Tree	Dicot	31.94
98	*Mallotus philippensis* (Lam.) Müll.Arg.	Euphorbiaceae	Tree	Dicot	48.61
99	*Mazus pumilus* (Burm. f.) Steenis	Mazaceae	Herb	Dicot	69.44
100	*Melia azedarach* L.	Meliaceae	Tree	Dicot	20.83
101	*Mesosphaerum suaveolens* (L.) Kuntze	Lamiaceae	Shrub	Dicot	36.11
102	*Millettia extensa* (Benth.)Baker	Fabaceae	Woody climber	Dicot	73.61
103	*Mitragyna parvifolia* (Roxb.) Korth.	Rubiaceae	Tree	Dicot	20.83
104	*Murraya koenigii* (L.) Spreng.	Rutaceae	Tree	Dicot	36.11
105	*Natsiatum herpeticum* Buch.-Ham. exArn.	Icacinaceae	Climber	Dicot	9.72
106	*Ophioglossum reticulatum* L.	Ophioglossaceae	Herb	Pteridophyte	40.27
107	*Oplismenus compositus* (L.) P.Beauv.	Poaceae	Herb	Monocot	20.83
108	*Pelatantheria insectifera* (Rchb.f.) Ridl.	Orchidaceae	Herb	Monocot	25
109	*Persicaria barbata* (L.) H.Hara	Polygonaceae	Herb	Dicot	38.88
110	*Phyllanthus emblica* L.	Phyllanthaceae	Tree	Dicot	11.11
111	*Phyllanthus urinaria* L.	Phyllanthaceae	Herb	Dicot	43.05
112	*Phyllodium pulchellum* (L.) Desv.	Fabaceae	Shrub	Dicot	65.27
113	*Piper longum* L.	Piperaceae	Herb	Dicot	40.27
114	*Platostoma hispidum* (L.) A.J.Paton	Lamiaceae	Herb	Dicot	22.22
115	*Pogostemon benghalensis* (Burm.f.) Kuntze	Lamiaceae	Shrub	Dicot	26.38
116	*Polygala crotalarioides* Buch.-Ham. ex DC.	Polygalaceae	Herb	Dicot	37.5
117	*Poranopsis paniculata* (Roxb.) Roberty	Convolvulaceae	Climber	Dicot	25
118	*Rauvolfia serpentina* (L.) Benth. ex Kurz	Apocynaceae	Herb	Dicot	23.61
119	*Schleichera oleosa* (Lour.) Merr.	Sapindaceae	Tree	Dicot	68.05
120	*Scleromitrion diffusum* (Willd.) R.J.Wang	Rubiaceae	Herb	Dicot	15.27
121	*Scoparia dulcis* L.	Plantaginaceae	Herb	Dicot	33.33
122	*Semecarpus anacardium* L. f.	Anacardiaceae	Tree	Dicot	4.16
123	*Senegalia catechu* (L.f.) P.J.H. Hurter & Mabb.	Fabaceae	Tree	Dicot	25
124	*Senegalia rugata* (Lam.) Britton & Rose	Fabaceae	Woody climber	Dicot	34.72
125	*Senna occidentalis* (L.)Link	Fabaceae	Shrub	Dicot	22.22
126	*Senna tora* (L.) Roxb.	Fabaceae	Herb	Dicot	36.11
127	*Shorea robusta* Gaertn.	Dipterocarpaceae	Tree	Dicot	86.11
128	*Sida acuta* Burm. f.	Malvaceae	Subshrub	Dicot	52.77
129	*Sida cordata* (Burm. f.) Borss.Waalk.	Malvaceae	Herb	Dicot	22.22
130	*Sida cordifolia* L.	Malvaceae	Subshrub	Dicot	30.55
131	*Smilax ovalifolia* Roxb. ex D.Don	Smilacaceae	Climber	Monocot	16.66
132	*Solanum erianthum* D. Don	Solanaceae	Shrub	Dicot	11.11
133	*Solanum virginianum* L.	Solanaceae	Herb	Dicot	13.88
134	*Spatholobus parviflorus* (DC.)Kuntze	Fabaceae	Woody climber	Dicot	70.83

S. N.	Plant species	Family	Habit	Life form	Frequency (%)
135	*Stephania pierrei* Diels	Menispermacxweae	Climber	Dicot	27.77
136	*Syzygium cumini* (L.) Skeels	Myrtaceae	Tree	Dicot	19.44
137	*Terminalia alata* Heyne ex Roth	Combretaceae	Tree	Dicot	73.61
138	*Terminalia bellirica* (Gaertn.) Roxb.	Combretaceae	Tree	Dicot	15.27
139	*Tinospora sinensis* (Lour.) Merr.	Menispermaceae	Climber	Dicot	12.5
140	*Triumfetta rhomboidea* Jacq.	Malvaceae	Subshrub	Dicot	45.83
141	*Uraria lagopodioides* (L.) DC.	Fabaceae	Herb	Dicot	41.66
142	*Urena lobata* L.	Malvaceae	Subshrub	Dicot	52.77
143	*Vanda tessellata* (Roxb.) Hook. ex G.Don	Orchidaceae	Herb	Monocot	38.88
144	*Vandellia anagallis* (Burm. f.) T. Yamaz.	Linderniaceae	Herb	Dicot	25
145	*Woodfordia fruticosa* (L.) Kurz	Lythraceae	Large shrub	Dicot	63.88
146	*Zingiber capitatum* Roxb.	Zingiberaceae	Herb	Monocot	29.16
147	*Zingiber chrysanthum* Roscoe	Zingiberaceae	Herb	Monocot	59.72
148	*Zizyphus mauritiana* Lam.	Rhamnaceae	Shrub	Dicot	36.11

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Wetland Plants and their Ethnobotanical Uses in Raja-Rani Tal, Letang, Morang, Nepal

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Abstract

Wetland plants play vital role in ecosystems and also provide various basic needs such as food, medicine, fodder, green manure, raw materials etc. A total of 108 plant species were documented from the wetland and periphery of Raja-Rani Tal, Morang in April and December 2018.We documented angiosperms (Dicots 48, Monocots 32), Pteridophytes 10, Bryophytes 1, and algae 17 species, belonging to 91 genera and 56 families. Among the total species, 70 species are hydrophytes including algae having the different growth form likeEmergent (43) > Floating (5) > Floating-Leaved (3) > Submerged (2)while 43 species are found around the periphery of wetland. There were 77 species native including one endemic flowering plant species, and 12 species exotic including 6 invasive alien species. These wetland plants were used by local people for fodder (31), medicine (7), fermenting agent (3), vegetable (3), beekeeping (2), compost (1), fish poison (1), ripening (1), cultural use.

Keywords: Exotic, Growth form, Macrophytes, Native, Use

Introduction

Wetland plants are those plants that normally grow in or on the water or where soils are flooded or saturated long enough for anaerobic condition to develop in the root zone (Cronk & Fennessy, 2009). They are also known as hydrophytes. Thousands of plant species grow in wetlands, ranging from algae, mosses and grass to shrub and trees. The richness of plant species in aquatic and wetland habitats is relatively low compared with most terrestrial communities (Richardson & Vymazal, 2001). Most are rooted, but a few species float freely in the water. There are 318 wetland dependent plant species in Tairairegion of Nepal which include 6 climbers, 287 herb, 9 shrub and 16 tree species (Siwakoti, 2006) Wetland plants play several vital roles in wetland ecosystems as they are the base of food chain, and providecritical habitat for other taxonomic groups, such as bacteria, epiphyton (algae that grow on the surface of plants), macro invertebrates, fish, and birds. Beside these roles, wetland plants also provide various basic needs such as food, medicine, fodder, green manure, raw materials etc. (Siwakoti, 2006; Dangol, 2014).

Wetlands are considered to be one of the most threatened of all major natural ecosystems. Similarly, the ethnobotanical knowledge and practices are also in danger of being lost in country.Therefore, effort should be made to document the wetland plants and their ethnobotanical uses before the extinction of these useful plants and their associated knowledge. Some studies on documentation of wetland flora and their use have been conducted in some wetlands of Morang districts (Niroula & Singh, 2010; Koirala & Jha, 2011).

The Raja- Rani Tal is a very important wetland in the Chure region of the eastern Nepal due to its biodiversity, environmental and cultural values. It was recommended to explore the possibility to designate the wetland as a Ramsar Site and found two Ramsar criteria qualifying to this wetland (Siwakoti et al., 2012). Enumeration of orchid species and algal flora (Chlorophyceae) species has been done in Raja-Rani wetland. Shakya & Bajracharya (2005) recorded 45 orchid species from Raja-Rani wetland with most of the species in good number, whereas,Godar & Rai (2018) reported 72 freshwater chlorophycean algae belonging to 33 genera, 12 families and 7 orders from Raja-Rani wetland. We documented few algal species, bryophytes, pteridophytes and angiosperm from the Raja-Rani wetland and their ethnobotanical uses from the Magar community residing in Letang village.

Materials and Methods

Study area

Raja-Rani wetland (26°44'53"N and 87°28'54"E, 467 m asl) situated along the Chure hill range in Letang Municipality-1, Morang district, Nepal (Figure 1). It is located in north-west from Letangat Dhimal Danda surrounded by mixed forest of *Shorea robusta*, *Cassia fistulosa*, *Syzygium cuminii* etc. The wetland cover three ponds named as Raja, Rani and Chori. The largest one is Raja Pokhari which is located in the western side having the depth of 7 m. in rainy and 3 m. in winter and 2 m. in summer season.The Rani Pokhari located in eastern side and Chori in north eastern side which is almost dried in condition. Nearby settlement of wetland is dominated by Magar community. This wetland has been managed by Raja Rani Community Forest User's Group. The three lakes cover about 20 ha of land along the catchment area of about 133 ha. It feeds by ground source "Jaruwa", seasonal forest fed streams and direct precipitation (Godar & Rai, 2018). Some Tree species in surroundings of Raja Rani wetland are *Albizia* sp., *Alstonia scholaris, Castanopsis indica,Cassia fistula, Mallotus phillipensis, Schima wallichii, Shorea robusta, Syzygium cuminii, Sterculia* sp., *Trewia nudiflora* etc.

Field visit and data collection

Two field trips were done in April and December 2018 for the study. The hydrophytes specimens and their photographs were collected from wetland as well as other macrophytes from the surroundings up to 5 m from the periphery of the wetland. Some plants were identified in field and unidentified plants were collected, prepared herbarium and identified by comparing it with deposited herbarium at National Herbarium and Plant Laboratories (KATH), Godawari, Lalitpur, Nepal and housed at KATH. http://www.theplantlist.org was followed for nomenclature. On the basis of their growth form, hydrophytes excluding algae are classified as submerged, floating, floating-leaved and emergent (Materac & Sobiecka, 2017).

1) Submerged: These plants usually grow under water and are rooted in mud.
2) Floating-Leaved: Their leaves are floating on the water surface, and they contain anchoring roots on the surface of soil.
3) Floating: The stem and leaves of these plants float, they submerged in water without any attachment to the sediment.
4) Emergent: These plants spread their shoots and leaves above the water, while keeping their roots beneath the surface.

Other angiospermic plants collected from periphery of the wetland are classified as tree, shrub, herb, climber, creeper. Similarly, pteridophytes are classified asepiphytic, terrestrial and creeping.

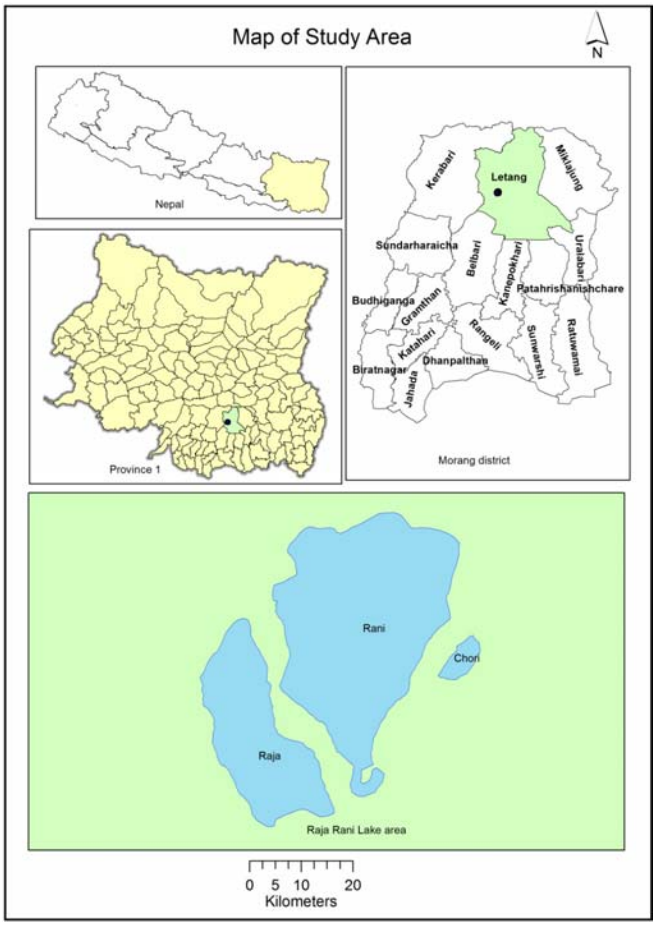

Figure1: Map of study sites.

Plant categorization

Plants were classified into the categories of native, exotic on the basis of its origin by consulting Global Register of Introduced and Invasive Species-Nepal (Shrestha et al., 2019) and for invasive plant (Shrestha et al., 2017).

Ethnobotanical data collection

Ethnobotanical data of wetland plants were collected from the Magar community residing nearby the wetland. Questionnaires were prepared for interviews and informants were chosen randomly. Prior to interview, the purpose of research background was explained to each informant. Ethnobotanical data of wetland plants were collected through interview with local people and key informants. During the interview, collected plants or photographs were shown.

Results and Discussion

Species diversity

Altogether 108 plant species were documented from the wetland and around the periphery of wetland comprising angiosperms (Dicots 48, Monocots 32), Pteridophytes 10, Bryophytes 1, and algae 17, belonging to 91 genera and 56 families (Table 1 and Figure 2). Among the total species, 70 species are hydrophytes including algae while 43 species are found around the periphery of wetland. 77 species were native including one endemic flowering plant species, 12 species were exotic including six invasive alien species. Koirala & Jha (2011) had reported 149 species of macrophytes from the low land wetlands of Morang which compromise Angiosperms (dicots 60, monocots 78), Pteridophytes 7, Bryophytes 2, and algae 2. Similarly, Niroula & Singh (2011) reported 84 aquatic plant species belonging to 71 genera and 41 families from the Betana wetland, Morang. This show high diversity of macrophytes in Raja-Rani wetland. One of the endemic species (*Eriocaulon exsertum)* has been reported from this wetland.

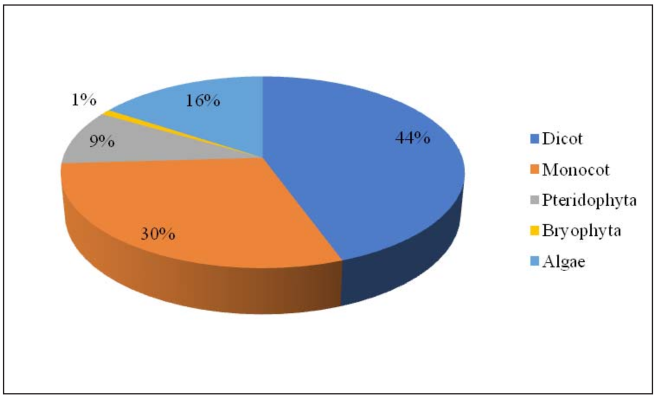

Figure 2: Species diversity in different taxonomic groups

Growth form

The growth forms of the hydrophytes excluding algae recorded in the present study were Emergent (43) >Floating (5) >Floating-Leaved (3) > Submerged (2) (Figure 3). Similarly the growth form of Angiospermic plant around the periphery of wetland were Shrub (14) > Herb (10) >Climber (6) > Tree (2) like-wise the growth form of pteridophytes were Terrestrial (3) >Epiphytic (1),Creeping (1) and Climber (1). As in findings of Burlakoti & Karmacharya (2004) and Dangol et al. (2014) the emergent species were higher in number which is followed by Floating, Floating –leaved and Submerged. The aquatic tree species *Cephalanthus tetrandra* has covered about one third of Rani Tal also in some portion in Raja Tal. Higher the number and dominance of emergent species may be the indicator of the wetland sedimentation reducing the core area of the lake and showing the trend of succession towards marsh meadow condition.

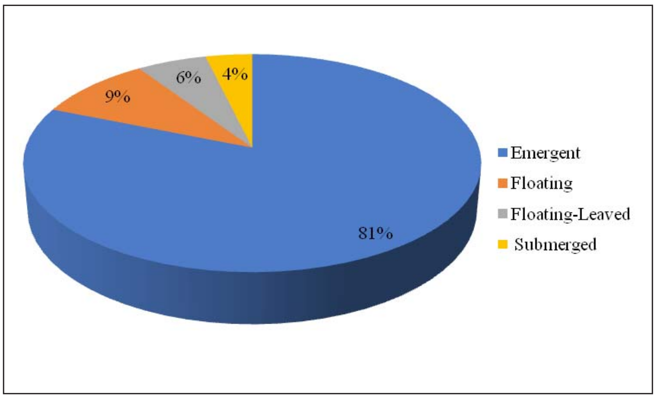

Figure 3: Growth forms of the hydrophytes

Ethnobotanical use

The local Magar communities residing near the wetland interviewed about the use of wetland plants.

They use 31 dicot plants and 16 monocot plants for different purpose like for Fodder (31), Medicine (7), Fermenting agent (3), Vegetable (3), Beekeeping (2), Compost (1), Fish poison (1), Ripening (1), Cultural use (1), Local tools (1). They use stem of 34 species, leaves of 33 species, Root of 6 species, Flower of 4 species, Fruit of 3 species, latex of 2 species, tuber of 1 species and young shoot of 2 species and whole plants of 1 species.

Conclusion

The presence of wide diversity and locally important plants, as well as endemic plants in the wetland, reveal that the wetland is ecologically important. The intrusion of invasive species like *Eichhornia crassipes* and *Pistia stratiotes* threatening the wetland plants. The study had recorded one endemic species in small patch.The conservation of this plant is our duty. So for the conservation of such important species the local community should be made awarefor restoration and sustainable conservation of wetlandto reduce the eutrophication and succession rate.

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References

Basnet, B. (2013). Documentation of flora of Rara lake and adjoining areas in mid-western region of Nepal. *BankoJanakari,21*(1), 41-47.

Burlakoti, C. & Karmacharya, S. B. (2004). Quantitative analysis of macrophytes of Beeshazar Tal, Chitwan, Nepal. *Himalayan Journal of Sciences,2*(3), 37-41.

Dangol, D. R., Gautam, B. & Oli, B. B. (2018).Wetland Plants and their local use: Observation from Rampur Ghol, Chitwan, Nepal. *Nat. Hist. Mus*., *28*,142-159.

Godar, K. & Rai, S.K. (2018). Freshwater Green Algae from Raja-Rani Wetland, Bhogateni-Letang, Morang, Nepal. *Journal of Plant Resoureces*. *16*(1), 1-17.

Cronk J.K. & Fennessy M.S. . (2009). Encyclopedia of Inland Waters.

Koirala, U. & Jha, S. (2011). Macrophytes of the lowland wetland in Morang district, Nepal. *Nepalese Journal of Bioscience, 1*, 131-139.

Materac, M. & Sobiecka, E. (2017). The efficiency of macrophytes for heavy metals removal from water. *Biotechnol. Food Sci.*, 81 (1), 35-40.

Niroula, B. & Singh, K.L.B. (2010).Contribution to aquatic macrophytes of Biratnagar and adjoining areas, eastern Nepal. *Ecoprint*, *17*, 23-34.

Niroula, B. & Singh, K.L.B. (2011).Aquatic Plant Resources of Betana Wetland, Morang Nepal. *Our Nature*, 9, 146-155.

Press, J. R., Shrestha, K. K. & Sutton, D. A. (2000). *Annotated checklist of the flowering plants of Nepal*, London: The Natural History Museum.

Richardson, C.J., & J. Vymazal. (2001). Sampling macrophytes in wetlands. Pp. 297-338 (Chapter 14) in Rader, R., D.P. Butzer, and Scott A. Wissinger (eds.). *Bioassessment and Management of North American Freshwater Wetlands* (pp. 469). New York: John Wiley & Sons.

Shakya, L. R., & Bajracharya, D.M. (2005). Orchid sanctuary Raja Rani (Morang District), East Nepal: an effort toward habitat conservation. *Selbyana*, *26*, 236-239.

Siwakoti, M. (2006). An overview of floral diversity in wetlands of Terai region of Nepal. *Our Nature, 4*, 83-90.

Siwakoti, M., Sherchan, R., Basnet, B.T. & Sharma, N.L. (2012). *Report on Wetlands Inventory of Chure Region (East of Narayani)*. Kathmandu:

The Rastrapati Chure Conservation Program Coordination Unit.

Shrestha, B.B, Budha, P.B, Wong, L.J.& Pagad, S. (2019). *Global Register of Introduced and Invasive Species*- Nepal. Version 2.4. Invasive Species Specialist Group ISSG. Checklist dataset https://doi.org/10.15468/4r0kkr accessed via GBIF.org on 2020-01-12.

Shrestha, B.B., Siwakoti, M. & Ranjit, J.D. (2017). Status of Invasive alien Plant Species in Nepal. Dhulikhel: *Conservation and Utilization of Agriculural Plant Genetic Resources in Nepal. 2nd National Workshop* 22-23 May 2017. Proceeding.

Table 1: Wetland Plants and their ethnobotanical uses

S. N.	Family	Scientific Name	Growth Form	Parts Used	Uses	Collection No.	IUCN Conservation Status	Native/ exotic
	Algae							
1	Desmidiaceae	*Closterium dianae* Ehrenberg ex Ralfs						
2	Desmidiaceae	*Closterium gracile* Brébisson ex Ralfs						
3	Desmidiaceae	*Cosmarium pyramidatum* Brébisson ex Ralfs						
4	Desmidiaceae	*Cosmarium* sp.						
5	Desmidiaceae	*Cosmarium speciosum* P.Lundell						
6	Desmidiaceae	*Euastrum turneri* West						
7	Desmidiaceae	*Micrasterias foliacea* Bailey ex Ralfs						
8	Desmidiaceae	*Pleurotaenium baculoides* (J.Roy & Bisset) Playfair						
9	Desmidiaceae	*Staurastrum* cf *margaritaceum* Meneghini ex Ralfs						
10	Desmidiaceae	*Staurastrum* sp.						
11	Euglenidae	*Euglena gracilis* G.A.Klebs						
12	Nostocaceae	*Anabaena* sp.						
13	Oscillatoriaceae	*Oscillatoria* sp.						
14	Oscillatoriaceae	*Phormidium* sp.						
15	Phacidae	*Phacus* sp.						
16	Scenedesmaceae	*Scenedesmus acutiformis* Schröder						
17	Spirulinaceae	*Spirulina* sp.						
	Bryophyta							
1	Ricciaceae	*Riccia fluitans* L.	F				LC	Na
	Pteridophytes							
1	Pteridaceae	*Onychium siliculosum* (Desv.) C. Chr.	Terrestrial			RJ0054		Na
2	Pteridaceae	*Pteris biaurita* L.	Terrestrial	Rhizome	Paste is applied in sprain and fracture	RJ0042/L2		Na
3	Thelypteridaceae	*Cyclosorus interruptus* (Willd.) H. Ito	E			RN034	LC	Na
4	Pteridaceae	*Adiantum lunulatum* Burm. f.	E			RN013		Na
5	Dennstaedtiaceae	*Microlepia setosa* Alston	E	Leaves	Used as cattle bed	RJ0041		Na
6	Dennstaedtiaceae	*Microlepia speluncae* (L.) T. Moore	Terrestrial			RJ0047/L38		Na
7	Lycopodiaceae	*Lycopodiella cernua* (L.) Pic. Serm.	Creeping			RJ0034/L12	LC	Na
8	Lygodiaceae	*Lygodium flexuosum* (L.) Sw.	Climber	Rhizome	Paste is applied on wound.	RJ0043		Na
9	Polypodiaceae	*Microsorum punctatum* (L.) Copel.	Epiphytic	Leaves	Fodder	RJ0012		Na
10	Salviniaceae	*Azolla pinnata* R.Br.subsp. *Asiatica* R.M. K. Saunders & K. Fowler	F			RJ0100		Na
	Angiosperms							
	Dicotyledons							
1	Amaranthaceae	*Achyranthes bidentata* Blume	E	Stem, leaves	Fodder	RJ0011		Na
2	Lauraceae	*Actinodaphne gullavara* (Buch.-Ham. ex Nees) M.R.Almeida	Tree			L39,RJ0062		Na
3	Compositae	*Adenostemma lavenia* (L.) Kuntze	Herb			RJ0046		Na
4	Amaranthaceae	*Alternanthera sessilis* (L.) R.Br. ex DC.	E	Stem, leaves	Fodder	RJ0026	LC	Ex
5	Primulaceae	*Ardisia solanacea* (Poir.) Roxb.	Shrub			RJ0050		Na
6	Convolvulaceae	*Argyreia hookeri* C. B. Clarke	Climber			RJ0009		Na
7	Acanthaceae	*Barleria cristata* L.	Shrub	Stem, leaves	Fodder	RJ0006		Na
8	Compositae	*Bidens pilosa* L.	E	Stem, leaves	Fodder	RN024		Ex (Invasive)
9	Urticaceae	*Boehmeria macrophylla* Hornem.	Herb			RN018		Na
10	Lamiaceae	*Callicarpa macrophylla* Vahl	Herb	Root, fruit	Paste is eaten against gastritis and in fever	RJ0039	LC	Na

S. N.	Family	Scientific Name	Growth Form	Parts Used	Uses	Collection No.	IUCN Conservation Status	Native/ exotic
11	Celastraceae	*Celastrus* sp.	E			RJ0057		Na
12	Rubiaceae	*Cephalanthus tetrandra (*Roxb.) Ridsdale & Bakh.f.	E			RJPh2		Na
13	Compositae	*Chromolaena odorata* (L.) R.M.King & H.Rob.	Shrub	Whole plant	Used to make compost.	RJ0044		Ex (Invasive)
14	Lamiaceae	*Clerodendrum bracteatum* Wall. ex Walp.	Shrub	Stem, leaves	Fodder for pigs only.	RJ0058		Na
15	Lamiaceae	*Colebrookea oppositifolia* Sm.	Shrub	Leaves; flower	Used for ripening of banana, useful for bee keeping	RJ0053		Na
16	Leguminosae	*Crotalaria alata* D.Don	E			RJ0004		Na
17	Compositae	*Eclipta prostrata (L.) L.*	E	Leaves	Paste is applied in cut and wounds	RJ0040	LC	Ex
18	Compositae	*Elephantopus scaber* L.	E	Root	Paste is used as fermenting agent	RJ0016		Na
19	Compositae	*Emilia sonchifolia* (L.) DC. ex DC.	E	Root, stem	Paste is used as fermenting agent	RJ0005		Na
20	Acanthaceae	*Eranthemum pulchellum* Andrews	E	Stem, leaves	Fodder	RJ0018		Na
21	Euphorbiaceae	*Euphorbia hirta* L.	E	Stem, leaves, latex	Fodder; latex is used in cut and wound and in opaqueness of eye of cattle.	RN029		Ex
22	Leguminosae	*Flemingia strobilifera* (L.) W.T.Aiton	Shrub	Stem, leaves	Fodder	RJ0048		Na
23	Compositae	*Gnaphalium* sp.	Herb	Stem, leaves	Fodder	RJ0030		Na
24	Apocynaceae	*Ichnocarpus frutescens* (L.) W.T.Aiton	Climber			RN016		Na
25	Compositae	*Inula cappa* (Buch.-Ham. ex D.Don) DC	Shrub	Flower	Flower is used as fermenting agent	RJ0017		Na
26	Rubiaceae	*Knoxia sumatrensis* (Retz.) DC.	Herb			RJ0035		Na
27	Vitaceae	*Leea macrophylla* Roxb. ex Hornem.	Shrub	Leaves	Fodder	RJ0036		Na
28	Onagraceae	*Ludwigia perennis* L.	E	Stem, leaves	Fodder	RJ0021	LC	Na
29	Primulaceae	*Maesa chisia* Buch.-Ham. ex D. Don	Tree	leaves, fruit	Fodder, fruit paste is applied on skin of hand and legs for good looks.	RJ0055		Na
30	Leguminosae	*Millettia fruticosa* (DC.) Baker	Shrub			RJ0052	DD	Na
31	Compositae	*Mikania micrantha* Kunth	Climber	Stem, leaves	Fodder, flower is useful for honey bee to make honey	RJ0010		Ex (Invasive)
32	Leguminosae	*Mimosa pudica* L.	Shrub	Root	paste is eaten to cure throat ache; applied on wound.	RN022		Ex (Invasive)
33	Nelumbonaceae	*Nelumbo nucifera Gaertn.*	FL			RJPh3		Na
34	Menyanthaceae	*Nymphoides hydrophylla* (Lour.) Kuntze	FL			RN005	LC	Na
35	Apiaceae	*Oenanthe javanica* (Blume) DC.	E	Stem, leaves	Fodder	RJ0007	LC	Na
36	Polygonaceae	*Persicaria barbata* (L.) H.Hara	E			L19	LC	Na
37	Polygonaceae	*Persicaria hydropiper* (L.) Delarbre	E	Root, stem	Paste is used as fish poison for fishing	RJ0022	LC	Na
38	Piperaceae	*Piper longum* L.	Climber	Stem, leaves	Fodder	RJ0023		Na
39	Lamiaceae	*Pogostemon benghalensis* (Burm.f.) Kuntze	Herb			RJ0059		Na
40	Linaceae	*Reinwardtia indica* Dumort.	Shrub			RN009		Na
41	Lythraceae	*Rotala indica* (Willd.) Koehne	E			RN003	LC	Na
42	Acanthaceae	*Rungia pectinata* (L.) Nees	E	Stem, leaves	Fodder	RJ0027		Na

S. N.	Family	Scientific Name	Growth Form	Parts Used	Uses	Collection No.	IUCN Conservation Status	Native/ exotic
43	Plantaginaceae	*Scoparia dulcis* L.	Herb			RN026		Ex
44	Malvaceae	*Sida cordata* (Burm.f.) Borss.Waalk.	Shrub	Young shoot	Used as vegetable	RN023		Ex
45	Rubiaceae	*Spermacoce alata* Aubl.	Herb	Stem, leaves	Fodder	RJ0045		Ex. (Invasive)
46	Menispermaceae	*Stephania japonica* (Thunb.) Miers	Climber	Root	Paste is eaten against gastritis and stomach problems	RJ0025		Na
47	Acanthaceae	*Strobilanthes abbreviata* Y.F. Deng & J.R.I. Wood	Shrub	Stem, leaves	Fodder	RJ0014		Na
48	Malvaceae	*Urena lobata* L.	Shrub	Leaves, stem	Fodder	RJ0037		Na
	Monocotyledons							
1	Commelinaceae	*Commelina* sp.	E	Stem, leaves	Fodder, young leaves are used as vegetable	RJ0033		
2	Cyperaceae	*Courtoisina cyperoides* (Roxb.) Sojak	E			L7	LC	Na
3	Cyperaceae	*Cyperus platystylis* R.Br.	E			L24		Na
4	Cyperaceae	*Cyperus corymbosus* Rottb.	E			RJ0032		Na
5	Cyperaceae	*Cyperus esculentus* L.	E			RJ0038	LC	Na
6	Cyperaceae	*Cyperus pilosus* Vahl	E			RN032	LC	Na
7	Poaceae	*Dichanthelium* sp.	E			RN012		Na
8	Poaceae	*Digitaria longiflora* (Retz.) Pers.	E			L20		Na
9	Pontederiaceae	*Eichhornia crassipes* (Mart.) Solms	F			RJ0003		Ex
10	Eriocaulaceae	*Eriocaulon exsertum* Satake	E	Stem, leaves	Fodder	RN030		Na. En.
11	Cyperaceae	*Fimbristylis aestivalis* Vahl	E			L6		Na
12	Cyperaceae	*Fimbristylis dichotoma* (L.) Vahl	E			RN027	LC	Na
13	Commelinaceae	*Floscopa scandens* Lour.	E	Whole plant	Fodder	RJ0013	LC	Na
14	Poaceae	*Hymenachne amplexicaulis* (Rudge) Nees	E	Stem, leaves	Fodder	RN004		Na
15	Poaceae	*Isachne* sp.	E	Stem, leaves	Fodder	RN006		Na
16	Juncaceae	*Juncus* sp.	E	Stem, leaves	Fodder	RJ0015		Na
17	Juncaceae	*Juncus* sp.	E			RN031		Na
18	Juncaceae	*Juncus* sp.	E			RN033		Na
19	Zingiberaceae	*Kaempferia rotunda* L.	Herb	Tuber	Paste is applied to cure fracture	RN019		Na.
20	Araceae	*Lasia spinosa* (L.) Thwaites	E	Young shoot	Used as vegetable	RN008/L27	LC	Na
21	Apiaceae	*Oenanthe javanica* (Blume) DC.	E	Stem, leaves	Fodder	RJ0007	LC	Na
22	Hydrocharitaceae	*Ottelia alismoides* (L.) Pers.	S			RJ0001	LC	Na
23	Poaceae	*Panicum sumatrense* Roth	E	Stem, leaves	Fodder	RJ0061	LC	Na
24	Araceae	*Pistia stratiotes* L.	FL	Stem, leaves	Fodder for pigs	RN002	LC	Ex (Invasive)
25	Poaceae	Poa annua L.	Herb	Stem, leaves	Fodder	RJ0060		Na
26	Araceae	*Pothos chinensis* (Raf.) Merr.	Climber	Stem	Used to make local tools *nanglo*	RJ0049		Na
27	Poaceae	*Sacciolepis indica* (L.) Chase	E	Stem, leaves	Fodder	RN025		Na
28	Poaceae	*Setaria pumila* (Poir.) Roem. & Schult.	E	Stem, leaves	Fodder	RN028		Na
29	Smilacaceae	*Smilax ovalifolia* Roxb. ex D.Don	Climber	Stem	Cultural value; stick to play *dhangro* used by *dhami*	RJ0056		Na
30	Hydrocharitaceae	*Vallisneria natans* (Lour.) H. Hara	S			RN035		Na
31	Pandanaceae	*Pandanus furcatus* Roxb.	E			RNPh1		Na
32	Araceae	*Spirodela polyrrhiza* (L.) Schleid.	F					Na

Note: Life forms: E-emergent; FL-floating -leafed; F-floating; S-submerged; Na-Native; Ex-Exotic; LC least concern

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Orchids of Panchase Forest, Central Nepal: A Checklist

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Abstract

The checklist of orchids distributed in Panchase forest, Central Nepal is updated, with 52 genera, 142 species and a natural hybrid. Notes on the altitudinal range, habit, habitat, phenology and distribution are given.

Keywords: Hotspot, Mid-hill of Nepal, Orchidaceae, Orchid checklist, Orchids of Nepal

Introduction

Orchidaceae in Nepal is one of the largest plant families with about 506 species (Bhandari et al., 2015; Bhandari et al., 2016b; Subedi et al., 2017; Bhandari et al., 2019a; Bhandari et al., 2019b; Raskoti & Ale, 2019a; Raskoti & Ale, 2019b; Bhandari et al., 2020) distributed in range of habitats from tropical low land to high Himalaya (Acharya et al., 2011; Rokaya et al., 2013; Rajbhandari & Rai, 2017; Shrestha et al., 2018). Central Nepal is considered the high species-rich region for orchids with peaks at about 1600 m corresponding to 16°C mean annual temperature (Acharya et al., 2011). Panchase forest, lying at the mid-hill of central Nepal, with annual mean temperature about 17°C (Dixit et al., 2015), is being a centre of scientific research since early 2000, as several novel orchid species and new findings are reported from the forest (Bajracharya et al., 2003; Subedi et al., 2011; Raskoti & Ale, 2013; Raskoti & Jin, 2013; Bhandari et al., 2015; Raskoti & Kurzweil, 2015; Raskoti, 2015; Bhandari et al., 2020). This research was, therefore, carried out to contribute to the orchid conservation in Panchase by preparing an updated orchid checklist within the altitudinal range of 900-2500 m asl (meter above sea level). Incorporating the previous findings, this paper attempt to give comprehensive information's on orchid of Panchase, with updated nomenclature, habit, habitat, phenology, the elevation range and other relevant information.

Materials and Methods

Study area

The Panchase forest is located in the mid-hills of Central Nepal connecting the three districts; Kaski, Parbat and Syangja within the elevation range of 900 to 2500 m asl (Figure 1). The forest is characterized by the presence of different terraces of terrain with a range of habitat including forest, rangeland, shrub land, wetland and agriculture. The climate in the lower elevation is subtropical while it is moist temperate in the higher elevation above 2000 m. Hill Sal forest, *Schima-Castanopsis* forest dominate the lower subtropical belt, while *Daphniphyllum* forest, Alder forest, Oak- Laurel forest and Oak-*Rhododendron* forest dominate the upper temperate belt. Apart from its natural beauty, Panchase is famous for its religious value. Each year, hundreds

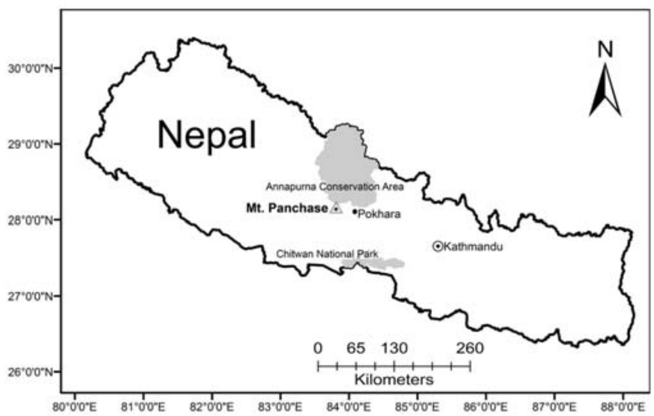

Figure 1: Location of Panchase in Nepal

of Hindus and Buddhist pilgrimages climb to Panchase to visit the holy sites; the Sidha-baba mandir and Panchase tal.

Home to about 718 vascular plant species (Subedi et al., 2007; Mikage, 2008; Bhandari, 2015; Bhandari et al., 2015; Bhandari & Shrestha, 2016; Bhandari et al., 2016-18; Thakur & Rajbhandary, 2018; Bhandari et al., 2020), Panchase forest is often considered as a hotspot of plant diversity in Central Nepal (World Wide Fund For Nature [WWF], 2013). Panchase also provides shelter to a number of endangered wildlife species including the Himalayan Black Bear (*Ursus thibetanus*), the common leopard (*Panthera pardus*), 152 species of birds, and 8 species of bats (Aryal & Dhungel, 2009; Malla et al., 2013; Baral, 2018). Owing to the biological uniqueness of the forest, Panchase is regarded as the biological corridor between southern plain Tarai and northern Himalaya; within a broad conservation regime, the landscape is popularly known as Chitwan-Annapurna Landscape (CHAL) (WWF, 2013).

Data collection

The present checklist is based on the published literature (Subedi et al., 2007; Raskoti, 2015), herbarium consultation and field visits. A bi-monthly field trip was organized for specimens collection between 2017 and 2018. At the meantime, the ecological parameters, for example, forest type, host tree, phenology, and altitudinal range were also recorded. The National Herbarium and Plant Laboratories (KATH) and Tribhuvan University Central Herbarium (TUCH) were visited for identifying the collected samples and cross-checking the vouchers collected from the study site. Information on herbarium specimens housed outside Nepal was obtained through Flora of Nepal webpage (http://www.floraofnepal.org/). Nomenclature and author citation of plant species followed the International Plant Name Index (https://www.ipni.org/), 'The Plant List' (http://theplantlist.org/) and TROPICOS (http://tropicos.org/). The species number for the genera and the broad distribution of the species followed Govaerts et al., (2019) and Chen et al., (2009), while the geographical regions are adopted from Watson et al., (2011), Rajbhandari & Rai (2017), Shrestha et al., (2018), Raskoti & Ale (2019a), Raskoti & Ale (2019b) and Bhandari et al., (2020) were followed for Nepal's distribution.

Results and Discussion

The present study reported 142 wild orchid species and a natural variety, within 52 genera from Panchase forest. *Bulbophyllum* (20 species), *Dendrobium* (15 species), *Pinalia* (8 species), *Oberonia* (7 species), *Coelogyne* (6 species), *Cymbidium* (6 species), *Gastrochilus* (5 species), *Liparis* (5 species), *Calanthe* (4 species), *Goodyera* (4 species), *Herminium* (4 species), *Odontochilus* (4 species), *Pholidota* (4 species), *Zeuxine* (4 species) and *Otochilus* (3 species), were the abundant genera, while, the remaining genera have either two or single species. Three Orchids (*Gastrochilus nepalensis* Raskoti, *Odontochilus nandae* Raskoti & H.Kurzweil, and *Pinalia pokharensis* (Bajrach., Subedi & K.K.Shrestha) Schuit., Y.P.Ng & H.A.Pedersen) are reported to be endemic to Panchase forest (Bajracharya et al., 2003; Raskoti & Kurzweil, 2015; Raskoti, 2015). *Oberonia nepalensis* L.R.Shakya & R.P.Chaudhary, endemic to central Nepal (Shakya & Chaudhary, 1999), was also found in good population in the subtropical (900 to 1500 m asl) Panchase forest. Of the total reported species, nine are Himalayan endemic, two distributed in South Asia (Nepal and India), and 118 species shows East and South Asian affinities, while the remaining 10 species shows the broad distribution. *Panisea panchaseensis* Subedi, named after the type locality Panchase forest (Subedi et al., 2011), was, later reported from Dolakha, Nepal (Bhandari et al., 2016) and Assam, India (Odyuo et al., 2016).

Majority of the orchid species prefers forest to rangeland and shrubland. A large number of the species favours epiphytic habitat (113 species), while 40 species prefer terrestrial habitat. Eight of the epiphytic species (*Bulbophyllum caudatum*, *B. hirtum*, *B. leopardinum*, *B. reptans*, *Coelogyne fuscescens*, *Dendrobium eriiflorum*, *Pholidota pallida*, *Porpax muscicola*) were occasional lithophyte, preferring the rock crevices. Interestingly, a terrestrial orchid, *Satyrium nepalense* var. *nepalense,* was observed in epiphytic habitat in the lower branches of *Daphniphyllum himalayense*, *Quercus semecarpifolia*, *Symplocos ramosissima*, and *Viburnum erubescens* as an occasional climber; however, the substratum was rich with mosses and litter. The epiphytic species tend to grow in damp forest, mostly in the lower tree trunk and branches rich with mosses. *Cymbidium aloifolium* and *C. manii* were observed growing in the open canopy, showing the high light demand. Few epiphytes like *Gastrochilus distichus*, *Oberonia falcata*, *Vandopsis undulata* were sometimes observed in *Berberis* bushes. The terrestrial species were often growing in the forest floor, forest edges, and grassy slope. The *Odontochilus* species, *Goodyera* species, *Liparis petiolata*, *Tainia minor* prefers to grow in the *Daphniphyllum* forest, floor rich with litter and mosses. Similarly, *Nervilia* species and *Zeuxine* species dominate the floor of *Schima-Castanopsis* forest within 900 to 1100 m asl. While, the species like *Herminium lanceum*, *Peristylis aristatus*, *Platanthera biermanniana*, *Satyrium nepalense* var. *nepalense*, *Spiranthes australis*, *S. flexuosa* prefers the grassy slope, forest edges and along the walking trail.

Schima wallichii, *Castanopsis indica*, *Diospyrus lancifolia* were the major host in the lower elevation, while *Bombax ceiba*, *Engelhardia spicata*, *Myrica esculenta*, *Macaranga pustulata* were the occasional host. In the temperate belt, *Daphniphyllum himalayense*, *Rhododendron arboreum*, *Quercus* species, *Lindera pulcheriima*, *Neolitsea pallens*, *Lyonia ovalifolia* were the major host. *Pyrularia edulis*, *Viburnum erubescens*, *Rhus* species served as rare host for the epiphytic species in the temperate belt. Some orchids were host-specific, for example, *Goodyera recurva* prefers *Viburnum erubescens* and *Neolitsea pallens*; *Panisea panchaseensis* prefers laurels and oaks, and *Taeniophyllum scaberulum* prefers *Daphniphyllum himalayense* and *Ilex excelsa*.

The peak flowering season for orchids in Panchase was observed between July to August, which can be linked with the rate of precipitation and temperature (Rokaya et al., 2013). More than 60% of orchids bloom in this period. The incoming spring brings the *Coelogyne* species to bloom, making the whole environment whitish. While, during August, the forest turns colourful (Subedi et al., 2007) with the blooming of different species of *Cymbidium*, *Bulbophyllum*, *Goodyera* and *Panisea*. Therefore, March-April and July-September could be the best season for the orchid enthusiast to visit Panchase.

Checklist

This checklist includes 142 wild species and a natural hybrid orchid species growing in Panchase forest, Central Nepal.

Acampe Lindl. (7/2/1 species)

Acampe rigida (Buch.-Ham. ex Sm.) P.F.Hunt; *Aerides rigida* Buch.-Ham. ex Sm.

Epiphytic on tree trunks or major branches (*Castanopsis indica*, *Schima wallichii*, *Diospyrus lancifolia*) between 900-1700 m. Fl. Aug-Oct. Ghatichida, Kaski, 900 m, 18 August 2015, P. Bhandari P1408 (KATH). [CE Nepal, E Himalaya, Assam-Burma, S Asia, E Asia, SE Asia and Africa].

Aerides Lour. (28/2/2 species)

***Aerides multiflora** Roxb.

Epiphytic on *Shorea robusta* within 1000-1500 m. Fl. May-Jun. [WCE Nepal, E Himalaya, S Asia and SE Asia].

Aerides odorata Lour.; *Aerides cornuta* Roxb.

Epiphytic on tree trunks or major branches (*Castanopsis indica*, *Schima wallichii*) between 900-1500 m. Fl. Jun-Jul. Ghatichida, NW Pokhara, Kaski, 900-1000 m, 28 June 2018, P. Bhandari & B. Ojha P1637 (KATH). [WCE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Agrostophyllum Blume (139/2/2 species)

Agrostophyllum callosum Rchb.f.

Epiphytic on lower branches (*Quercus* species, *Rhododendron arboreum*, *Daphniphyllum himalayense*, *Ilex fragilis*, *Lyonia ovalifolia*, *Myrica esculenta*, *Schima wallichii*) between 1700-2300 m. Fl. Jun-Jul. Panchase forest, Kaski, 2200 m, 10 August 2014, P. Bhandari & A. Bhandari P774 (TUCH). [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Ania Lindl. (6/1/1 species)

Ania penangiana (Hook.f.) Summerh.

Terrestrial on the floor of *Schima-Castanopsis* forest between 1000-1100 m. Fl. Mar-Apr. Between Ghatichida to Damdame, Panchase forest, Kaski, 1045 m, 17 March 2018, P. Bhandari P1505, KATH030392. [C Nepal, E Himalaya, E Asia and SE Asia].

Anthogonium Lindl. (1/1/1 species)

Anthogonium gracile Lindl.
Terrestrial on grassy slopes, in rock crevices between 1300-1800 m. Fl. Aug-Sep. Panchase forest, Kaski, 1800 m, 07 September 2014, P. Bhandari & S. Budhamagar P800 (KATH & TUCH). [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, S Asia, E Asia and SE Asia].

Apostasia Blume (8/1/1 species)

Apostasia wallichii R.Br.
Terrestrial on *Schima-Castanopsis* forest between 900-1000 m. Fl. May-Jun. Between Damdame to Ghatichida, Panchase forest, Kaski, 900-1000 m, 28 June 2018, P. Bhandari & B. Ojha P1632 (KATH). [C Nepal, Assam-Burma, S Asia, E Asia, SE Asia and Australasia].

Bulbophyllum Thouars (ca. 1900/36/20 species)

Bulbophyllum affine Wall. ex Lindl.
Epiphytic on tree trunks (*Schima wallichii*, *Castanopsis indica*) between 1100-1500 m. Fl. Jun-Jul. Panchase forest, Kaski, 1100-1500 m, 27 June 2018, P. Bhandari P1657 (KATH). [CE Nepal, E Himalaya, E Asia and SE Asia].

Bulbophyllum bisetum Lindl.
Epiphytic on tree trunks (*Daphniphyllum himalayense*) between 1900-2100 m. Fl. Sep. Panchase forest, Kaski, 1950 m, 22 September 2017, P. Bhandari P1696 (KATH). [CE Nepal, E Himalaya and SE Asia].

Bulbophyllum careyanum (Hook.) Spreng.; *Anisopetalum careyanum* Hook.
Epiphytic on tree trunks and branches (*Schima wallichii*, *Castanopsis* species, *Diospyrus lancifolia*, *Myrsine capitellata*) between 900-1700 m. Fl. Sep-Nov. Thulakhet, NW Pokhara, Kaski, 1000 m, 10 October 2014, P. Bhandari P1495 (KATH). [CE Nepal, E Himalaya, Assam-Burma and SE Asia].

***Bulbophyllum caudatum** Lindl.
Epiphytic or Lithophytic. Fl. Jun-Jul. [CE Nepal, E Himalaya and Tibetan Plateau].

Bulbophyllum cylindraceum Wall. ex Lindl.
Epiphytic on tree trunks (*Daphniphyllum himalayense*) at 1700 m. Fl. Oct-Nov. Panchase forest, Kaski, 1700 m, 10 November 1999, A. Subedi 347 (TUCH!). [CE Nepal, E Himalaya and E Asia].

***Bulbophyllum hirtum** (Sm.) Lindl. ex Wall.
Stelis hirta Sm.
Epiphytic or Lithophytic. Fl. Jan-Feb. [Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

Bulbophyllum leopardinum (Wall.) Lindl. ex Wall.; *Dendrobium leopardinum* Wall.
Epiphytic on tree trunks (*Daphniphyllum himalayense*, *Rhododendron arboreum*, *Schima wallichii*, *Alnus nepalensis*, *Engelhardia spicata*, *Ilex fragilis*, *Lyonia ovalifolia*, *Myrica esculenta*) or lithophytic between 1500-2000 m. Fl. Jun-Jul. Panchase forest, Kaski, 1900 m, 05 October 2014, P. Bhandari P1567 (TUCH). [CE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, E Asia and SE Asia].

***Bulbophyllum moniliforme** C.S.P.Parish & Rchb.f.
Epiphytic on tree trunks. Fl. Oct. [C Nepal, Assam-Burma and SE Asia].

Bulbophyllum muscicola Rchb.f.
Epiphytic on tree trunks and lower branches (*Quercus semecarpifolia*, *Rhododendron arboreum*, *Lyonia ovalifolia*, *Quercus lamellosa*) between 2300-2500 m. Fl. Sep. Panchase danda, Kaski, 2500 m, 09 September 2014, P. Bhandari & S. Budhamagar P827 (TUCH). [C Nepal, W Himalaya, E Himalaya and Assam-Burma].

Bulbophyllum odoratissimum (Sm.) Lindl.; *Stelis odoratissima* Sm.
Epiphytic on tree trunks and lower branches (*Schima wallichii*, *Castanopsis indica*) between 1400-1700 m. Fl. Jun-Jul. Panchase forest, Kaski, 1400-1700 m, 06 June 2017, P. Bhandari & A. Bhandari P1664 (KATH). [C Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

***Bulbophyllum polyrhizum** Lindl.
Epiphytic on tree trunks. Fl. Apr. [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia].

Bulbophyllum purpureofuscum J.J.Verm., Schuit. & de Vogel; *Sunipia cirrhata* (Lindl.) P.F.Hunt
Epiphytic on tree trunks and lower branches (*Daphniphyllum himalayense*, *Quercus lamellosa*, *Rhus succedanea*) between 1900-2300 m. Fl. Dec-Jan. Panchase forest, Kaski, 1900-2300 m, 08 January 2015, P. Bhandari P1692 (KATH). [C Nepal, E Himalaya, Assam-Burma and E Asia].

Bulbophyllum reptans (Lindl.) Lindl.; *Tribrachia reptans* Lindl.
Epiphytic on tree trunks (*Schima wallichii*, *Castanopsis indica*, *Daphniphyllum himalayense*, *Rhododendron arboreum*, *Quercus semecarpifolia*, *Lyonia ovalifolia*, *Ilex fragilis*, *Rhus succedanea*) or lithophytic between 1300-2500 m. Fl. Mar-Jun. Panchase forest, Kaski, 1600 m, 20 March 2015, P. Bhandari, S. Budhamagar & A. Bhandari P1276 (KATH & TUCH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Bulbophyllum retusiusculum Rchb.f.
Epiphytic on tree trunks and lower branches (*Quercus semecarpifolia*, *Rhododendron arboreum*, *Ilex fragilis*, *Lindera pulcherrima*) between 2300-2500 m. Fl. Jul-Aug. Panchase forest, Kaski, 2500 m, 10 July 2000, A. Subedi 430 (TUCH!). [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Bulbophyllum roseopictum J.J.Verm., Schuit. & de Vogel; *Sunipia bicolor* Lindl.
Epiphytic on tree trunks and lower branches (*Daphniphyllum himalayense*, *Quercus semecarpifolia*, *Viburnum erubescens*) between 2000-2500 m. Fl. Nov-Dec. Panchase forest, Kaski, 1500-2100 m, 08 January 2015, P. Bhandari & Y.N. Paudel P1691 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, S Asia, E Asia and SE Asia].

Bulbophyllum secundum Hook.f.
Epiphytic on tree trunks (*Schima wallichii*) between 900-

1000 m. Fl. Jun-Jul. Ghatichida, Panchase forest, Kaski, 900 m, 28 June 2018, P. Bhandari & B. Ojha P1634 (KATH). [C Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Bulbophyllum striatum (Griff.) Rchb.f.; *Dendrobium striatum* Griff.

Epiphytic on tree trunks at 2000 m. Fl. Jan-Feb. Panchase forest, Kaski, 2000 m, 11 January 2002, A. Subedi, R.P. Chaudhary & L.R. Shakya 1035 (TUCH!). [C Nepal, E Himalaya, E Asia and SE Asia].

***Bulbophyllum umbellatum** Lindl.

Epiphytic on tree trunks. Fl. Mar-Jun. [Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Bulbophyllum viridiflorum (Hook.f.) Schltr.; *Cirrhopetalum viridiflorum* Hook.f.

Epiphytic on tree trunks (*Daphniphyllum himalayense*) between 2000-2200 m. Fl. Sep-Oct. Panchase forest, Kaski, 2100 m, 05 October 2014, P. Bhandari & A. Bhandari P1235 (KATH). [CE Nepal, E Himalaya, S Asia and E Asia].

Bulbophyllum wallichii (Lindl.) Rchb.f.; *Cirrhopetalum wallichii* Lindl.

Epiphytic on tree trunks (*Daphniphyllum himalayense*) between 2000-2100 m. Fl. Mar-Apr. Panchase forest, Kaski, 2000 m, 22 March 2015, P. Bhandari, S. Budhamagar & A. Bhandari P1496 (TUCH). [WCE Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

Calanthe R.Br. (ca. 150/16/4 species)

Calanthe mannii Hook.f.

Terrestrial on Oak forest floor at 2300 m. Fl. May-Jun. Panchase forest, Kaski, 2300 m, 27 June 2018, P. Bhandari P1646 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Calanthe plantaginea Lindl.

Terrestrial on *Rhododendron*-Oak forest floor between 2400-2500 m. Fl. Mar-Apr. Panchase forest, Kaski, 2450 m, 22 March 2015, P. Bhandari & S. Budhamagar P1547 (TUCH). [WC Nepal, W Himalaya, E Himalaya, Tibetan Plateau and E Asia].

Calanthe puberula Lindl.; *Alismorkis puberula* (Lindl.) Kuntze

Terrestrial on *Rhododendron*-Oak forest floor between 1900-2500 m. Fl. Jul-Aug. Panchase forest, Kaski, 1900 m, 16 August 2017, P. Bhandari, K. Shrestha, G. Thapamagar & D. Pathak 1008P (KATH). [CE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, E Asia and SE Asia].

Calanthe tricarinata Lindl.

Terrestrial on Oak forest floor at 2300 m. Fl. May-Jun. Panchase forest, Kaski, 2300 m, 17 April 2018, P. Bhandari P1607 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau and E Asia].

Cheirostylis Blume (55/1/1 species)

Cheirostylis griffithii Lindl.

Saprophytic growing on the floor of *Schima-Castanopsis* forest between 900-2000 m. Fl. Sep. Panchase forest, Kaski, 900 m, 17 March 2018, P. Bhandari P1702 (KATH). [C Nepal, W Himalaya, E Himalaya, Assam-Burma, S Asia, E Asia and SE Asia].

Chiloschista Lindl. (20/3/1 species)

Chiloschista usneoides (D.Don) Lindl.; *Epidendrum usneoides* D.Don

Epiphytic on tree trunks (*Castanopsis indica*) at 900-1900 m. Fl. May-Jun. Thulakhet, Bhadaure Tamage VDC, Kaski, 900 m, 04 May 2015, P. Bhandari P1356 (TUCH). [C Nepal, W Himalaya and E Himalaya].

Cleisostoma Blume (99/4/1 species)

***Cleisostoma filiforme** (Lindl.) Garay; *Sarcanthus filiformis* Lindl.

Epiphytic on tree trunks and branches within 1000-1200 m. Fl. Aug-Sep. [CE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Coelogyne Lindl. (ca. 200/11/6 species)

***Coelogyne corymbosa** Lindl.

Epiphytic on tree trunks (Rhododendron forest) within 1600-2300 m. Fl. Mar-Apr. [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, S Asia and E Asia].

Coelogyne cristata Lindl.

Epiphytic on tree trunks and major branches (*Schima wallichii*, *Daphniphyllum himalayense*, *Rhododendron arboreum*, *Alnus nepalensis*, *Ilex fragilis*) between 1600-2300 m. Fl. Mar-Apr. Panchase forest, Kaski, 2000 m, 21 March 2015, P. Bhandari, S. Budhamagar & A. Bhandari P1289 (KATH & TUCH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau and S Asia].

***Coelogyne flaccida** Lindl.

Epiphytic on tree trunks near streams and water bodies. Fl. Mar-Apr. [C Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Coelogyne fuscescens Lindl.; *Coelogyne fuscescens* var. *viridiflorum* Pradhan

Epiphytic on tree trunks and lower branches (*Schima wallichii*, *Castanopsis* species, *Daphniphyllum himalayense*, *Rhododendron arboreum*, *Alnus nepalensis*, *Engelhardia spicata*, *Ilex fragilis*, *Lyonia ovalifolia*, *Myrica esculenta*, *Prunus cerasoides*) or lithophytic between 1100-2100 m. Fl. Dec-Feb. Panchase forest, Kaski, 1940 m, 09 January 2015, P. Bhandari & Y.P. Paudel P1256 (TUCH). [CE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Coelogyne nitida (Wall. ex D.Don) Lindl.; *Cymbidium nitidum* Wall. ex D.Don

Epiphytic on tree trunks and lower branches (*Schima wallichii*, *Castanopsis indica*, *Daphniphyllum himalayense*, Oaks, Laurels) between 1600-2500 m. Fl. May-Jul. Panchase forest, Kaski, 1950 m, 02 June 2014, P. Bhandari & S. Budhamagar P417 (TUCH). [WCE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Coelogyne prolifera Lindl.; *Coelogyne flavida* Hook.f. ex Lindl.

Epiphytic on tree trunks and branches (*Schima wallichii*, *Castanopsis indica*, *Diospyrus lancifolia*, *Ficus neerifolia*, *Daphniphyllum himalayense*, *Lyonia ovalifolia*, *Engelhardia spicata*) between 900-1900 m. Fl. May-Jun. Panchase forest, Kaski, 1510 m, 04 June 2014, P. Bhandari, S. Budhamagar & R. Chhetri P509 (TUCH & KATH). [CE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Crepidium Tausch (280/2/1 species)

Crepidium acuminatum (D.Don) Szlach.; *Malaxis acuminata* D.Don

Terrestrial on *Daphniphyllum* forest between 1400-1900 m. Fl. Jun-Jul. Panchase forest, Kaski, 1400 m, 11 September 2014, P. Bhandari & S. Budhamagar P873 (KATH & TUCH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia, SE Asia and Australasia].

Cryptochilus Wall. (8/2/1 species)

Cryptochilus luteus Lindl.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, *Quercus semecarpifolia*, *Lyonia ovalifolia*) between 1900-2500 m. Fl. Jun-Aug. Panchase forest, Kaski, 2200 m, 10 August 2014, P. Bhandari & A. Bhandari P773 (KATH & TUCH). [WCE Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia SE Asia].

Cymbidium Sw. (55/9/6 species)

Cymbidium aloifolium (L.) Sw.; *Epidendrum aloifolium* L.

Epiphytic on tree trunks and branches (*Castanopsis indica*, *Schima wallichii*) or lithophytic to 1200 m. Fl. Jun-Jul. Thulakhet to Harpan, Panchase, Kaski, 1200 m, 27 June 2018, P. Bhandari P1637 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Assam-Burma, S Asia, E Asia and SE Asia].

Cymbidium elegans Lindl.; *Cymbidium longifolium* D.Don

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, Oak, *Lindera pulcherrima*) between 2000-2500 m. Fl. Oct-Jan. Panchase forest, Kaski, 2260 m, 05 October 2014, P. Bhandari & A. Bhandari P1220 (TUCH). [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma and E Asia].

Cymbidium erythraeum Lindl.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, *Quercus semecarpifolia*) between 2200-2300 m. Fl. Oct-Nov. Panchase forest, Kaski, 2260 m, 05 October 2014, P. Bhandari & A. Bhandari P1219 (TUCH). [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, S Asia, E Asia and SE Asia].

Cymbidium × gammieanum King & Pantl.; *Cyperorchis gammieana* (King & Pantl) Schltr.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, Oak) at 2200 m. Fl. Oct-Nov. Panchase forest, Kaski, 2200 m, 09 November 2002, A. Subedi, R.P. Chaudhary & L.R. Shakya 1023 (TUCH!). [CE Nepal and E Himalaya].

Cymbidium lancifolium Hook.

Terrestrial on *Schima-Castanopsis* forest floor between 1800-1900 m. Fl. Sep-Oct. Panchase forest, Kaski, 1900 m, 15 July 2002, A. Subedi, R.P. Chaudhary & L.R. Shakya 94 (TUCH!). [C Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Cymbidium mannii Reichnb.; *Cymbidium bicolor* Lindl. var. *obtusum* Du Puy & P.J.Cribb

Epiphytic on tree trunks and branches (*Castanopsis indica*) or lithophytic between 1000-1300 m. Fl. Mar-May. Harpan village, Kaski, 1300 m, 23 March 2015, P. Bhandari & S. Budhamagar P1548 (TUCH). [WCE Nepal, Assam-Burma, S Asia and SE Asia].

Dendrobium Sw. (ca. 1100/26/15 species)

Dendrobium amoenum Wall. ex Lindl.; *Callista amoena* (Wall. ex Lindl.) Kuntze

Epiphytic on tree trunks and branches (*Schima wallichii*, *Daphniphyllum himalayense*, *Alnus nepalensis*) between 1500-1800 m. Fl. May-Jun. Panchase forest, Kaski, 1700 m, 04 June 2014, P. Bhandari & S. Budhamagar P507 (TUCH). [CE Nepal, W Himalaya, E Himalaya, Assam-Burma and S Asia].

Dendrobium amplum Lindl.; *Epigeneium amplum* (Lindl.) Summerh.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*) between 1800-1900 m. Fl. Aug-Oct. Panchase forest, Kaski, 1890 m, 06 October 2014, P. Bhandari P1239 (TUCH). [CE Nepal, E Himalaya, Tibetan Plateau, E Asia and SE Asia].

***Dendrobium aphyllum** (Roxb.) C.E.C.Fisch.; *Limodorum aphyllum* Roxb.

Epiphytic on tree trunks (*Castanopsis* forest) up to 1400 m. Fl. May-Jun. [CE Nepal, E Himalaya, Assam-Burma, S Asia, SE Asia and Australasia].

Dendrobium chryseum Rolfe

Epiphytic on tree trunks and branches (*Schima wallichii*, *Castanopsis indica*) between 1100-1200 m. Fl. Aug-Sep. Ghatichida to Makwanpur, Panchase forest, Kaski, 1100 m, 15 September 2017, P. Bhandari, K. Shrestha, A. Bhandari, & M. Paudel, P1698 (KATH). [C Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

***Dendrobium crepidatum** Lindl. & Paxton

Epiphytic on tree trunks (*Castanopsis* forest) up to 1400 m. Fl. Apr. [C Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

***Dendrobium densiflorum** Wall.

Epiphytic on tree trunks on *Castanopsis* forest. Fl. Apr-May. [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Dendrobium denudans D.Don

Epiphytic on tree trunks and branches (*Schima wallichii*) between 1500-1600 m. Fl. Aug-Sep. Panchase forest, Kaski, 1500 m, 10 September 2014, P. Bhandari & A. Bhandari P853 (TUCH). [WCE Nepal, W Himalaya and E Himalaya].

Dendrobium eriiflorum Griff.

Epiphytic on tree trunks and branches (*Schima wallichii*, *Castanopsis indica*, *Rhododendron arboreum*, *Alnus* *nepalensis*, *Albizia* species) or lithophytic between 900-1900 m. Fl. Sep-Oct. Panchase forest, Kaski, 1900 m, 05 October 2014, P. Bhandari P1549 (TUCH). [WCE Nepal, E Himalaya, Assam-Burma, S Asia and SE Asia].

Dendrobium heterocarpum Wall. ex Lindl.

Epiphytic on tree trunks and branches (*Schima wallichii*, *Castanopsis indica*, *Lyonia ovalifolia*) between 1300-1600 m. Fl. Mar-Apr. Panchase forest, Kaski, 1600 m, 20 March 2015, P. Bhandari, S. Budhamagar & A. Bhandari P1283 (KATH & TUCH). [WCE Nepal, W Himalaya, E Himalaya, Assam-Burma, S Asia, E Asia and SE Asia].

Dendrobium longicornu Lindl.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, *Ilex* species, Laurel, *Eurya cerasifolia*, *Quercus lamellosa*) between 1800-2500 m. Fl. Oct-Nov. Panchase forest, Kaski, 1800 m, 04 October 2014, P. Bhandari & A. Bhandari P1211 (TUCH). [CE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

***Dendrobium moniliforme** (L.) Sw.; *Epidendrum moniliforme* L., *Dendrobium candidum* Wall ex Lindl.

Epiphytic on Oak and *Rhododendron* forest. Fl. May-Jun. [CE Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

***Dendrobium moschatum** (Buch.-Ham.) Sw.

Epiphytic on tree trunks (*Castanopsis* forest) up to 1400 m. Fl. Jun-Jul. [CE Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

***Dendrobium polyanthum** Wall. ex Lindl.; *Dendrobium primulinum* Lindl.

Epiphytic on tree trunks (*Castanopsis* forest) up to 1400 m. Fl. Apr. [WC Nepal, W Himalaya, Assam-Burma, E Asia and SE Asia].

Dendrobium porphyrochilum Lindl.

Epiphytic on tree trunks (*Daphniphyllum himalayense*, *Lindera pulcherrima*, *Rhododendron arboreum*, *Lyonia ovalifolia*, *Quercus semecarpifolia*, *Symplocos ramosissima*) between 2300-2500 m. Fl. Jun-Jul. Panchase forest, Kaski, 2500 m, 07 June 2014, P. Bhandari, R. Chhetri & S. Budhamagar P599 (KATH & TUCH). [CE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

***Dendrobium transparens** Wall. ex Lindl.

Epiphytic on tree trunks (*Castanopsis* forest) up to 1400 m. Fl. May-Jun. [WCE Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

Dienia Lindl. (6/4/2 species)

Dienia ophrydis (J.Koenig) Seidenf.; *Epidendrum ophrydis* J.Koenig, *Malaxis ophrydis* (J.Koenig) Ormerod

Terrestrial on *Schima-Castanopsis* forest floor between 900-1500 m. Fl. Jun-Jul. Ghatichida, Panchase forest, Kaski, 900 m, 28 June 2018, P. Bhandari & B. Ojha P1663 (KATH). [CE Nepal, W Himalaya, E Himalaya, Assam-Burma, S Asia, E Asia, SE Asia and Australasia].

Eria Lindl. (ca. 54/3/1 species)

Eria coronaria (Lindl.) Rchb.f.; *Coelogyne coronaria* Lindl.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, *Quercus lamellosa*, *Schima wallichii*) between 1500-2300 m. Fl. Nov-Dec. Panchase forest, 7500 ft., 1954, J.D.A. Stainton 9331 (BM, E n.v.). Panchase forest, Kaski, 1800 m, 14 December 2015, P. Bhandari & K. Shrestha P1693 (KATH). [CE Nepal, E Himalaya, Tibetan Plateau, E Asia and SE Asia].

Galeola Lour. (10/1/1 species)

Galeola lindleyana (Hook.f. & Thomson) Rchb.f.; *Cyrtosia lindleyana* Hook.f. & J.W.Thomson

Saprophytic growing *Daphniphyllum* forest floor at 1950 m. Fl. Jun-Jul. Panchase forest, Kaski, 1950 m, 27 June 2018, P. Bhandari P1656 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, E Asia and SE Asia].

Gastrochilus D.Don (ca. 66/7/5 species)

Gastrochilus affinis (King & Pantl.) Schltr.; *Saccolabium affine* King & Pantl.

Epiphytic on lower branches (*Quercus semecarpifolia*, *Rhododendron arboreum*, *Lindera pulcherrima*) above 2400 m. Fl. May-Jun. Panchase forest, Kaski, 2490 m, 07 June 2014, P. Bhandari, S. Budhamagar & R. Chhetri P600 (KATH). [CE Nepal, E Himalaya and E Asia].

Gastrochilus calceolaris (Buch.-Ham. ex Sm.) D.Don; *Aerides calceolaris* Buch.-Ham. ex Sm, *Gastrochilus calceolaris* var. *biflora* L.R.Shakya & M.R.Shrestha

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, *Castanopsis indica*, *Schima wallichii*, *Rhus succadaena*) between 1100-2000 m. Fl. Mar-Apr. Panchase forest, Kaski, 1940 m, 21 March 2015, P. Bhandari, S. Budhamagar & A. Bhandari P1288 (KATH & TUCH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Gastrochilus distichus (Lindl.) Kuntze; *Saccolabium distichum* Lindl.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, *Neolitsea pallens*, *Quercus semecarpifolia*, *Rhododendron arboreum, Berberis* species) between 1800-2500 m. Fl. Jan-Feb. Panchase forest, Kaski, 2500 m, 09 January 2015, P. Bhandari & Y.P. Paudel P1264 (TUCH). [CE Nepal, W Himalaya, E Himalaya, Tibetan Plateau and E Asia].

Gastrochilus nepalensis Raskoti

Epiphytic on tree branches at about 2350 m. Fl. Mar. Above Deurali on the way to Kande, Kaski, 2350 m, 22 March 2007, BB Raskoti 00745 (KATH!). **Endemic to Panchase**. *Type*: Central Nepal, Kaski, above Deurali on the way to Kande, 2350 m, 22 March 2007, BB Raskoti 00745 (holotype: KATH!).

Gastrochilus pseudodistichus (King & Pantl.) Schltr.; *Saccolabium pseudodistichum* King & Pantl.

Epiphytic on tree trunks (*Daphniphyllum himalayense, Berberis* bushes) between 1800-2500 m. Fl. Sep-Oct. Panchase forest, Kaski, 2020 m, 05 October 2014, P. Bhandari & A. Bhandari P1217 (TUCH). [C Nepal, E Himalaya, E Asia and SE Asia].

Goodyera R.Br. (ca. 100/10/4 species)

Goodyera foliosa (Lindl.) Benth. ex C.B.Clarke; *Georchis foliosa* Lindl.

Terrestrial on *Daphniphyllum* and Oak forest floor between 2000-2200 m. Fl. Sep-Oct. Panchase forest, Kaski, 2100 m, 05 October 2014, P. Bhandari & A. Bhandari P1233 (TUCH). [CE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Goodyera procera (Ker Gawl.) Hook.; *Neottia procera* Ker Gawl.

Terrestrial on grassy slopes, roadsides, forest edges (*Schima-Castanopsis* forest) between 900-1300 m. Fl. May-Jun. Panchase forest, Kaski, 900-1200 m, P. Bhandari P1449 (KATH). [WC Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, S Asia, E Asia and SE Asia].

Goodyera recurva Lindl.

Epiphytic on tree trunks of *Neolitsea pallens*, *Viburnum erubescens* between 2300-2500 m. Fl. Sep-Oct. Panchase forest, Kaski, 2300-2450 m, 11 September 2017, P. Bhandari & A. Bhandari P1610 (KATH). [C Nepal, E Himalaya and E Asia].

Goodyera viridiflora (Blume) Lindl. ex D.Dietr.; *Neottia viridiflora* Blume

Terrestrial on the forest floor (*Schima-Castanopsis*, *Daphniphyllum* forest) between 1300-2000 m. Fl. Jul-Aug. Panchase forest, Kaski, 1300-2000 m, 17 August 2017, P. Bhandari and G. Thapamagar 1010P (KATH). [CE Nepal, W Himalaya, E Himalaya, E Asia, SE Asia and Australasia].

Habenaria Willd. (ca. 891/18/1 species)

Habenaria intermedia D.Don

Terrestrial on grassy slopes between 2000-2300 m. Fl. Jul-Aug. Bhanjyang, Panchase, Kaski, 2000 m, 18 August 2015, P. Bhandari 1448 (KATH). [WC Nepal, W Himalaya and Tibetan Plateau].

Hemipilia Lindl. (12/1/1 species)

Hemipilia cordifolia Lindl.

Terrestrial on temperate grassy slopes at 2100 m. Fl. Jul-Aug. Bhanjyang, Panchase, Kaski, 2100 m, 27 July 2002, A. Subedi 960 (TUCH!). [WC Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma and E Asia].

Herminium L. (50/24/4 species)

Herminium lanceum (Thunb. ex Sw.) Vuijk; *Ophrys lancea* Thunb. ex Sw.

Terrestrial on grassy slopes and forest floor (*Daphniphyllum* forest) between 1800-2000 m. Fl. Jul-Aug. Sidhane, Bhadaure Tamage VDC, Kaski, 1820 m, 09 August 2014, P. Bhandari & A. Bhandari P735 (TUCH). [WCE Nepal, W Himalaya, E Himalaya, Assam-Burma, Assam-Burma, E Asia and SE Asia].

***Herminium latilabre** (Lindl.) X.H.Jin, Schuit, Raskoti & L.Q.Huang; *Platanthera latilabris* Lindl.

Terrestrial on grassy slopes between 1700-2100 m. Fl. Jul. [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau and E Asia].

Herminium mackinonii Duthie

Epiphytic on tree trunk of *Quercus semecarpifolia*, *Ilex fragilis* between 2300-2500 m. Fl. Jul-Aug. Panchase danda, Kaski, 2500 m, 17 August 2015, P. Bhandari & S. Budhamagar P1452 (TUCH & KATH). [WCE Nepal, W Himalaya and E Himalaya].

Herminium souliei (Finet) Rolfe; *Herminium angustifolium* var. *souliei* Finet

Terrestrial on grassy slopes at 2400 m. Fl. Jul-Aug. Panchase forest, Kaski, 2400 m, 12 July 2008, B. Raskoti 515 (KATH). [WCE Nepal, Tibetan Plateau and E Asia].

Liparis Rich. (ca. 436/18/5 species)

Liparis bootanensis Griff.

Epiphytic on tree trunks of *Quercus semecarpifolia* at 1600-2200 m. Fl. Aug-Sep. Panchase forest, Kaski, 02 August 2002, A. Subedi, R.P. Chaudhary & L.R. Shakya, 972 (TUCH!). [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Liparis nervosa (Thunb.) Lindl.; *Ophrys nervosa* Thunb.

Terrestrial on the floor of *Schima-Castanopsis* forest at 1300 m. Fl. Jun-Jul. Panchase forest, Kaski, 1300 m, 27 June 2018, P. Bhandari P1660 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, S Asia, E Asia, SE Asia; widespread in Old and New World tropics].

Liparis petiolata (D.Don) P.F.Hunt & Summerh.; *Acianthus petiolatus* D.Don

Terrestrial on the forest floor (Oak-*Rhododendron* forest) between 2300-2500 m. Fl. Jul-Sep. Panchase forest, Kaski, 2410 m, 10 August 2014, P. Bhandari & A. Bhandari P765 (TUCH & KATH). [C Nepal, E Himalaya, Tibetan Plateau, E Asia and SE Asia].

Liparis resupinata Ridl.

Epiphytic on tree trunks (*Alnus nepalensis*, *Macaranga pustulata*, *Daphniphyllum himalayense*, *Lyonia ovalifolia*, *Rhododendron arboreum*) between 1500-2100 m. Fl. Oct-Jan. Bhadaure Deurali, Panchase, Kaski, 1850 m, 08 January 2015, P. Bhandari & Y.P. Paudel P1250 (KATH & TUCH). [CE Nepal, W Himalaya, E Himalaya, Tibetan Plateau and E Asia].

***Liparis viridiflora** (Blume) Lindl.

Epiphytic on Oak. Fl. Nov-Dec. [CE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, S Asia, E Asia, SE Asia and Australasia].

Luisia Gaudich. (ca. 44/2/1 species)

Luisia tristis (G.Forst.) Hook.f.; *Epidendrum triste* G.Forst

Epiphytic on tree trunks (*Schima wallichii*, *Castanopsis indica*, *Ficus religiosa*, *Bombax ceiba*) between 900-1200 m. Fl. Apr-May. Thulakhet, Panchase, Kaski, 1200 m, 27 June 2018, P. Bhandari P1641 (KATH). [WCE Nepal, W Himalaya, E Himalaya and S Asia].

Mycaranthes Blume (37/1/1 species)

Mycaranthes floribunda (D.Don) S.C.Chen & J.J.Wood.; *Dendrobium floribundum* D.Don

Epiphytic on tree trunks and upper branches (*Schima* *wallichii*, *Castanopsis indica*, *Lyonia ovalifolia*, *Diospyrus lancifolia*) between 900-1500 m. Fl. Apr-May. Between Ghatichida to Damdame, Panchase, Kaski, 900-1000 m, 06 May 2018, P. Bhandari P1623 (KATH). [CE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Neottia Guett. (72/8/1 species)

Neottia longicaulis (King & Pantl.) Szlach.; *Listera longicaulis* King & Pantl.

Terrestrial on Oak-*Rhododendron* forest floor between 2400-2500 m. Fl. Jun-Jul. Panchase forest, Kaski, 2400-2500 m, 27 June 2018, P. Bhandari P1711, KATH030407. [WC Nepal, E Himalaya and Tibetan Plateau].

Nervilia Comm. ex Gaudich. (75/6/2 species)

Nervilia crociformis (Zoll. & Moritzi) Seidenf.; *Bolborchis crociformis* Zoll. & Moritzi

Terrestrial on *Schima-Castanopsis* forest floor between 900-1000 m. Fl. Apr-May. Ghatichida, Panchase forest, Kaski, 900-1000 m, 28 June 2018, P. Bhandari & B. Ojha P1635 (KATH). [E Nepal, W Himalaya, E Himalaya, E Asia, SE Asia, Africa and Australasia].

Nervilia macroglossa (Hook.f.) Schltr.; *Pogonia macroglossa* Hook.f.

Terrestrial on *Schima-Castanopsis* forest floor at 1300 m. fl. Apr-May. Panchase forest, Kaski, 1300 m, 27 June 2018, P. Bhandari P1659 (KATH). [E Nepal, E Himalaya, Assam-Burma and SE Asia].

Oberonia Lindl. (ca. 306/18/7 species)

***Oberonia acaulis** Lindl.; *Oberonia myriantha* Lindl.

Epiphytic on tree branches within 1400-1800 m. Fl. Sep-Oct. [CE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Oberonia ensiformis (Sm.) Lindl.; *Malaxis ensiformis* Sm.

Epiphytic on tree trunks (*Schima wallichii*, *Castanopsis indica*) between 1400-1800 m. Fl. Sep-Oct. Panchase forest, Kaski, 1400-1800 m, P. Bhandari 1495 (KATH). [CE Nepal, W Himalaya, E Himalaya, S Asia, E Asia and SE Asia].

Oberonia falcata King & Pantl.

Epiphytic on tree trunks, branches and shrubs (*Daphniphyllum himalayense*, *Quercus lamellosa*, *Rhododendron arboreum*, *Berberis* species) between 1700-2300 m. Fl. Jul-Sep. Sidhane, Bhadaure Tamage VDC, Kaski, 1820 m, 09 August 2014, P. Bhandari & A. Bhandari P736 (KATH & TUCH). [CE Nepal, E Himalaya, E Asia and SE Asia].

Oberonia nepalensis L.R.Shakya & R.P.Chaudhary

Epiphytic on tree trunks (*Schima wallichii*, *Castanopsis indica*, *Ficus lacor*, *Albizia* species) between 900-1500 m. Fl. Mar-Apr. Panchase forest, Kaski, 1460 m, 22 March 2015, P. Bhandari & S. Budhamagar P1329 (KATH & TUCH). **Endemic to Central Nepal**. *Type*: *Kaski*, Pokhara (around Annapurna Conservation Zone), ca. 1100 m, L.R. Shakya, R.L. Singh & R.P. Chaudhary 27 (holotype: KATH!; isotype: TUCH!).

Oberonia obcordata Lindl.; *Malaxis obcordata* (Lindl.) Rchb.f.

Epiphytic on tree branches and trunks (*Lyonia ovalifolia*, *Schima wallichii*) between 1300-1600 m. Fl. Mar-Apr. Bhadaure Deurali, Kaski, 1600 m, 20 March 2015, P. Bhandari, S. Budhamagar & A. Bhandari P1272 (KATH & TUCH). [CE Nepal, E Himalaya, Tibetan Plateau, S Asia, E Asia and SE Asia].

Oberonia pachyrachis Rchb.f. ex Hook.f.

Epiphytic on tree trunks (*Schima wallichii*) between 1300-1600 m. Fl. Mar-Apr. Harpan, Kaski, 1360 m, 22 March 2015, P. Bhandari & S. Budhamagar P1332 (KATH & TUCH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Oberonia recurva Lindl.; *Oberonia parvula* King & Pantl.

Epiphytic on tree trunks (*Schima wallichii*) between 1300-1400 m. Fl. Mar-Apr. Harpan, Kaski, 1360 m, 22 March 2015, P. Bhandari & S. Budhamagar P1333 (KATH & TUCH). [C Nepal, E Himalaya and E Asia].

Odontochilus Blume (ca. 55/4/4 species)

Odontochilus crispus (Lindl.) Hook.f.; *Anoectochilus crispus* Lindl.

Terrestrial on *Daphniphyllum* forest floor at 1900 m. Fl. Aug-Sep. Panchase forest, Kaski, 1900 m, 17 August 2017, P. Bhandari, K. Shrestha, G. Thapamagar & D. Pathak 1006P (KATH). [CE Nepal, E Himalaya, Tibetan Plateau and E Asia].

Odontochilus elwesii C.B.Clarke ex Hook.f.

Terrestrial on the forest floor, near streamside (*Daphniphyllum* forest) at 1900 m. Fl. Jul-Aug. Panchase forest, Kaski, 1900 m, 20 July 2018, P. Bhandari P1702, KATH030406. [C Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Odontochilus lanceolatus (Lindl.) Blume; *Anoectochilus lanceolatus* Lindl.

Terrestrial on the forest floor and rock crevices (*Daphniphyllum himalayense*, *Quercus lamellosa*) between 1700-2100 m. Fl. Jul-Aug. Panchase forest, Kaski, 1700 m, 17 August 2017, P. Bhandari, K. Shrestha, G. Thapamagar & D. Pathak P1669 (KATH). [CE Nepal, E Himalaya, Assam-Burma, S Asia, E Asia and SE Asia].

Odontochilus nandae Raskoti & H.Kurzweil

Terrestrial on the forest floor (*Daphniphyllum* forest, Oak-*Rhododendron* forest) between 2100-2400 m. Fl. Jul-Aug. Panchase forest, Kaski, 2100-2400 m, 17 August 2017, P. Bhandari, K. Shrestha, G. Thapamagar & D. Pathak 1009P (KATH). **Endemic to Panchase**. *Type*: Kaski, Panchase, 2400 m, BB Raskoti 119 (holotype: KATH!).

Otochilus Lindl. (5/4/3 species)

***Otochilus albus** Lindl.

Epiphytic on tree trunks and lower branches (*Schima wallichii*, *Castanopsis indica*) between 1000-1700 m. Fl. Oct-Nov. [CE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Otochilus fuscus Lindl.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*) between 1700-1900 m. Fl. Dec-Jan. Bhadaure Deurali, Kaski, 1850 m, 08 January 2015, P. Bhandari & Y.P. Paudel P1252 (TUCH & KATH). [CE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Otochilus lancilabius Seidenf.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, Oak, *Castanopsis* species, *Schima wallichii*, *Lyonia ovalifolia*, *Rhus succedanea*, *Engelhardia spicata*, *Myrica esculenta*, *Diospyrus lancifolia*) between 900-2400 m. Fl. Dec-Jan. Panchase forest, Kaski, 2350 m, 13 December 1973, D.P. Joshi & M.M. Amatya 73/1171 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma and SE Asia].

Panisea (Lindl.) Lindl. (13/3/2 species)

Panisea demissa (D.Don) Pfitzer; *Dendrobium demissum* D.Don

Epiphytic on tree trunks and lower branches (*Daphniphyllum himalayense*, *Quercus lamellosa*) between 2100-2300 m. Fl. Sep-Oct. Panchase forest, Kaski, 2260 m, 05 October 2014, P. Bhandari & A. Bhandari P1221 (KATH & TUCH). [CE Nepal, E Himalaya, Assam-Burma, S Asia, E Asia and SE Asia].

Panisea panchaseensis Subedi

Epiphytic on tree trunks and lower branches (Laurels, *Quercus lamellosa*, *Viburnum erubescens*) between 2300-2450 m. Fl. Nov-Dec. Panchase forest, Kaski, 2300-2450 m, 14 December 2015, P. Bhandari & K. Shrestha P1710 m (KATH). [C Nepal and E Himalaya]. **Panchase is the type locality for this species**. *Type*: Kaski, Panchase forest, 2200-2450 m, A. Subedi 1780 (holotype: KATH n.v.).

Papilionanthe Schltr. (10/2/1 species)

Papilionanthe uniflora (Lindl.) Garay; *Mesoclastes uniflora* Lindl.

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*, *Ilex fragilis*) between 2000-2100 m. Fl. Sep-Oct. Panchase forest, Kaski, 2100 m, 05 October 2014, P. Bhandari & A. Bhandari P1236 (TUCH). [CE Nepal and E Himalaya].

Peristylus Blume (103/11/1 species)

Peristylus aristatus Lindl.

Terrestrial on grassy slopes and *Daphniphyllum* forest between 1600-2100 m. Fl. Aug-Sep. Panchase forest, Kaski, 1900 m, 07 September 2014, P. Bhandari & S. Budhamagar P788 (TUCH). [CE Nepal, E Himalaya and S Asia].

Phalaenopsis Blume (74/3/2 species)

***Phalaenopsis difformis** (Wall. ex Lindl.) Kocyan & Schuit.; *Aerides difformis* Wall. ex Lindl., *Ornithochilus difformis* (Wall. ex Lindl.) Schltr.

Epiphytic on tree trunks between 1500-1700 m. Fl. Jun-Jul. [CE Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

Phalaenopsis taenialis (Lindl.) Christenson & Pradhan; *Aerides taenialis* Lindl., *Kingidium taenialis* (Lindl.) P.F.Hunt

Epiphytic on tree trunks and branches (*Ficus neerifolia*, *F. lacor*, *F. glaberrima*) between 1400-1800 m. Fl. Apr-Jul. Bhadaure Deurali, Panchase, Kaski, 1650 m, 05 May 2018, P. Bhandari P1622 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Pholidota Lindl. (40/5/4 species)

Pholidota articulata Lindl.; *Coelogyne articulata* (Lindl.) Rchb.f.

Epiphytic on tree trunks and branches (*Quercus lamellosa*, *Daphniphyllum himalayense*, *Rhododendron arboreum*, *Alnus nepalensis*, *Engelhardia spicata*, *Ilex fragilis*, *Lindera pulcherrima*, *Schima wallichii*) between 1100-1800 m. Fl. Jul-Aug. Panchase forest, Kaski, 02 August 2014, A. Subedi, 973 (TUCH!). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

***Pholidota imbricata** Hook.

Epiphytic on *Schima-Castanopsis* forest between 1000-1800 m. Fl. Jul-Aug. [CE Nepal, E Himalaya, Assam-Burma, S Asia, E Asia and SE Asia].

Pholidota pallida Lindl.

Epiphytic on tree trunks and branches (*Castanopsis indica*, *Schima wallichii*, *Engelhardia spicata*, *Myrica esculenta*) or lithophytic between 1100-1400 m. Fl. Jun-Jul. Panchase forest, Kaski, 1200-1400 m, 06 June 2017, P. Bhandari & A. Bhandari P1665 (KATH). [WCE Nepal, E Himalaya, Assam-Burma, E Asia and SE Asia].

Pholidota protracta Hook.f.

Epiphytic on tree trunks (*Daphniphyllum himalayense*, *Rhododendron arboreum*, *Ilex* species) between 1900-2100 m. Fl. Sep-Oct. Panchase forest, Kaski, 2020 m, 05 October 2014, P. Bhandari & A. Bhandari P1218 (TUCH & KATH). [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Phreatia Lindl. (ca. 219/1/1 species)

Phreatia elegans Lindl.; *Thelasis elegans* (Lindl.) Blume

Epiphytic on tree trunks and branches (*Daphniphyllum himalayense*) between 1600-1700 m. Fl. Aug-Sep. Chapakot VDC, Panchase Protected Forest, Kaski, 1600 m, 09 August 2014, P. Bhandari & A. Bhandari P721 (TUCH & KATH). [C Nepal, E Himalaya, Tibetan Plateau, S Asia and SE Asia].

Pinalia Lindl. (ca. 160/18/8 species)

***Pinalia amica** (Rchb.f.) Kuntze; *Eria amica* Rchb.f.

Epiphytic on *Shorea robusta* forest within 900-1200 m. Fl. Mar-Apr. [C Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

***Pinalia apertiflora** (Summerh.) A.N.Rao; *Eria apertiflora* Summerh.

Epiphytic on *Schima-Castanopsis* and *Daphniphyllum* forest within 1400-1800 m. Fl. Jul-Aug. [WCE Nepal, E Himalaya, Assam-Burma and SE Asia].

Pinalia bipunctata (Lindl.) Kuntze; *Eria bipunctata* Lindl.

Epiphytic on tree trunks and branches (*Schima wallichii*, *Daphniphyllum himalayense*, *Engelhardia spicata*) between 1400-1800 m. Fl. Sep-Oct. Panchase forest, Kaski, 1500 m, 10 September 2014, P. Bhandari & S. Budhamagar P852 (TUCH & KATH). [C Nepal, W Himalaya, E Himalaya, E Asia and SE Asia].

***Pinalia bractescens** (Lindl.) Kuntze; *Eria bractescens* Lindl.
Epiphytic on *Schima-Castanopsis* and *Daphniphyllum* forest within 1400-1800 m. Fl. Jul-Aug. [CE Nepal, Assam-Burma and SE Asia].

Pinalia excavata (Lindl.) Kuntze; *Eria excavata* Lindl.
Epiphytic on tree trunks and branches (*Quercus semecarpifolia*, *Rhododendron arboreum*, *Daphniphyllum himalayense*) within 2300-2500 m. Fl. May-Jun. Panchase forest, Kaski, 2300 m, 02 August 2002, A. Subedi 957 (TUCH!). [C Nepal, E Himalaya and Tibetan Plateau].

Pinalia graminifolia (Lindl.) Kuntze; *Eria graminifolia* Lindl.
Epiphytic on branches (*Rhododendron arboreum*, *Quercus semecarpifolia*, *Lyonia ovalifolia*, *Daphniphyllum himalayense*) between 2300-2500 m. Fl. Jul-Aug. Panchase forest, Kaski, 2500 m, 10 August 2014, P. Bhandari & A. Bhandari P757 (TUCH & KATH). [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma and E Asia].

Pinalia pokharensis (Bajrach., Subedi & K.K.Shrestha) Schuit., Y.P.Ng & H.A.Pedersen, *Eria pokharensis* Bajrach., Subedi & K.K.Shrestha
Epiphytic on tree trunks (*Daphniphyllum himalayense*, *Lindera pulcherrima*, *Quercus lamellosa*) between 2000-2100 m. Fl. Jun-Jul. Panchase forest, Kaski, 2020 m, 06 June 2017, P. Bhandari & A. Bhandari P1666 (KATH). **Endemic to Central Nepal**. *Type*: *Kaski*, Lumle, Pokhara, 900-1000 m, DM Bajracharya & A. Subedi CN220 (holotype: KATH!).

Pinalia spicata (D.Don) S.C.Chen & J.J.Wood; *Octomeria spicata* D.Don, *Eria spicata* (D.Don) Hand.-Mazz.
Epiphytic on tree trunk and branches (*Schima wallichii*, *Castanopsis indica*, *Daphniphyllum himalayense*, *Bombax ceiba*, *Diospyrus lancifolia*) between 900-2200 m. Fl. Jul-Aug. Panchase forest, Kaski, 1500-2000 m, 14 August 2017, P. Bhandari, A. Bhandari, K. Shrestha, G. Thapamagar & D. Pathak 1001P (KATH). [C Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Platanthera Rich. (123/13/1 species)

Platanthera biermanniana (King & Pantl.) Kraenzl.; *Habenaria biermanniana* King & Pantl.
Terrestrial on grassy slopes, edges of Oak forest between 2300-2400 m. Fl. Sep-Oct. Panchase forest, Kaski, 2300-2400 m, 08 October 2017, P. Bhandari, G. Thapamagar & D. Pathak P1704 (KATH). [CE Nepal and E Himalaya].

Pleione D.Don (24/5/2 species)

Pleione humilis (Sm.) D.Don; *Epidendrum humile* Sm.
Epiphytic on tree trunks, lower and middle branches (*Daphniphyllum himalayense*, Oak, *Lyonia ovalifolia*, *Rhododendron arboreum*, Laurel, *Ilex fragilis*, *Symplocos ramosissima*, *Viburnum erubescens*) between 2100-2500 m. Fl. Jan-Mar. Panchase forest, Kaski, 2500 m, 09 January 2015, P. Bhandari & Y.P. Paudel P1257 (TUCH & KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia and SE Asia].

Pleione praecox (Sm.) D.Don; *Epidendrum praecox* Sm.
Epiphytic on tree trunks, lower and middle branches (*Daphniphyllum himalayense*, Oak, *Rhododendron arboreum*, *Lyonia ovalifolia*, Laurel, *Viburnum erubescens*) between 2100-2500 m. Fl. Sep-Nov. Panchase forest, Kaski, 2300 m, November 2002, A. Subedi, R.P. Chaudhary & L.R. Shakya 996 (TUCH!). [CE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, S Asia, E Asia and SE Asia].

Podochilus Blume (65/1/1 species)

Podochilus cultratus Lindl.
Epiphytic on tree trunks and lower branches (*Schima wallichii*, *Castanopsis indica*) between 900-1000 m. Fl. Jul-Aug. Makwanpur to Ghatichida, Panchase forest, Kaski, 900-1000 m, 24 August 2016, P. Bhandari P1613 (KATH). [CE Nepal, E Himalaya, Assam-Burma and SE Asia].

Porpax Lindl. (53/4/2 species)

Porpax elwesii (Rchb.f.) Rolfe; *Eria elwesii* Rchb.f.
Epiphytic on tree trunks and lower branches (*Schima wallichii*, *Castanopsis indica*) between 900-1000 m. Fl. Jun-Jul. Below Damdame, Panchase, Kaski, 900-1000 m, 22 July 2018, P. Bhandari P1630 (KATH). [CE Nepal, E Himalaya, E Asia and SE Asia].

Porpax muscicola (Lindl.) Schuit, Y.P.Ng & H.A.Pedersen; *Dendrobium muscicola* Lindl., *Conchidium muscicola* (Lindl.) Rauschert, *Eria muscicola* (Lindl.) Lindl.
Epiphytic on tree trunks (*Daphniphyllum himalayense*, *Schima wallichii*, *Lyonia ovalifolia*, *Myrica esculenta*) or lithophytic between 1300-1500 m. Fl. Jul-Aug. Bhadaure Deurali, Kaski, 1500 m, 10 September 2014, P. Bhandari & S. Budhamagar P854 (KATH & TUCH). [CE Nepal, W Himalaya, E Himalaya, Assam-Burma, S Asia, E Asia and SE Asia].

Rhynchostylis Blume (5/1/1 species)

Rhynchostylis retusa (L.) Blume; *Epidendrum retusum* L.
Epiphytic on tree trunks and branches (*Castanopsis indica*, *Ficur lacor*, *Ficus religiosa*, *Diospyrus lancifolia*) between 1000-1800 m. Fl. Jun-Aug. Harpan, Panchase, Kaski, 1340 m, 22 March 2015, P. Bhandari & S. Budhamagar P1334 (TUCH). [WCE Nepal, W Himalaya, E Himalaya, Assam-Burma, S Asia, E Asia and SE Asia].

Satyrium Sw. (91/1/1 species)

Satyrium nepalense D.Don var. **nepalense**
Terrestrial on grassy slopes, sometimes epiphytic (*Daphniphyllum himalayense*, *Quercus semecarpifolia*, *Symplocos ramosissima*, *Viburnum erubescens*) between 2200-2500 m. Fl. Aug-Oct. Panchase forest, Kaski, 2280-2500 m, 09 September 2014, P. Bhandari & S. Budhamagar P752 (TUCH & KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, S Asia and E Asia].

Spiranthes Rich. (37/2/2 species)

Spiranthes australis (R.Br.) Lindl.; *Neottia australis* R.Br.; *Spiranthes sinensis* var. *amoena* (M.Bieb.) H.Hara

Terrestrial on grassy slopes, edges of *Daphniphyllum* forest between 1800-2200 m. Fl. Aug-Oct. Panchase forest, Kaski, 1800-2200 m, 07 September 2014, P. Bhandari & S. Budhamagar P789 (TUCH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, Assam-Burma, E Asia, SE Asia, N Asia, SW Asia and Australasia].

Spiranthes flexuosa (Sm.) Lindl.; *Neottia flexuosa* Sm.

Terrestrial on grass slopes and *Schima-Castanopsis* forest edges between 1300-1700 m. Fl. Apr-May. Between Damdame to Sidhane, Panchase, Kaski, 1300-1400 m, 06 May 2018, P. Bhandari P1619 (KATH). [C Nepal, SW Asia and Europe].

Taeniophyllum Blume (241/1/1 species)

Taeniophyllum scaberulum Hook.f.

Epiphytic on tree trunks (*Daphniphyllum himalayense*, *Ilex excelsa*) between 1700-2000 m. Fl. Jul-Aug. Panchase forest, Kaski, 1940 m, 21 March 2015, P. Bhandari, A. Bhandari & S. Budhamagar P1286 (KATH). [C Nepal and S Asia (S India)].

Tainia Blume (29/1/1 species)

Tainia minor Hook.f.

Terrestrial on *Daphniphyllum* forest floor between 1900-2100 m. Fl. May-Jun. Panchase forest, Kaski, 1900 m, 02 June 2014, P. Bhandari, S. Budhamagar & C.K. Thakur, P441 (KATH). [CE Nepal, E Himalaya, Tibetan Plateau, Assam-Burma and E Asia].

Thunia Rchb.f. (5/1/1 species)

Thunia alba (Lindl.) Rchb.f.; *Phaius albus* Lindl.

var. **bracteata** (Roxb.) N.Pearce & P.J.Cribb; *Limodorum bracteatum* Roxb.

Epiphytic on tree trunks (*Ficus neerifolia*, *Ficus lacor*, *Eurya acuminata*, *Castanopsis indica*, *Diospyrus lancifolia*, *Schima wallichii*) between 900-1800 m. Fl. Jul-Aug. Tarebhir, Pumdi Bhumdi VDC, Kaski, 1460 m, 08 August 2014, P. Bhandari & A. Bhandari P685 (TUCH). [C Nepal, E Himalaya, SE Asia].

Vanda Jones ex R.Br. (81/7/1 species)

Vanda cristata Wall. ex Lindl.; *Aerides cristata* (Wall. ex Lindl.) Wall. ex Hook.f.

Epiphytic on tree trunks and branches (*Schima wallichii*, *Ficus lacor*, *Prunus cerasoides*, *Daphniphyllum himalayense*, *Alnus nepalensis*, *Albizia* species, *Castanopsis indica*, *Diospyrus lancifolia*, *Engelhardia spicata*, *Ilex excelsa*, *Phoebe* species) between 900-1900 m. Fl. May-Jun. Panchase forest, Kaski, 1510 m, 04 June 2014, P. Bhandari, S. Budhamagar & R. Chhetri P511a (TUCH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau, S Asia, E Asia and SE Asia].

Vandopsis Pfitzer (4/1/1 species)

Vandopsis undulata (Lindl.) J.J.Sm.; *Vanda undulata* Lindl.

Epiphytic on trunks and branches (*Daphniphyllum himalayense*, *Quercus semecarpifolia*, *Berberis* scrubs) between 1800-2400 m. Fl. Apr-Jun. Bhanjyang to Panchase, Kaski, 2100-2400 m, 06 May 2018, P. Bhandari P1627 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Tibetan Plateau and E Asia].

Zeuxine Lindl. (76/10/4 species)

Zeuxine affinis (Lindl.) Benth. ex Hook.f.; *Monochilus affinis* Lindl.

Terrestrial on *Schima-Castanopsis* forest floor between 900-1100 m. Fl. Mar-Apr. Panchase forest, Kaski, 900-1100 m, 17 March 2018, P. Bhandari P1700 (KATH). [C Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

Zeuxine flava (Wall. ex Lindl.) Trimen; *Monochilus flavus* Wall. ex Lindl.

Terrestrial on *Schima-Castanopsis* forest floor between 900-1100 m. Fl. Feb-Mar. Panchase forest, Kaski, 900 m, 17 March 2018, P. Bhandari P1701 (KATH). [CE Nepal, W Himalaya, E Himalaya, Assam-Burma, E Asia and SE Asia].

Zeuxine longilabris Trimen

Terrestrial on *Schima-Castanopsis* forest floor between 900-1000 m. Fl. Apr-May. Ghatichida, Panchase, Kaski, 900-1000 m, 06 May 2018, P. Bhandari P1618 (KATH). [C Nepal, Assam-Burma and S Asia].

Zeuxine strateumatica (L.) Schltr.; *Orchis strateumatica* L.

Terrestrial on grassy slopes, near water bodies between 900-1000 m. Fl. Feb-Mar. Ghatichida, Panchase, Kaski, 900-1000 m, 14 February 2017, P. Bhandari P1603 (KATH). [WCE Nepal, W Himalaya, E Himalaya, Assam-Burma, S Asia, E Asia, SE Asia, SW Asia, S America and Australasia].

***Note**: '*' denote the species cited from Subedi* et al. *(2007).*

Format of Checklist

Genus (species number distributed worldwide/Nepal/ Panchase)

Species; *Basionym, Synonyms*

Habit/Habitat, Ecology (forest types, Host), Altitude. **Endemism** (if endemic to Nepal). Flowering month. Representative collection. [Broad distribution]. Type (if endemic to Panchase).

Conclusion

This paper attempted to revise the species status on orchids of Panchase and reported 143 orchid taxa, with three endemic to the forest. Representing, nearly one third orchid species (28%) of Nepal, Panchase can be regarded as a hotspot of orchid diversity in Nepal and the forest should be conserved in the form of 'Panchase Orchid Sanctuary'.

The way forward

Orchid explorations have been frequently carried out in Panchase but the research on the spatial and temporal distribution of orchid, the ecosystem services, their conservation and threat status in Panchase are lacking. The fulfilment of these gaps could give compelling information on orchids of Panchase and Central Nepal.

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References

Acharya, K.P., Vetaas, O.R. & Birks, J.B. (2011). Orchid species richness along Himalayan elevational gradients. *Journal of Biogeography*, *38*, 1821-1833.

Aryal, A. & Dhungel, S.K. (2009). Species diversity and distribution of bat in Panchase region of Nepal. *Conservation and Information Network of South Asia*, *1*(1), 11-14.

Bajracharya, D.M., Subedi, A. & Shrestha, K.K. (2003). *Eria pokharensis*, sp. nov. (Orchidaceae); A new species from Nepal Himalaya. *Journal of the Orchid Society*, *17*(1-2), 1-4.

Baral, R. (2018). Altitudinal diversity of birds in Panchase Protected Forest, Nepal. *Journal of Ecosystem and Ecography*, *8*, 24. 10.4172/2157-7625-C4-041

Bhandari, P. & Shrestha, K.K. (2016). *Dischidia bengalensis* (Apocynaceae), a New Record for the Flora of Nepal. *The Journal of Japanese Botany*, *91*(5), 310-313.

Bhandari, P. (2015). *Flora of Panchase Protected Forest, Kaski District (Central Nepal)*. (Master's Thesis). Central Department of Botany, Tribhuvan University, Kirtipur, Kathmandu.

Bhandari, P., Budhamagar, S. & Shrestha, K.K. (2016-18). A Checklist of Flowering Plants of Panchase Protected Forest, Kaski District, Central Nepal. *Journal of Natural History Museum*, *30*, 55-84.

Bhandari, P., Karki, S. & Budhamagar, S. (2016a). A new locality record for the endemic *Panisea panchaseensis* Subedi (Orchidaceae) in Nepal. *Pleione*, *10*(1), 183-185.

Bhandari, P., Raskoti, B.B. & Shrestha, K.K. (2015). *Pheretia elegans* Lindley (Orchidaceae)- a new record for the flora of Nepal. *Pleione*, *9*(2), 365-368.

Bhandari, P., Shakya, L.R. & Chaudhary, R.P. (2019a). *Zeuxine lindleyana* (Orchidaceae) - A New Record for the Flora of Nepal. *The Journal of Japanese Botany*, *94*(1), 45-46.

Bhandari, P., Shakya, L.R. & Shrestha, K.K. (2016b). *Zeuxine membranacea* (Orchidaceae) - An Addition to the Orchid Flora of Nepal. *The Journal of Japanese Botany*, *91*(5), 317-320.

Bhandari, P., Shrestha, K., KC, A. & Shrestha, K.K. (2020). Three New Records of Orchids from Nepal. *The Journal of Japanese Botany*, *95*(1), 41-46.

Bhandari, P., Uprety, Y. & Shakya, L.R. (2019b). *Platanthera carnosilabris* (Orchidaceae), an Addition to the Flora of Nepal. *The Journal of Japanese Botany*, *94*(1), 42-44.

Dixit, A., Karki, M. & Shukla, A. (2015). *Vulnerability and Impacts Assessment for Adaptation Planning in Panchase Mountain Ecological Region, Nepal*, Kathmandu, Nepal: Government of Nepal, United Nations Environment Programme, United Nations Development Programme, International Union for Conservation of Nature, German Federal Ministry for the Environment, Nature Conservation, Building and Nuclear Safety and Institute for Social and Environmental Transition-Nepal.

Govaerts, Bernet, P., Kratochvil, K., Gerlach, G., Carr, G., Alrich, P., Pridgeon, A.M., Pfahl, J., Campacci, M.A., Baptista, D.H., Tigges, H., Shaw, J., Cribb, P., George, A., Kreuz, K. & Wood, J. (2019). *World Checklist of Orchidaceae*. UK: The Royal Botanic Gardens, Kew. http://wcsp.science.kew.org/ Retrieved 14 December 2019.

Malla, D.B., Aryal, A. & Kuwor, K.J. (2013). Distribution and food resources of Asiatic black bear (*Ursus thibetanus*) and human-bear conflict in the Panchase Protected Forest of Nepal. *International Bear News*, *22*(2), 33-35.

Mikage, M., (2008). Field Research on the Medicinal Plant Resources around the Tamage Village, Kaski District, West Nepal. *Newsletter of Himalayan Botany No. 27*, Tokyo: The Society of Himalayan Botany.

Odyuo, N., Deori, C. & Daimary, R. (2016). *Panisea panchaseensis* (Orchidaceae): an addition to the flora of India, *Richardiana*, *16*, 141-145.

Rajbhandari, K.R. & Rai, S.K. (2017). *A Handbook of the Flowering Plants of Nepal*, Vol. 1. Kathmandu, Nepal: Department of Plant Resources.

Raskoti, B.B. & Ale, R. (2013). Four New Additions to the Orchid Flora of Nepal. *The Journal of Japanese Botany*, *88*, 378-382.

Raskoti, B.B. & Ale, R. (2019a). Molecular phylogeny and morphology reveal a new epiphytic species of *Habenaria* (Orchidaceae; Orchideae; Orchidinae) from Nepal. *PLoS ONE*, *14*(10), e0223355. https://doi.org/10.1371/journal.pone.0223355.

Raskoti, B.B. & Ale, R. (2019b). New species of orchids and notes on Orchidaceae of Nepal. *Phytotaxa*, *394*(4), 257-266.

Raskoti, B.B. & Jin, X-H. (2013). *Herminium alaschanicum* and *Herminium souliei* (Orchidaceae) – Two New Records for the Flora of Nepal. *The Journal of Japanese Botany*, *88*, 383-385.

Raskoti, B.B. & Kurzweil, H. (2015). *Odontochilus nandae* (Orchidaceae; Cranichidae; Goodyerinae), a new species from Nepal. *Phytotaxa*, *233*(3), 293-297.

Rastoki, B.B. (2015). A new species of *Gastrochilus* and new records for the orchids of Nepal. *Phytotaxa*, *233*(3), 179-184.

Rokaya, M.B., Raskoti, B.B., Timsina, B. & Münzbergová, Z. (2013). An annotated checklist of the orchids of Nepal. *Nordic Journal of Botany*, *31*, 511-550.

Shrestha, K.K., Bhattarai, S. & Bhandari, P. (2018). *Handbook of Flowering plants of Nepal*, vol. 1. Jodhpur, India: Scientific Publishers.

Subedi, A., Chaudhary, R.P., Vermeulen, J.J. & Gravendeel, B. (2011). *Panisea panchaseensis* sp. nov. (Orchidaceae) from central Nepal. *Nordic Journal of Botany*, *29*, 361-365.

Subedi, A., Subedi, N. & Chaudhary, R.P. (2007). Panchase Forest: an extraordinary place for wild orchids in Nepal. *Pleione*, *1*, 23-31.

Subedi, C.K., Bhandari, P., Thapa, S., Pandey, M. Gurung, J., Shakya, L.R. & Chaudhary, R.P. (2018). *Cephalanthera erecta* var. *oblanceolata* (Orchidaceae) - A New Record for the Flora of Nepal. *The Journal of Japanese Botany*, *93*(4), 287-290.

Thakur, C. & Rajbhandary, S. (2018). Fern and fern allies of Panchase Protected Forest, Central Nepal. *Journal of Plant Resources, 16*(1), 39-45.

Watson, M.F., Akiyama, S., Ikeda, H., Pendry, C., Rajbhandari, K.R. & Shrestha, K.K. (Eds.) (2011). *Flora of Nepal.* Vol. 3. UK: The Royal Botanic Garden, Edinburgh.

World Wide Fund for Nature. (2013). *Chitwan-Annapurna Landscape Biodiversity Areas and Linkages*. Hariyo Ban Program, Kathmandu, Nepal: Author.

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Diversity of Phorophytes Selected by Epiphytic Orchid *Vanda cristata* Wall. ex Lindl. (Orchidaceae) in Central Nepal

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Abstract

Epiphytic orchids, which live non-parasitically on another plant (host or phorophyte) are the important component of global plant diversity, and occurdiversely in tropical and sub-tropical forest of Nepal. However, little is known about the specificity of the relationship between epiphytic orchids and their host tree species in Nepal. Host specificity relationship was assessed for the epiphytic orchid *Vanda cristata* Wall. ex Lindl. in tropical and sub-tropical forest of Jugu, Central Nepal. To record the local abundance and geographical distribution, a rating system was used. The result showed that majority of orchid-host associations were locally sparse and restricted geographically (42.10 %), followed by locally sparse and widespread (21.05 %). *Vanda cristata* was seen as generalist species because it was found colonized on 19 different host tree species (phorophytes). Thus, non-specific nature of association of this orchid with host trees was detected.

Keywords: Epiphytes, Host species, Host specificity

Introduction

Epiphytes, which contribute about 10 percent of total vascular species (Kress, 1986) are diverse and important component of global plant diversity. They live on another plant (host plant or phorophytes) for the structural support and anchorage, thus known as structurally dependent plants (Wagner et al., 2015). Epiphytic orchids, which belongs to the family Orchidaceae, includes some threatened species and encompass up to 70% of total vascular epiphytic species (Madison, 1977), occur diversely in tropical and subtropical forest of the world (Nieder et al., 2001).

Epiphytes are broadly classified into two groups: holo-epiphytes (spending their entire life on the host tree) and hemi-epiphytes (spending only part of their life cycle on host tree) (Benzing, 1990; Nieder et al., 2001; Bhatt et al., 2015). Epiphytic orchids are vascular holo-epiphytes. Due to complex habit, perfect suit of environment is necessary for their survival and growth. Therefore, to adapt in epiphytic habit, they have developed some special morphological and physiological adaptations. For example, succulent leaves and stems, pseudobulb as storage organ, sunken stomata, CO_2 uptake via. CAM pathway, roots with velamen redicum and impermeable cuticles, which help them to adapt successfully even in the poor nutrient and water supply conditions and led them to an outstanding success in epiphytic habitat (Zotz & Winkler, 2013).

Epiphytic orchids constitute an important position in the flora of Nepal. So far, 256 species of epiphytic orchids have been discovered from the country, and still are in the process of documentation (Rajbhandari & Rai, 2017; Shrestha et al., 2018; Raskoti & Ale, 2019). They are distributed from 60 m to 5200 m asl in Nepal (Rokaya et al., 2013), growing on various habitats and microclimatic conditions. Most of the epiphytic orchids occur in tropical and sub-tropical forests of the country (Acharya et al., 2011).

Studies on host specificity analysis are very few in the Nepalese context. In other parts of the world, studies have shown that epiphytic orchids have host preference and that depends on the host traits (Callaway et al., 2002; Merwin et al., 2003). It has been suggested that successful seedling establishment on a phorophyte depends on biotic and abiotic factors like tree bark roughness, host size, bark porosity, height, bark chemistry (e.g., pH, plant exudates) and suitable mycorrhizae (Johanson, 1974; Callaway et al., 2002; Trapnell & Hamrick, 2006;

Adhikari & Fischer, 2011). Trapnell & Hamrick (2006) suggested that presence of suitable mycorrhizae is the most critical factor because orchid seed lacks endosperm and requires suitable mycorrhizal infection for carbon uptake before germination. This study is aimed to assess the diversity of phorophytes colonized by epiphytic orchid *Vanda cristata* in tropical and sub-tropical forest of Central Nepal.

Materials and Methods

Study species

Vanda cristata is the perennial epiphytic orchid first collected in 1818 AD by Nathaniel Wallich in Nepal and described by John Lindley in 1832. It is distributed in Bhutan, Nepal, India, N. Vietnam, Thailand, Myanmar, Bangladesh and China (geographical distribution) (Roskov et al., 2019). In Nepal, it is distributed in Central and Eastern Nepal from 920 m to 2300 m (Rajbhandari & Rai, 2017). It is monopodial epiphytic orchid (holo-epiphyte) with yellowish-green fragrant flowers (Figure 1). Single inflorescence can produce 1 to 3 flowers and usually 1 or rarely 2 healthy pods. Flowers are thickly textured with wide openings. Petals are falcate, lip golden yellow to white, three-lobed and spurred. This species loves middle and lower trunks of host trees. Flowering time of *V. cristata* is from May to July.

Figure 1. Study species *Vanda cristata* Wall. ex Lindl.

Vanda cristata Wall. ex Lindl. has medicinal importance. The paste prepared from the root is used to treat dislocated bones, cut and wounds and leaf powder as expectorant (Manandhar, 2002; Subedi et al., 2013). Similarly, the plant bear attractive flowers, so used as ornamental plant in home gardens. Thus, concerning the diverse use-values, this species is recognized asone of the most important orchid species of Nepal.

Study area

The study was carried out in tropical and sub-tropical forest of Jugu (now Gaurishankhar Rural Municipality) Central Nepal, with coordinate between 27º 53' N to 27.88º N latitudes and 86º 24' E to 86.40º E longitudes (Figure 2). The climate ranges from warm tropical, subtropical monsoon and cool temperate. There is high rainfall during monsoon period (June to September) and account for about 80 percent of the total annual rainfall. According to the report of the nearest Jiri weather station, the area receive an average precipitation of 2427.817 mm. The annual mean minimum and maximum temperature are 8.17°C and 20.71°C, respectively (Karki, 2019).

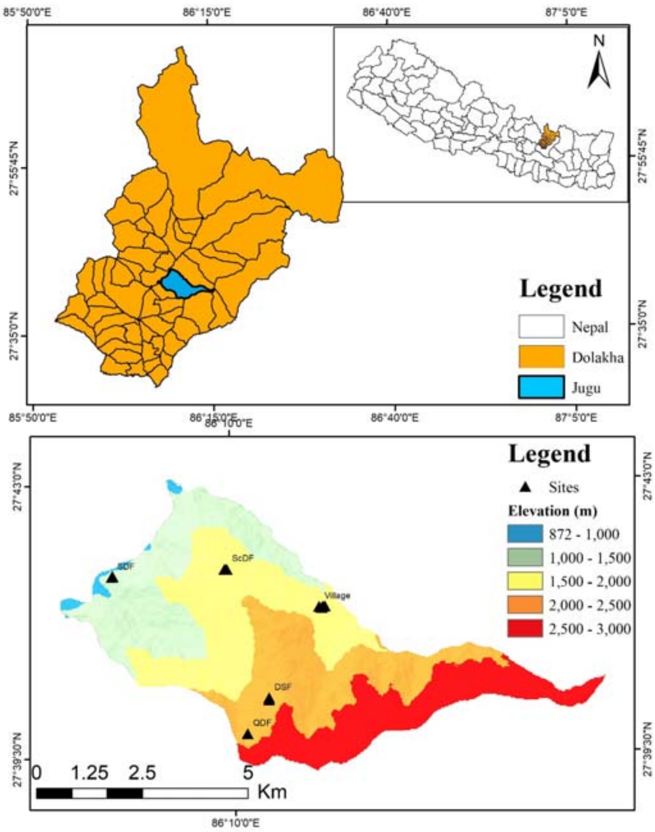

Figure 2: Map of Dolakha district showing study area

Data collection

The study was based on both repeated field surveys and literature reviews. Observations of different forest types (e.g., sal forest, riverine forest, *Schima-Castanopsis* forest and mixed pine forest) of Jugu were systematically made. The population of epiphytic orchid *Vanda cristata* along with their host trees and locations were recorded. The variety of phorophyte species colonized by *Vanda cristata* was photographed and identification of host species was made. The family of each host species was also identified.

In each site, the number of host individuals supporting *Vanda cristata* was recorded. A method known as rating system (Trapnell & Hamrick, 2006), similar to that of Rabinowitz (1981), was used to account the local abundance of host trees supporting *V. cristata* and geographical distribution of that orchid-host association (Table 1). In this method, based on the abundance of epiphytic orchid on host trees, theirlocal status was categorized into three different categories viz. locally abundant (A), locally sparse (S) and locally occasional (O). For occurrence of *V. cristata* on a particular tree species to be rated as 'A'; several to many individuals of that tree species had to support *V. cristata* at each site. A rating 'S' indicates that, if only few individuals of that tree species support *V. cristata* at each site. Similarly, rating 'O' was given in cases where some locations had less than three trees with orchids, while other locations had many trees of that species supporting *V. cristata* (Trapnell & Hamrick, 2006). Similarly, following Trapnell & Hamrick (2006), the geographic distribution of trees having *V. cristata* was recorded for each host/ phorophyte species as widespread (W) or restricted (R). A rating 'W' indicated that, the host tree has wide distribution, whereas rating 'R' indicated the narrow distribution of host species in the study area.

Results and Discussion

Vanda cristata was found colonized on 19 different host species (Table 2). This shows *V. cristata* is not host-specific orchid species. Although, this number is not exact because many areas were not systematically studied. Most common host species colonized by *V. cristata* was *Prunus cerasoides* (Rosaceae). However, *V. cristata* was also frequently observed on *Shorea robusta, Ficus nerifolius* and *Schima wallichii*. The association of *Vanda cristata* with multiple host species indicated that, it has high ability to establish population on several phorophytes, which is a good emblem of long-term survival potential of this species.

These results concur with the distribution of another epiphytic orchid species *Rhynchostylis retusa*, which is morphologically similar with *V. cristata* (Adhikari & Fischer, 2011). In many host species, *V. cristata* was observed at only once in the study sites. Although, there was good abundance of other trees (*Pinus roxburghii, Dalbergia sissoo* and *Myrica esculenta*) in the study sites, but we didn't find the association of *V. cristata* on those tree species. It seems unlikely that seed of *V. cristata* doesn't fall on the trunk of those trees, but it is predictable that such trunk lack appropriate substrate and mycorrhizae for the germination of *V. cristata* seedlings.

The result showed that the abundance of *V. cristata* on host trees at locality were locally sparse (63.15%), of which majority of orchid-host associations were locally sparse and restricted geographically (8 species, 42%), followed by locally sparse and widespread (4 species, 21.05%) (Table 2).Twelve associations were seen as locally sparse (63.15%), of which four (21%) were widespread and eight (42.10%) were recorded within restricted geographical range (Table 1 and 2). Similarly, three species have restricted and occasional distribution (15.78%) and two species have wide spread and locally abundant distribution (10.52%) (Table 1). This indicates, although *V. cristata* have wide range of host species, the abundancewas locally sparse and distribution was restricted to few sites only. Sparse local abundance of *V. cristata* in some sites was due to the sparse occurrence of host tree species (For example: *Ficus carica*, *F. sarmentosa* and *Fraxinus floribunda*). These findings agree with the findings of Trapnell & Hamrick, (2006) on the host-orchid associations of neotropical orchid *Laelia rubescens*.

However, in our study locally abundant *V. cristata* also have restricted geographical distribution (5.26 %), which differ from previous result of Trapnell & Hamrick, (2006).

In addition, it was observed that *V. cristata* was also found in degraded habitat. Thus, it is predictable that *Vanda cristata* can tolerate some level of disturbances and pollution. *Vanda cristata* has large, long roots and thick leaves, which might be advantageous to tolerate some degree of disturbances.In general, our study supports the previous findings, such as large-sized trees supports more species, high bark rugosity supports many species (Callaway et al., 2002; Hirata et al., 2008). Regardless, many individuals were seen adapted successfully even on trees with less rugose bark and on branches of small size. In some trees, even small twig (less than 10 cm diameter) also supported many individuals. It indicate that, in particular site, host bark rugosity and host size are less important for the occurrence of *V. cristata*. Other various microclimatic factors might be responsible for their occurrence in those sites.

In most of the study sites, *V. cristata* was recorded in the lower and middle trunk zones of host trees forming a patchy populations, covering the host trunk by their large roots, indicating that these layers are suitable for the growth. The reason behind the successful establishment of *V. cristata* on lower layer of host tree might be due to high substrate stability and moisture in lower trunk layers compared to upper layers. They are normally associated with other epiphytic orchid species, such as *Vandopsi sundulata*, *Oberonia pachyrachis*, *Pholidota articulata* and *Dendrobium heterocarpum* and hemi-parasitic species, such as *Hoya linearis, Lepisorus* sp., *Scrrula elata*, and *Drynaria mollis*.

Table 2: Phorohyte species of epiphytic orchid *Vanda cristata* Wall. ex Lindl.

S.N	Phorophyte	Family	Occurrence of *Vc* association
1.	*Acacia catechu*	Fabaceae	RS
2.	*Alnus nepalensis*	Betulaceae	WS
3.	*Buddleja asiatica*	Scrophulariaceae	RO
4.	*Engelhardia spicata*	Juglandaceae	WS
5.	*Euphorbia royelena*	Euphorbiaceae	RS
6.	*Eurya acuminata*	Theaceae	WS
7.	*Ficus carica*	Moraceae	RS
8.	*Ficus nerifolius*	Moraceae	RS
9.	*Ficus religiosa*	Moraceae	WO
10.	*Ficus sarmentosa*	Moraceae	RS
11.	*Fraxinus floribunda*	Oleaceae	RS
12.	*Juglans regia*	Juglandaceae	WS
13.	*Mangifera indica*	Anacardiaceae	RO
14.	*Prunus cerasoides*	Rosaceae	WA
15.	*Quercus glauca*	Fagaceae	RS
16.	*Sarauria napaulensis*	Saurauriaceae	RO
17.	*Schima wallichii*	Theaceae	WA
18.	*Shorea robusta*	Dipterocarpaceae	RA
19.	*Wendlandia heynei*	Rubiaceae	RS

Note: RS = Restricted, locally sparse, WS = Widespread, locally sparse, RO = Restricted, locally occasional, WO = Widespread, locally occasional, WA = Widespread, locally abundant, RA = Restricted, locally abundant

Conclusion

From the result, *Vanda cristata* showed generalist behavior and tend to establish population on more than single host species rather than on a particular

Table 1: Frequency of epiphytic orchid *Vanda cristata* (*Vc*) found on phorophytes locally and over the orchid's geographical range in forest of Central Nepal. Percentages are of 19 host tree species (see table 2) supporting *Vc* in each classification.

Abundance of *Vc* on phorophytes at locality	Geographical range of Phorophytes/ *Vc* association	
	Widespread (W)	Restricted (R)
Locally abundant (A)	*Vc* on many trees locally. Association with phorophyte widespread geographically. (10.52 %)	*Vc* on many trees locally. Association with phorophyte restricted geographically to one or few sites. (5.26%)
Locally occasional (O)	*Vc* on varying numbers of trees locally. Association with phorophyte widespread geographically. (5.26 %)	*Vc* on varying numbers of trees locally. Association with phorophyte restricted geographically to one or few sites. (15.78 %)
Locally sparse (S)	*Vc* on one to a few trees locally. Association with phorophyte widespread geographically. (21.05 %)	*Vc* on one to a few trees locally. Association with phorophyte restricted geographically to one or few sites. (42.10%)

host species. Therefore, it seems that the future perpetuation potential of this species is high, although orchid-host associations were locally sparse and restricted geographically in the study area. Host preference was biased towards the host trees having thick, rugose bark and tree withlarge size. However, different underlying mechanisms might be there for the successful host-epiphyte associations and host preference. Therefore, future research work should use a comprehensive approach for determining host traits, the role of mycorrhizae and microclimatic conditions in order to give some rigorous conclusions about the host specificity.

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References

Acharaya, K.P., Vetaas O.R. & Birks H.J.B. (2011). Orchid speciesrichness along Himalayan elevational gradients. *Journal of Biogeography*, *38*(9), 1821–1833.

Adhikari, Y.P. & Fischer, A. (2011). Distribution pattern of the epiphytic orchid Rhynchostylisretusa under strong human influence in Kathmandu valley, Nepal. *Botanica Orientalis- Journal of Plant Science*, *8*, 90–99.

Benzing, D.H. (1990). *Vascular Epiphytes*. Cambridge, UK: Cambridge University Press.

Bhatt, A., Gairola, S., Govender, Y., Baijnath, H. & Ramdhani, S. (2015). Epiphyte diversity on host trees in an urban environment, eThekwini Municipal Area, South Africa. *New Zealand journal of botany*, *53*(1), 24–37.

Callaway, R.M., Reinhart, K.O., Moore, G.W., Moore, D.J. & Pennings, S.C. (2002). Epiphyte host preferences and host traits: mechanisms for species-specific interactions. *Oecologia*, *132*,221–230.

Gentry, A.H. & Dodson,C.H. (1987). Diversity and biogeography of neotropical vascular epiphytes. *Ann. Missouri Bot. Gard*, *74*, 205–233.

Hirata, A., Kamijo, T. & Saito, S. (2008). Host trait preferences and distribution of vascular epiphytes in a warm temperate forest. *Plant Ecology*, *201*, 247–254.

Johansson, D. (1974). Ecology of vascular epiphytes in West African rain forest. *Acta Phytogeographica Suecica*, *59*, 1–123.

Karki, (2019). *Diversity, Distribution and Ethnobotany of Epiphytic Orchids in Jugu, Dolakha, Central Nepal*. (Master's Dissertation). Central Department of Botany, Tribhuvan University, Kirtipur, Kathmandu, Nepal.

Kress, W.J. (1986). The systematic distribution of vascular epiphytes: an update. *Selbyana*, *9*, 2–22.

Madison, M. (1977). Vascular epiphytes: their systematic occurrence and salient features. *Selbyana*, *2*, 1–13.

Manandhar, N.P. (2002). *Plants and people of Nepal.* Oregon, USA: Timber Press Portland.

Merwin, M.C.,Rentmeester, S.A. & Nadkarni, N. M. (2003). The influenceof host trees species on the distribution of epiphytic bromeliads in experimental monospecific plantations, LaSelva, Costa Rica. *Biotropica*, *35*, 37–47.

Nieder, J., Prosperi, J. & Michaloud, G. (2001). Epiphytes and their contributionto canopy diversity. *Plant Ecology*, *153*, 51–63.

Rajbhandari, K.R. & Rai, S.K. (2017). *A Handbook of Flowering Plant of Nepal*, Vol.1. Kathmandu, Nepal: Department of Plant Resources.

Raskoti, B.B. & Ale, R. (2019). New species of orchids and notes on Orchidaceae of Nepal. *Phytotaxa*, *394*(4), 257–266.

Rabinowitz, D.(1981). Seven forms of rarity. Pp. 205–218 In H. Spunge, ed. The Biological Aspectsof Rare Plant Conservation, New York: John Wiley and Sons.

Rokaya, M.B., Raskoti, B.B., Timsina, B. & Mu¨nzbergova´, Z. (2013). An annotated checklist of the orchids of Nepal. *Nordic Journal of Botany*, *31*, 511-550.

Roskov, Y., Ower, G., Orrell, T., Nicolson, D., Bailly, N., Kirk, P.M., Bourgoin, T., DeWalt, R. E., Decock, W., Nieukerken, E. van, Zarucchi J. & Penev L., Eds. (2019). Species 2000 & ITIS Catalogue of Life, 2019 Annual Checklist. Digital resource at www.catalogueoflife.org/annual-checklist/2019. Species 2000: Naturalis, Leiden, the Netherlands. ISSN 2405-884X.

Shrestha, K.K., Bhattarai, S. & Bhandari, P. (2018). *Handbook of Flowering Plants of Nepal: Gymnosperms and Angiosperms: Cycadaceae-Betulaceae*, Vol. 1. Jodhpur, India: Scientific Publishers.

Subedi, A., Kunwar, B., Vermeulen, J.J., Choi, Y., Andel, T., Chaudhary, R.P. & Gravendeel, B. (2013). Medicinal use and trade of wild orchids in Nepal. *Journal of Ethnobiology and Ethnomedicine*, *9*(1), 64.

Trapnell, D.W. & Hamrick, J.L. (2006). Variety of phorophyte species colonized by the neotropical epiphyte, *Laeliarubescens* (Orchidaceae). *Selbyana*, *27*(1), 60-64.

Wagner, K., Mendieta-Leiva., G. & Zotz, G. (2015). Host specificity in vascular epiphytes: a review of methodology, empirical evidence and potential mechanisms. *AoBPLANTS,7,* plu092; doi:10.1093/aobpla/plu092.

Zotz, G. & Winkler, U. (2013). Aerial roots of epiphytic orchids the velamen redicum and its role in water and nutrient uptake. *Oecologia*, *171*, 733–741.

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Status of an Endemic Taxa *Rhododendron cowanianum* in Langtang National Park, Central Nepal

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Abstract

A systematic investigation of status of *Rhododendron cowanianum* Davidian from Rasuwa district, Central Nepal was carried out. Study aimed at identifying the occurrence status, mean density, and habitat characteristics of the endemic taxa from the Langtang National Park. It was found in scattered forests under open canopy from 2898 m to 3600 m asl. The mean density of *R. cowanianum* was found to be 0.73 individuals/m^2 which shows a good status of this taxa in Langtang National Park. Altogether, 113 species of vascular plants, belonging to 88 genera and 53 families, were found to be associated with *R. cowanianum*, where Ericaceae with five genera and 12 species was found to be the largest family, followed by Rosaceae, Fabaceae, Ranunculaceae and so on. *Rhododendron* with seven species was the dominant genera of vascular plants in the study area. Usually, endemic species are confined to specific areas and thus they are the first to be affected by land use and other global changes. The current human population explosion, alarming rate of deforestation, habitat fragmentation and modern-day environmental changes are posing greater threats to these endemic plant species worldwide. But, the population status, conservation status and threats to these peculiar plant species are yet underrated. Therefore, local authorities, government and non-governmental organizations should pay special attention towards the study and conservation of such endemic plant species.

Keywords: Conservation status, Taxonomy, Threats, Vascular plants

Introduction

Nepal is located in the central main Himalaya thus it is characterized by the presence of six adjoining floristic regions. Together, the altitudinal range extends from 60m to 8848 m asl which has made Nepal rich in plant biodiversity, with 5309 species under 1515 genera of 193 families (Rajbhandari et al., 2017). Some plant species are found only in Nepal and not in other parts of the world, such plants are called as endemic to Nepal. Endemic taxa refers to any species, or a taxonomic unit, whose distribution is confined within a restricted geographical area (Gaston, 1991) and this phenomenon is known as endemism. In other words, endemic plant species are the plants that exist only in one geographical region. The diversity in topography due to altitudinal variations and climatic fluctuations has greatly contributed to the occurrence of a wide variety of endemic plants in Nepal. As per the recent estimate, a total of 312 endemic flowering plant species are found in Nepal belonging to 126 genera of 46 families (Rajbhandari et al., 2016) with higher endemism around the elevation of 3800–4200 m at sea level (Tiwari et al., 2019).

Usually an area that contains endemic species is isolated in some way, so that plant species have difficulty spreading to other areas, or it has unusual environmental characteristics to which endemic species are uniquely adapted. Most of such species and their habitats in many areas are under serious threat. Consequently, many of the endemic and useful species are now threatened, endangered and facing extinction. Particularly, the endemic species in mountains are more vulnerable to rapid climate change (Suwal & Vetaas, 2017). No precise estimate can be made of the number of species being endangered and lost in major habitats, but evidences indicate that the situation is alarming. If the current trends of habitat destruction and over-exploitation practices persist, majority of the endemic species will be lost forever. Hence, there is an urgent need to conserve the valuable but fast disappearing species for the sake of establishing ecological balance. For this purpose, it is necessary to study the geographical location, population ecology, conservation status and threats of the endemic taxa.

Rhododendron cowanianum Davidian is a member of family Ericaceae, the Gurans family and is an endemic taxon reported from Nepal, named after Dr. J. M. Cowan (Milleville, 2002). It is a shrub predominantly occurring in scattered patches in *Betula- Abies* forests. This species is endemic to Western and Central Nepal, where it extends from the south of Dhaulagiri to Langtang and Gosainkunda, from 3000 to 4000 m asl (Milleville, 2002; Department of Plant Resources [DPR], 2005). This study was carried out to document the occurrence status, mean density, and habitat characteristics of *Rhododendron cowanianum* from the Langtang National Park.

Taxonomy of Rhododendron cowanianum

Upright shrub, 0.5– 3 m tall, deciduous; stem branched, branchlets scaly, sparsely bristly or, glabrous; thin, oval to elliptic leaves, 2- 6.5 × 1.2- 3 cm, apex rounded, mucronate, base obtuse or, cuneate, margins ciliate, upper surface dark green and lepidote, lower surface pale green, lepidote with broadly rimmed scales; inflorescences raceme, racemes terminal, 3-5 flowered, umbellate, pedicels, 1- 2 cm, longer than corolla, scaly; flowers pink to mauve, appearing with or before the new leaves; calyx 5-lobed, 0.4- 1 cm, reddish, rounded, ovate, oblong, margin usually ciliate; corolla campanulate, tube short, 1.3- 2 cm long, pink to mauve, with or without darker spots; stamens 10, 0.8- 1.5 cm, longer than corolla tube, anthers white, filaments reddish, densely wooly-hairy at base; ovary 3- 5 mm, 5-celled, scaly, style short, sharply bent, not scaly; fruits capsule, 0.5- 1 cm, conic or oblong, scaly (Cullen, 1980; Milleville, 2002; DPR, 2005).

Type: Nepal, Langtang lateral valley, 3650 m, 7 June 1949, Polunin 175 (Holotype: BM, Isotype: E).
Local name: Kisur.
Flowering: April to June
Distribution: Endemic to Western and Central Nepal (DPR, 2005).

***Conservation status and threat*:** This species of *Rhododendron* has been included in the list of endangered species (Milleville, 2002).

Materials and Methods

Study area

The spatial location of Nepal in the Himalaya made Central Nepal a meeting place of both western and eastern Himalayan floristic as well as a meeting place of characteristic taxa endemic to eastern and western Nepal. Thus, Central Nepal is rich in terms of biological diversity as well as endemic taxa, therefore Rasuwa district was selected for the present study. Rasuwa district lies within Central Nepal and covers an area of 1,544 km² and has a population of 43,300 as per the census of 2011 (Central Bureau of Statistics [CBS], 2012). Langtang valley of Rasuwa district which lies within the territory of Langtang National Park was selected for the present study. Langtang National Park (LNP, Nepal) provides unique habitat for a number of highly valuable medicinal and aromatic plants (MAPs) which have intense local utilization as well as high trade demand (Shrestha & Shrestha, 2012). The map of study area is given in Figure 1.

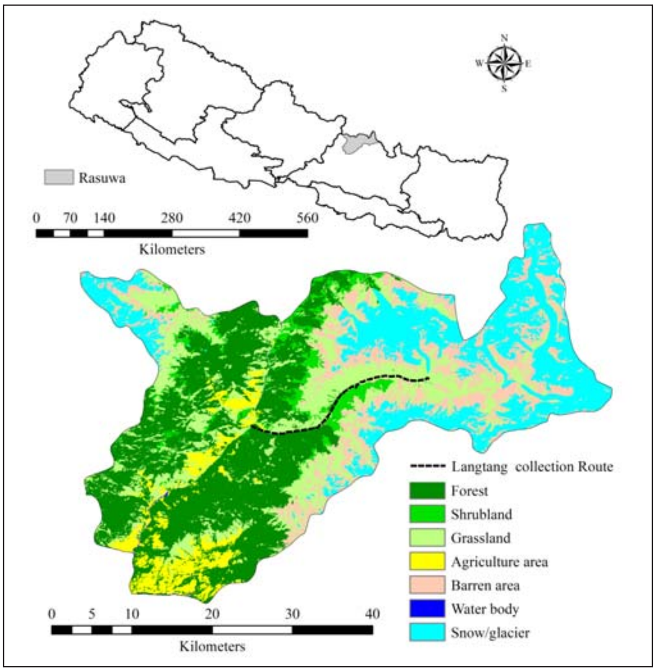

Figure 1: Map of study area showing Rasuwa district with different land use types and the collection route in Langtang valley.

Methods

The field visit was carried out during June 2019 in Rasuwa district, Central Nepal. During this visit, 5m × 5m quadrats were laid down at every 200 m elevation interval from 2900 m to 3500 m in three different transects to access the population status of *Rhododendron cowanianum*. The plots were replicated twice such that three plots each with an area of 25 m^2 was studied in each transect. Hence a total of nine such plots each covering areas of 25 m^2 were studied during this period. The associated species recorded in the study plots as well as along the walking trail (flowering or fruiting) were collected and herbarium specimens are deposited at National Herbarium and Plant Laboratories (KATH). However, the vascular plant species which were not on the state of flowering or, fruiting were simply noted to determine the species diversity of vascular plants in the study area. The identification of vascular plant species was carried out based on standard literatures (Grierson & Long 1983-2000; Polunin & Stainton, 1984; Stainton, 1988; Press et al., 2000; Ohba et al., 2008; Fraser-Jenkins et al., 2015). Nomenclature follows Catalogue of life (http://www.catalogueoflife.org) and the Plants of World Online (http://www.plantsoftheworldonline.org).

The data gathered from the field were tabulated in MS Excel and the necessary calculations were made. The frequency and density of *R. cowanianum* were calculated by using the following formulas.

$$\text{Frequency} = \frac{\text{No. of plots with species}}{\text{Total No. of plots taken}} \times 100\%$$

$$\text{Density (individual/m}^2\text{)} = \frac{\text{Total No. of individual of the species}}{\text{Total No. of plots} \times \text{Size of quadrats}}$$

The pattern of distribution of the species along the elevation was analyzed by linear regression. The strength of which is given by regression equation and R^2 value. A positive regression equation indicates positive relationship between the parameters. The R^2 value lies between 0 to 1 where,a value close to 1 indicates strong relationship between the parameters.

Results and Discussion

R. cowanianum was found in scattered forests under open canopy from 2898 m to 3600 m asl. It resembles with another *Rhododendron* species; *R. lepidotum* but can be distinguished by its oval to elliptic leaves with ciliated margins; pink to mauve flowers borne in clusters of three to five flowers; anthers with filaments bearing feathers at base only; carpel with characteristic style which is sharply bent (Figure 2).

A total of 49 individuals of *R. cowanianum* were recorded in the study plots. The mean density of *R. cowanianum* was 0.73 individuals/m^2 and the frequency of the study species was found to be 100%. The total number of individuals of *R. cowanianum* increased with increasing elevation (Figure 3). The positive regression equation and R^2 value of 0.75 shows a strong relationship between the number of individuals and elevation. Similarly, the p-value of 0.0025 indicates that the number of individuals significantly increases with increase in elevation.

A total of 113 species of vascular plants, belonging to 88 genera and 53 families, were recorded during the present study. Ericaceae with 5 genera and 12 species was found to be the largest family, followed by Rosaceae (7 genera, 9 species), Fabaceae (6 genera, 8 species), Ranunculaceae (3 genera, 5 species) and so on (Figure 4). A total of 87 species of dicotyledonous plants, 17 monocotyledonous plants, 5 gymnosperms and 4 pteridophytes were recorded, where Ericaceae (5 genera and 12 species) was the largest family of dicots and Asparagaceae and Zingiberaceae (3 genera and 4 species each) were the largest family of monocots. Similarly, Pinaceae (3 genera and 4 species) was the largest family of gymnosperms.

Rhododendron with 7 species was the dominant genera of vascular plants, which were followed by *Clematis* (4 species), *Primula*, *Astragalus* (3 species each), *Salix*, *Gaultheria*, *Polygonatum*, *Daphne*, etc. (2 species each) and so on. The list of associated vascular plant species recorded during the study is presented in the Tables.

Figure 2: *Rhododendron cowanianum* Davidian. **a- b.** Plant in its habitat (**a.** Flowering twig, **b.** Vegetative branch**)**, **c.** Lepidote leaves with brown scales and hairy margin, **d.** inflorescence bearing multiple flowers in a cluster, **e.** Stamens feathery at the base only, **f.** Carpel with a bent style.

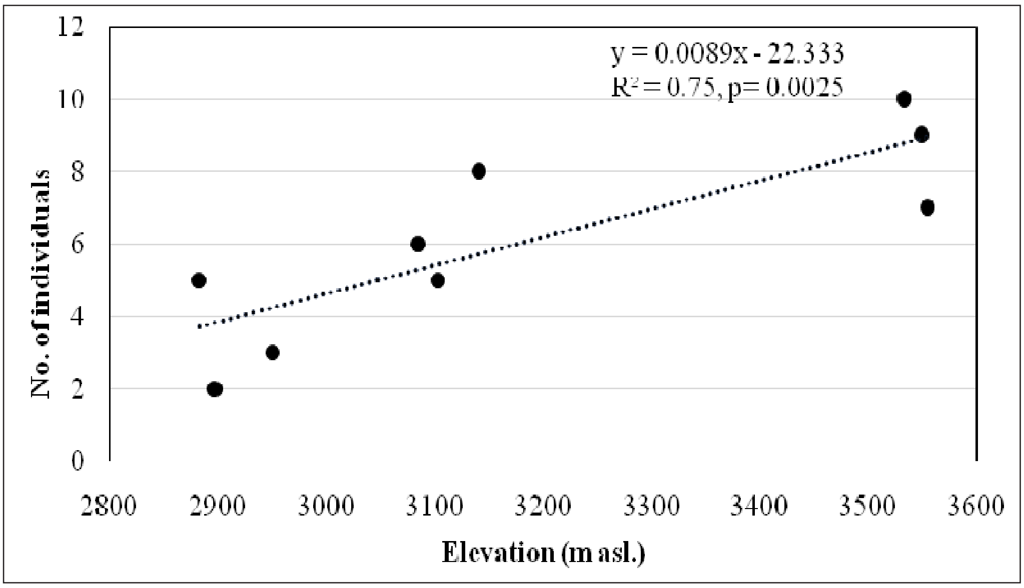

Figure 3: Variation in number of individuals with elevation

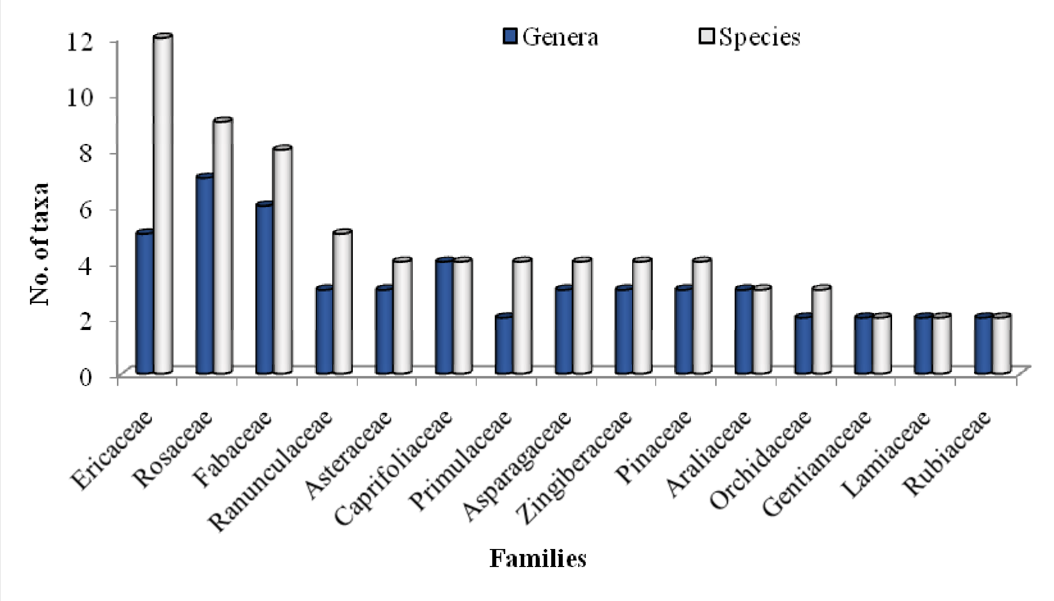

Figure 4: Distribution of families, genera and species of vascular plants

Conclusion

The study came up with the basic idea about the occurrence status of an endemic plant species *Rhododendron cowanianum* from Langtang National Park in Rasuwa district. It was found in scattered forests under open canopy from 2898 m to 3600 m asl. With the mean density of 0.73 individuals/m^2 which shows a good status of this taxa in Langtang National Park. Apart from *R. cowanianum*, another taxa *Sorbus sharmae* M. F. Watson, V. Manandhar and Rushforth which is endemic to Eastern and Central Nepal was also recorded during this study. Since it is a tall tree and a single individual of this plant was recorded, the population status of this plant could not be studied in detail. Usually, endemic species are confined to specific areas and thus they are the first to be affected by land use and other global changes. The current human population explosion, alarming rate of deforestation, habitat fragmentation and modern-day environmental changes are posing greater threats to these endemic plant species worldwide. But, the population status, conservation status and threats to these peculiar plant species are yet underrated. Therefore, local authorities, government and non-governmental organizations should pay special attention towards the study and conservation of such endemic plant species.

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References

Central Bureau of statistics. (2012). *National Population and Housing Census 2011*. National report. Kathmandu, Nepal: Author.

Cullen, J. (1980). A Revision of *Rhododendron* 1: Subgenus Rhododendron Sections Rhododendron and Pogonanthum. *Notes from the Royal Botanic Garden Edinburgh, 39*(1).

Department of Plant Resources. (2005) *Rhododendrons of Nepal (Fascicle of Flora of Nepal. Vol 5 No.6 partly)*, Kathmandu, Nepal: Author.

Fraser-Jenkins, C.R., Kandel, D.R., & Pariyar, S. (2015). *Ferns and Fern-allies of Nepal* – 1, (pp. 508). Kathmandu, Nepal: Department of Plant Resources.

Gaston, K.J. (1991). How large is a species' geographical range. *Oikos*, *61*, 329–335.

Grierson, A.J.C., & Long, D.G. (1983-2000). *Flora of Bhutan. Vol. 1, Part 1-3; Vol. 2, Part 1-3*. Edinburgh: Royal Botanic Garden & Bhutan: Royal Government of Bhutan.

Milleville, R. (2002). *The Rhododendrons of Nepal*. Lalitpur, Nepal: Himal Books.

Ohba, H., Iokawa, Y. & Sharma, L.R. (2008). *Flora of Mustang, Nepal*. Tokyo: Kodansha Scientific Ltd.

Polunin, O. & Stainton, A. (1984). *Flowers of the Himalaya*. New Delhi, India: Oxford University Press.

Press, J.R., Shrestha, K.K. & Sutton, D.A. (2000). *Annotated Checklist of the Flowering Plants of Nepal*. London: The Natural History Museum.

Rajbhandari K.R., Rai S.K., & Bhatt G.D. (2016). Endemic Flowering Plants of Nepal: An update. *Bulletin of Department of Plant Resources*, *38*,106-144.

Rajbhandari K.R., Rai S.K., Bhatt G.D., & Chhetri, R. (2017). *Flowering Plants of Nepal: An introduction*. Kathmandu, Nepal: Department of Plant Resources.

Shrestha, N. & Shrestha, K. K. (2012). Vulnerability assessment of high-valued medicinal plants in Langtang National Park, Central Nepal. *Biodiversity*, *13*(1), 24-36.

Stainton, A. (1988). *Flowers of the Himalaya: A supplement*. New Delhi, India: Oxford University Press.

Suwal, M.K. & Vetaas, O.R. (2017). Climatic variables determining Rhododendron sister taxa distributions and distributional overlaps in the Himalayas. *Frontiers of Biogeography*, *9*(3), e34911. doi: 10.21425/F59334911.

Tiwari, A., Uprety, Y. & Rana, S. (2019). Plant Endemism in the Nepal Himalayas and Phytogeographical Implications. *Plant Diversity*, *41*(3), 174-182.

http://www.catalogueoflife.org (retrived on various dates)

http://www.plantsoftheworldonline.org (retrived on various dates)

Table 1: List of Angiosperms (Dicots)

S.N.	Scientific Names	Family	Latitude	Longitude	Altitude (m asl)	Collection No.
1.	*Acer sterculiaceum* Wall.	Aceraceae	28°9.5278' N	85°25.2601' E	2743.1	R0007
2.	*Achyranthes aspera* L.	Amaranthaceae	-	-	-	-
3.	*Ainsliaea latifolia* (D. Don) Sch. Bip.	Asteraceae	28°9.5278' N	85°25.2601' E	2786.7	R0010
4.	*Alnus nepalensis* D.Don	Betulaceae	-	-	-	-
5.	*Anaphalis margaritacea* (L.) Benth. and Hook.f	Asteraceae	-	-	-	-
6.	*Anaphalis triplinervis* Sims. ex C.B. Clarke.	Asteraceae	-	-	-	-
7.	*Androsace sarmentosa* Wall.	Primulaceae	28°11.7479' N	85°27.1352' E	2979	R0016
8.	*Astragalus rhizanthus* Benth.	Fabaceae	28°12.6362' N	85°33.1590' E	3713.6	R0052
9.	*Astragalus* sp.	Fabaceae	28°12.9550' N	85°30.9268' E	3482.7	R0035
10.	*Astragalus* sp.	Fabaceae	28°12.6748' N	85°32.0843' E	3599	R0046
11.	*Aralia leschenaultii* (DC.) J. Wen.	Araliaceae	28°11.5285' N	85°26.8841' E	2800	R0011
12.	*Begonia picta* Sm.	Begoniaceae	-	-	-	-
13.	*Berberis* sp.	Berberidaceae	28°12.6645' N	85°32.2054' E	3616	R0047
14.	*Bergenia ciliata* (Haw.) Sternb.	Saxifragaceae	-	-	-	-
15.	*Bupleurum longicaule* Wall. ex DC.	Apiaceae	-	-	-	-
16.	*Campanula pallida* Wall.	Campanulaceae	-	-	-	-
17.	*Caragana sukiensis* C. K. Schneid	Fabaceae	28°12.2776' N	85°27.9631' E	3097.2	R0029
18.	*Cassiope fastigiata* (Wall.) D. Don	Ericaceae	28°12.6748' N	85°32.0843' E	3599	R0045
19.	*Ceropegia pubescens* Wall.	Apocynaceae	28°9.5278' N	85°25.2601' E	2654	R0005
20.	*Clematis barbellata* Edgew.	Ranunculaceae	28°12.7171' N	85°32.0594' E	3596.1	R0041
21.	*Clematis buchananiana* DC.	Ranunculaceae	-	-	-	-
22.	*Clematis connata* DC.	Ranunculaceae	-	-	-	-
23.	*Clematis montana* Buch.-Ham. ex DC.	Ranunculaceae	28°12.9091' N	85°30.9238' E	3489.7	R0037
24.	*Clinopodium umbrosum* (M. Bieb.) K. Koch	Lamiaceae	-	-	-	-
25.	*Cotoneaster microphyllus* Wall. ex Lindl.	Rosaceae	28°12.4531' N	85°28.4808' E	3170.1	R0024
26.	*Crotalaria* sp.	Fabaceae	28°11.8098' N	85°27.1951' E	2995.7	R0022
27.	*Cynoglossum zeylanicum* (Sw. ex Lehm.) Thunb. ex Brand	Boraginaceae	-	-	-	-
28.	*Daphne bholua* Buch.-Ham. ex D. Don.	Thymelaeaceae	-	-	-	-
29.	*Daphne papyracea* Wall. ex G. Don.	Thymelaeaceae	-	-	-	-
30.	*Dipsacus inermis* Wall.	Caprifoliaceae	-	-	-	-
31.	*Epilobium wallichianum* Hausskn.	Onagraceae	-	-	-	-
32.	*Euphorbia stracheyi* Boiss.	Euphorbiaceae	28°11.5022' N	85°34.8554' E	4081	R0063
33.	*Gaultheria fragrantissima* Wall.	Ericaceae	-	-	-	-
34.	*Gaultheria trichophylla* Royle.	Ericaceae	28°12.7204' N	85°33.8289' E	3836.9	R0056

S.N.	Scientific Names	Family	Latitude	Longitude	Altitude (m asl)	Collection No.
35.	*Gentiana capitata* Buch.-Ham. ex D.Don	Gentianaceae	-	-	-	-
36.	*Geranium nepalense* Sweet	Geraniaceae	-	-	-	-
37.	*Hedera nepalensis* K.Koch	Araliaceae	-	-	-	-
38.	*Hemiphragma heterophyllum* Wall.	Plantaginaceae	-	-	-	-
39.	*Hypericum elodeoides* Choisy.	Hypericaceae	-	-	-	-
40.	*Hypericum uralum* Buch.-Ham. ex D. Don	Hypericaceae	-	-	-	-
41.	*Impatiens racemosa* DC.	Balsaminaceae	-	-	-	-
42.	*Lonicera* sp.	Caprifoliaceae	28°12.9310' N	85°31.2119' E	3491.8	R0038
43.	*Papaver paniculatum* D.Don	Papaveraceae	28°12.8526' N	85°29.7165' E	3390.5	R0034
44.	*Morina polyphylla* Wall. ex DC.	Caprifoliaceae	28°12.7171' N	85°32.0594' E	3596.1	R0044
45.	*Myrica esculenta* Buch.-Ham. ex D.Don	Myricaceae	-	-	-	-
46.	*Myricaria rosea* W. W. Sm.	Tamaricaceae	28°12.6362' N	85°33.1590' E	3713.4	R0053
47.	*Neohymenopogon parasiticus* (Wall.) Bennet	Rubiaceae	-	-	-	-
48.	*Panax pseudoginseng* Wall.	Araliaceae	28°11.6913' N	85°27.0389' E	2883.5	R0012
49.	*Parochetus communis* Buch.-Ham. ex D.Don	Fabaceae	28°9.5278' N	85°25.2601' E	2760.3	R0008
50.	*Pieris formosa* (Wall.) D. Don	Ericaceae	28°9.5278' N	85°25.2601' E	2738.5	-
51.	*Piptanthus nepalensis* (Hook.) Sweet	Fabaceae	-	-	-	-
52.	*Dasiphora fruticosa* (L.) Rydb.	Rosaceae	28°13.0087' N	85°35.9546' E	4787.7	R0070
53.	*Potentilla* sp.	Rosaceae	28°12.7171' N	85°32.0594' E	3596.1	R0042
54.	*Primula atrodentata* W. W. Sm.	Primulaceae	28°12.9460' N	85°36.0433' E	4864.7	R0071
55.	*Primula minutissima* Jacquem. ex Duby.	Primulaceae	28°12.7948' N	85°36.0271' E	4984.2	R0065
56.	*Primula sikkimensis* Hook.	Primulaceae	28°12.5774' N	85°32.7900' E	3646.5	R0049
57.	*Prunella vulgaris* L.	Lamiaceae	-	-	-	-
58.	*Prunus napaulensis* (Ser.) Steud.	Rosaceae	-	-	-	-
59.	*Quercus semecarpifolia* Sm.	Fagaceae	-	-	-	-
60.	*Ranunculus diffusus* DC.	Ranunculaceae	-	-	-	-
61.	*Rhodiola bupleuroides* (Wall. ex Hook.f. and Thomson) S.H.Fu	Crassulaceae	28°12.6647' N	85°32.2057' E	3624.9	R0048
62.	*Rhododendron anthopogon* D. Don	Ericaceae	-	-	-	-
63.	*Rhododendron arboreum* Sm.	Ericaceae	-	-	-	-
64.	*Rhododendron campanulatum* D. Don	Ericaceae	28°12.6362' N	85°33.1590' E	3713.4	R0058
65.	*Rhododendron cowanianum* Davidian	Ericaceae	28°11.7598' N	85°27.1387' E	2978.3	R0018
66.	*Rhododendron lepidotum* Wall. ex G.Don	Ericaceae	28°11.7479' N	85°27.1352' E	2979	R0014
67.	*Rhododendron nivale* Hook.f	Ericaceae	28°12.9188' N	85°36.0469' E	4888.5	R0068
68.	*Rhododendron setosum* D. Don	Ericaceae	28°12.7204' N	85°33.8289' E	3836.9	R0055
69.	*Ribes griffithii* Hook.f and Thomson	Grossulariaceae	28°11.5285' N	85°26.8841'	2800.	R0009

S.N.	Scientific Names	Family	Latitude	Longitude	Altitude (m asl)	Collection No.
				E	4	
70.	*Rosa macrophylla* Lindl.	Rosaceae	28˚11.8098' N	85˚27.1951' E	2995.7	R0021
71.	*Rosa sericea* Wall. ex Lindl.	Rosaceae	28˚12.7171' N	85˚32.0594' E	3596.1	?
72.	*Rubia manjith* Roxb.	Rubiaceae	-	-	-	-
73.	*Rubus ellipticus* Sm.	Rosaceae	-	-	-	-
74.	*Salix daltoniana* Andersson.	Salicaceae	28˚12.2769' N	85˚27.9652' E	3071.6	R0025
75.	*Salix lindleyana* Wall. ex Andersson	Salicaceae	28˚12.5069' N	85˚34.7966' E	4045.4	R0066
76.	*Schisandra grandiflora* (Wall.) Hook.f & Thomson	Schisandraceae	28˚9.5278' N	85˚25.2601' E	2738.5	R0006
77.	*Sibbaldia cuneata* Hornem. ex Kuntze	Rosaceae	28˚12.8569' N	85˚29.7073' E	3358.5	R0033
78.	*Sorbus sharmae* M. F. Watson, V. Manandhar & Rushforth.	Rosaceae	28˚12.3977' N	85˚28.4052' E	3102	R0031
79.	*Stauntonia latifolia* (Wall.) R.Br. ex Wall.	Lardizabalaceae	-	-	-	-
80.	*Swertia chirayita* (Roxb.) H.Karst.	Gentianaceae	-	-	-	-
81.	*Taraxaccum* sp.	Asteraceae	28˚11.8301' N	85˚27.2072' E	3001.7	R0023
82.	*Tetrastigma serrulatum* (Roxb.) Planch	Vitaceae	-	-	-	-
83.	*Tetrastigma* sp.	Vitaceae	28˚9.2622' N	85˚22.4208' E	1929.3	R0003
84.	*Thalictrum foliolosum* DC.	Ranunculaceae	-	-	-	-
85.	*Thermopsis barbata* Benth.	Fabaceae	28˚12.2769' N	85˚27.9652' E	3071.6	R0027
86.	*Vaccinium nummularia* Hook.f. & Thomson ex C.B. Clarke	Ericaceae	-	-	-	-
87.	*Viburnum cotinifolium* D. Don	Caprifoliaceae	28˚12.2769' N	85˚27.9652' E	3071.6	R0028

Table 2: List of Angiosperms (Monocots)

S. N.	Scientific Names	Family	Latitude	Longitude	Altitude (m asl)	Collection No.
1.	*Asparagus racemosus* Willd.	Asparagaceae	-	-	-	-
2.	*Arisaema propinquum* Schott	Araceae	-	-	-	-
3.	*Bulbophyllum* sp.	Orchidaceae	-	-	-	-
4.	*Cautleya spicata* (Sm.) Baker	Zingiberaceae	-	-	-	-
5.	*Disporum cantoniense* (Lour.) Merr.	Colchicaceae	28˚11.8098' N	85˚27.1951' E	2995.7	R0020
6.	*Fritillaria cirrhosa* D. Don	Liliaceae	28˚12.6352' N	85˚33.1590' E	3714.5	R0051
7.	*Globba clarkei* Baker.	Zingiberaceae	-	-	-	-
8.	*Iris sp.*	Iridaceae	28˚12.2776' N	85˚27.9631' E	3097.2	R0030
9.	*Juncus thomsonii* Buchenau.	Juncaceae	28˚12.6362' N	85˚33.1590' E	3713.4	R0054
10.	*Calanthe* sp.	Orchidaceae	28˚9.2022' N	85˚22.9622' E	1831.1	R0001
11.	*Pleione hookeriana* (Lindl.) Rollisson	Orchidaceae	28˚9.5278' N	85˚25.2601' E	2739	R0004
12.	*Polygonatum hookeri* Baker.	Asparagaceae	28˚12.7406' N	85˚31.9564' E	3601.4	R0040
13.	*Polygonatum verticillatum* (L.) All.	Asparagaceae	28˚12.9091' N	85˚30.9238' E	3489.7	R0036
14.	*Roscoea alpina* Royle.	Zingiberaceae	28˚11.7479' N	85˚27.1352' E	2979	R0015

S. N.	Scientific Names	Family	Latitude	Longitude	Altitude (m asl)	Collection No.
15.	*Roscoea purpurea* Sm.	Zingiberaceae	-	-	-	-
16.	*Smilax elegans* Wall. ex Kunth.	Smilaceae	-	-	-	-
17.	*Theropogon pallidus* (Wall. ex Kunth) Maxim.	Asparagaceae	-	-	-	-

Table 3: List of Gymnosperms

S.N.	Scientific Names	Family	Latitude	Longitude	Altitude (m asl)	Collection No.
1.	*Abies spectabilis* (D. Don) Mirb.	Pinaceae	28°12.7171' N	85°32.0594' E	3596.1	R0043
2.	*Ephedra gerardiana* Wall. ex Klotzsch & Garcke	Ephedraceae	28°12.7406' N	85°31.9564' E	3601.4	R0039
3.	*Larix griffithii* Hook.f.	Pinaceae	28°12.4531' N	85°28.4808' E	3170.1	R0032
4.	*Pinus roxburghii* Sarg.	Pinaceae	-	-	-	-
5.	*Pinus wallichiana* A. B. Jacks	Pinaceae	-	-	-	-

Table 4: List of Pteridophytes

S. N.	Scientific Names	Family	Latitude	Longitude	Altitude (m asl)	Collection No.
1.	*Japanobotrychum lanuginosum* (Wall. ex Hook. and Grev.) M. Nishida ex Tagawa	Ophioglossaceae	-	-	-	-
2.	*Onychium cryptogrammoides* Christ.	Pteridaceae	-	-	-	-
3.	*Equisetum* sp.	Equisetaceae	-	-	-	-
4.	*Lepisorus* sp.	Polypodiaceae	-	-	-	-

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Altitudinal Pattern of Pteridophyte in Arghakhanchi district, West Nepal

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Abstract

Pteridophyte species richness pattern was carried out in Arghakhanchi district, West Nepal during October-November, 2018 and 2019. Main objectives of this study were to document the floristic composition of pteridophyte and to access the species richness pattern along altitude, temperature, rainfall and area per 100 m contour elevation. The pteridophyte species were recorded and collected after utilizing systematic random sampling strategies in each forest types into 10 x 10 m^2 area each 100 m altitudinal band from the bottom to the top of the mountain (240-2300 m asl). Herbarium of each collected pteridophyte sample present inside plots and along the track was identified with the help of the published literature and deposited in the TUTH. The pteridophyte species richness was plotted against altitude, interpolated temperature, rainfall and area per 100 m contour elevation through application of the Generalized Linear Model (GLM). The species environment relation was observed by application of CCA. A total of 75 pteridophyte species belonging to 18 families and 36 genera were reported among which Pteridaceae with 5 genera and 21 species was the richest family and *Thelypteris* with 9 species was the richest genera. Among those species 39 species were terrestrial followed by 12 species of lithophytes and 11 epiphytes. The pteridophyte richness showed statistically significant unimodal relationship with altitude and temperature (R^2 = 0.95 and $p < 0.001$).Most of the fern species were present at moist places of *Schima-Castonopsis-Quercus* forest above than 1500 m.

Keywords: Contour, Elevation, Habitat, Pteridaceae, Species richness,Unimodal

Introduction

Pteridophyte is a beautiful gift of nature which provides magnificent beauties without flowers. They form an attractive component of the vegetation of hills and forests (Gurung, 1991). They live in habitats from the tropics to polar latitudes unlike seeded plants; they reproduce by minute spores (Moran, 2004). The popular Nepalese local slogan "Unyu phulnu ra dhunga rasaunu ekai ho" says that ferns have neither seeds nor flowers but can reproduce offspring. They complete their life-cycle in two generations: sporophyte and gametophyte. Pteridophyte is a unique group of plant with two phases independent life cycle: gametophyte and sporophyte.

Pteridophytes are the earliest vascular plants originated during the Silurian period (400 million years ago) of Paleozoic Era and formed the dominant during Devonian to Permian period (Khare, 1996). They are the most diverse group and the oldest lineage of vascular plant and the second most species after angiosperms with an estimate of 12,000 species in the world (Vidyashree, 2018). Most of them are abundantly found in humid and shady forests area. Nowadays, pteridophyte found grows in different ecological habits thus classified as epiphyte, lithophytes, terrestrial, tree ferns, climbers and hydrophytes (Gurung 1984; Gurung, 1991; Moran, 2004), but some species occur in more than one habitat. Most of them are annual and some are perennial, but some are climbers, creepers and small tree (*Cyathea spinulosa*). They rarely form pure vegetation but form understory inside the forest. Pteridophyte foliage is highly considered as economically valuable in decoration, food, medicine, biofertilizer, ornamental and reclamation of contaminated soil (Vidyashree, 2018). Being very sensitive to direct sunlight, epiphytic ferns could be used as indicators of forest disturbances (Edward et al., 2003) andhave wide range of habitat preferences (Nagalingum & Cantrill, 2015).

Nepal has a rich and diversified bioresources due to varied topographical and climatic regions. The studies on species richness patterns of Medicinal plants (Acharya et al., 2010), orchids (Acharya et al., 2011), higher plants (Bhattarai & Vetaas, 2003), ferns (Bhattarai et al., 2004), bryophytes (Grau et al., 2007) in Nepal's mountains show unimodal pattern with variation in peak. Many studies in ferns (Bhattarai & Vetaas, 2003; Watkie et al., 2006; Nagalingum & Cantrill , 2015; Jeyalatchayan et al., 2020) showed that unimodal is common pattern of fern species against altitude. In many cases, area accounts the large percentage of variation than altitude (Xiang et al., 2017). The environmental gradient drives pattern of fern species richness on the spatial scale (Watkin et al., 2006). Climate is one of the strongest predictors for diversity of species and their richness, especially for epiphytic species (Zhang et al., 2015).

The Department of Plant Resources (DPR), Nepal (2002) reported 534 species of pteridophyte species in Nepal. Fraser-Jenkins et al. (2015) reported 580 species of pteridophyte in Nepal. These reports only generalized the number of pteridophyte species present in the whole country after enumeration. No direct ecological study has been taken yet so far focusing on the definite landscape. Arghakhanchi, one of the middle mountains in Nepal, lies in the borderline between tropical and subtropical ecological zones as well as beginning of west Nepal has been considered as important place to this study. The main objectives of this paper were documentation of pteridophyte species present in Arghakhanchi district and find out the pteridophyte species richness pattern along altitude temperature, rainfall and area per 100 m contour elevation.

Materials and Methods

Physiography and vegetation of the study area

Arghakhanchi, one of beautiful hilly districts, is located in providence no. 5, West Nepal. Arghakhanchi occupies an area of 1193 km^2 and extended between 27º45' to 28º06' N and 80º45' to 83º23' E (Figure 1). The population of this district was 197,632 (Central Bureau of Statistics [CBS], 2012). Other neighboring districts are Palpa in the east, Gulmi in the north, Kapilbastu and Rupandehi in the south and Pyuthan and Dang are in the west. About 68% land of this whole district lies within the Mahabharata range and remaining are in the Siwalik Hills. The elevation of this district ranges from 240 to 2515 masl (Alternative energy promotion center, [AEPC], 2016).

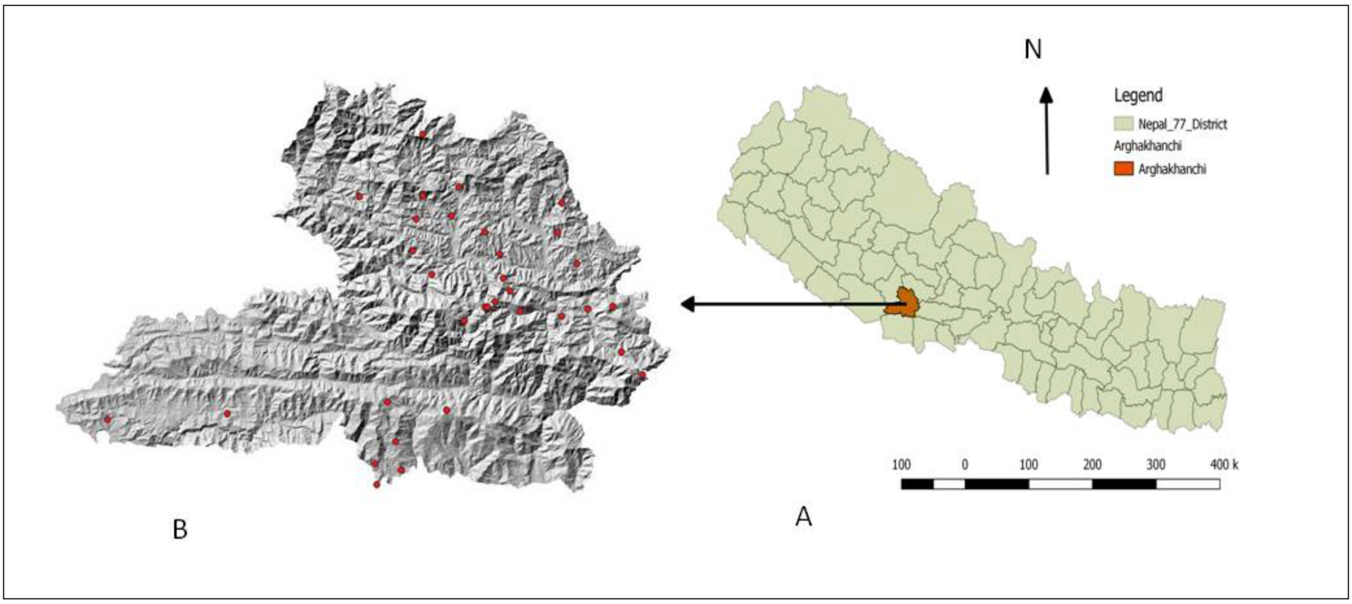

Figure 1: A. Arghakhanchi district in Nepal, B. Botanically explored sites (Drawn by using QGIS)

Physiographically, this district has four zones: lower tropical (less than 300 m asl) which covers 0.2%, upper tropical (300-1000 m asl) coves 51%, subtropical (1000-2000 m asl) which covers 49% and temperate zone (2000-3000 m asl) of 0.2% (CBS, 2012).The maximum and minimum average mean temperatures ranged between 25-27°C and 5-7°C respectively between May-July and January (Department of Hydrology and Meteorology [DHM, 2017). The average annual rainfall of this district remains around 1750 mm (CBS, 2012). Arghakhanchi district lies in the moderate temperate climatic zone of the country.

In Arghakhanchi, about 40 % of the total land is covered by the forest. Lowland (below 500 m asl) towards south consists of lower tropical *Shorea robusta* forest. Churia Sal forest lies between 500-1000 m asl and Inner hill Sal forest (up to 900 m). Northern slope of this district lies in the Mahabharat range (1000-1500 m) with the broad leaved forest intermixed with *Pinus* species. Between 1500-2000 masl, *Rhododendron arboreum, Schima wallichii, Pinus roxburghii, Myrica esculenta,* containing forest are present. The uppermost region (above 2000 m) in this district bear *Rhododendron-Quercus-Maesa* mixed forest with bushes (Panthi & Chaudhary, 2002).

Sampling design and primary data collection

The Arghakhanchi district is mainly occupied by two landscapes(Narapani-Masina landscape, 200-2200 m and Malarani-Gokhunga landscape, 700-2300 m) which are extended from East to West. The overall altitude range of this district was divided equally by 100 elevation band. The field was visited and sampled twice in the post-monsoon (October and November) of 2018 and 2019. 3-4 Sampling plot of 10 x 10 m^2 was placed at each altitudinal band of both aspects (north and south) focusing at the forest type. The distance between two sample plots varied from 100 to 150 m distance.

All pteridophyte species enrooted inside each plot was recorded and one sample of each species was collected. Photograph of each pteridophyte sample was taken. The coordinate of each plot location was also measured through GPS (*eTrex*). The pteridophyte species present outside the plots and along the track were also collected and their habitats and coordinate were also noted.

Herbarium of each properly dried and specimens were prepared in the laboratory. GPS data and other micro-ecological characters were also recorded to each herbarium specimen. All herbarium specimens were identified with the help of relevant taxonomic literature such as Gurung, 1984 & 1991; Fraser-Jenkins et al., 2015; Rajbhandary, 2016; Fraser-Jenkins & Kandel, 2019. Some species were also identified with the help of consulting experts and comparing with specimens deposited at National Herbarium and Plant Laboratories (KATH) and Tribhuvan University Central Herbarium (TUCH). All these identified herbarium were submitted in the TUCH. After identification, the altitudinal range of each species was determined on the basis of their maximum and minimum altitude.

Altitudinal area and climatic variables calculation

Species numbers tend to increase as the area increase (Rahbek, 1997), but area per 100 contour elevation does not contain equal areas due to complex pyramidal and topography of Himalayas along the elevational gradient. So, the area occupied per 100 m contour elevation of Arghakhanchi district was calculated by using Digital Elevational Model (DEM) in QGIS. Similarly, physiographic and climatic information obtained from the Department of Hydrology and Meteorology and their periodical publications such as Ministry of Agriculture and Co-operatives [MoAC], (2011),DHM (2017).The climatic variables used in this study are mean annual average temperature (AMT) and mean annual Total rainfall (MAR). The climatic records 20 years (1994-2013) of 11 stations of Arghakhanchi and surrounding districts were collected. The temperature was interpolated in 100 m counter elevation by linear regression ($r^2 = 0.99$; $p \leq 0.001$) at lapse rate of 0.5°C/100 m for mean annual temperature (AMT). Rainfall is not a simple linear function of elevation, and therefore a cubic smooth spline in Generalized Additive Model (Hastie & Tibshirami, 1990) was used with 4 degrees of freedom to estimate total annual rainfall in each 100 m counter elevation.

Statistical analysis

The patterns related to pteridophyte species as response and their altitudes, area per 100 m contour elevation, temperature and rainfall as predictor variables were analyzed after underlying the principal of Generalized Linear Model (GLM) in R 3.6.2 (R Core Team, 2019). The quasi-poisson family of error distribution was applied to remove over dispersion as Baniya et al. (2010). The assumption of normal distribution of error was conformed after Q-Q diagnostic plots plotted against residuals. The change in deviance follows the F-distribution. Canonical Correspondence Analysis (CCA) used to analyze species environmental composition (Kent & Carmel, 2011).

Results and Discussion

Pteridophyte diversity in Arghakhanchi district

A total of 75 species of pteridophytes belonging to 18 families and 36 genera were found in Arghakhanchi district. The largest families were: Pteridaceae (6 genera with 22 species), Polypodiaceae (8 genera with 11 species) (Figure 2).

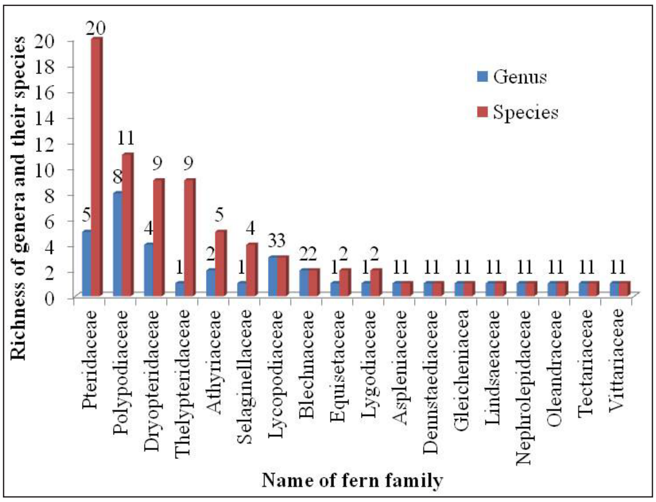

Figure 2: Richness of family wise genera and species of fern

There were 8 monogeneric (having single genus and single species) families: Aspleniaceae (*Asplenium ensiforme,)* Dennstaediaceae (*Pteridium revolutum*), Gleicheniaceae (*Dicranopteris lanigera)*, Lindsaeaceae (*Odontosoria chinensis*), Nephrolepidaceae (*Nephrolepis cordifolia*), Oleandraceae (*Oleandra wallichii*), Tectariaceae (*Tectaria gemmifera*), Vittariaceae (*Vittaria linearifolia* Ching) (Table 1). The largest species bearing genera were: *Thelypteris* (9 species), *Aleuritopteris* (6 species) and *Pteris* (5 species) (Table 1).

Pteridophyte distribution according to habitats

Pteridophyte of Arghakhanchi was found distributed into three broad habitats: terrestrial, epiphytes and lithophytes. This study found 39 terrestrial species followed 12 species of lithophytes and 11 species of epiphytes (Figure 3). The epiphytic species *Asplenium ensiforme*, *Lepisorus loriformis*, *Pyrrosia flocculosa*, *Pyrrosia porosa* were found above than 1500 m asl. The species of *Adiantum*, *Aleuritopteris Pteris, Thelypteris* were distributed at all types of forest from low to high altitude (Table 1). 47 species (63%) were present in moist places of *Schima-Castonopsis-Quercus* forests.

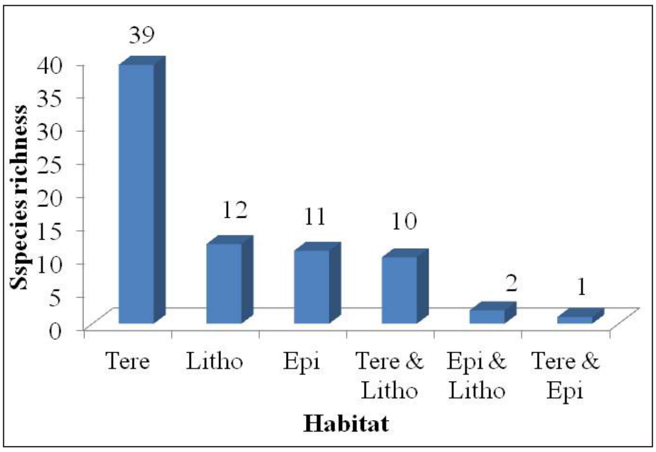

Figure 3: Pteridophyte species richness on the basis of habitat (Tere-Terestrial, Litho-Lithophyte & Epi-Epiphytes)

Species richness pattern and species environment relation

The pteridophyte species richness showed statistically highly significant unimodal richness pattern against both altitude and area per 100 m contour elevation in Arghakhanchi district (Figure 4 A and B). The pteridophyte species richness was found maximum spp. 66 at 1300 m asl and then declining (R^2 = *0.95 & p < 0.001*, Figure 4A). Similarly, species richness was found statistically unimodal pattern with area per 100 m asl contour elevation with the peak at 800 m (maximum species = 46&R^2 =*0.28*; Figure 3B). The species richness also showed the unimodal structure against temperature (*R = 0.95, p< 0.001)* and rainfall (R^2 = *0.42; p < 0.001)*.

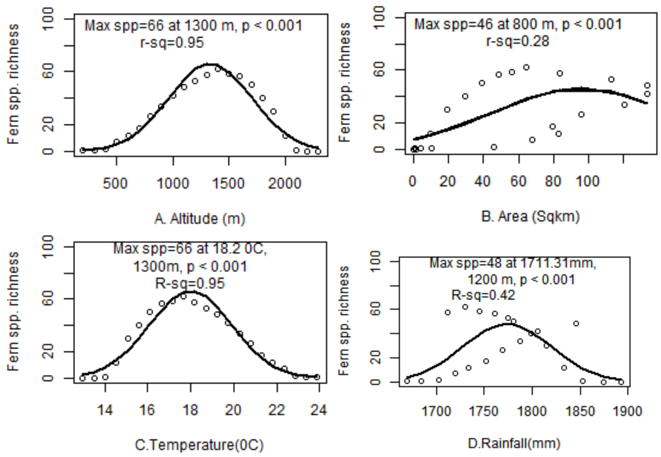

Figure 4: Scatter plots of the relationships between Pteridophyte species richness and (A) altitude & (B) area per 100 m contour elevation and (C) temperature & (D) rainfall which shows the second order polynomial fitting curves generated using *GLM* model.

The temperature and area showed the negative relation with altitude (Figure 4), but rainfall did not show any particular relation. The *Dicranopteris lanigera*, *Thelypteris glanduligera*, *Athyrium pectinatum*, *Polystichum discretum*, *Equisetum ramosissimum*, *Selaginella subdiaphana*, *Adiantum philippense*, *Lygodium japonicum* etc. were present dominant condition in Sal forest at low elevation. The species *Adiantum venustum*, *Asplenium ensiforme*, *Dryopteris chrysocoma*, *Lepisorus loriformis*, *Lepisorus scolopendrium*, *Polypodiodes lachnopus*, *Onychium lucidum*, *Pteris wallichiana*, *Vittaria linearifolia*, *Pyrrosia flocculosa*, *Woodwardia unigemmata* etc were present at moist places of *Schima-Castonopsi*, *Schima-Querscus*and *Schima-Diploknema* forests at high altitude (Figure 5 & Table 1).

The result shows that Arghakhanchi is rich in pteridophyte diversity due to presence of about 13% of total pteridophyte species of Nepal. Pteridaceae

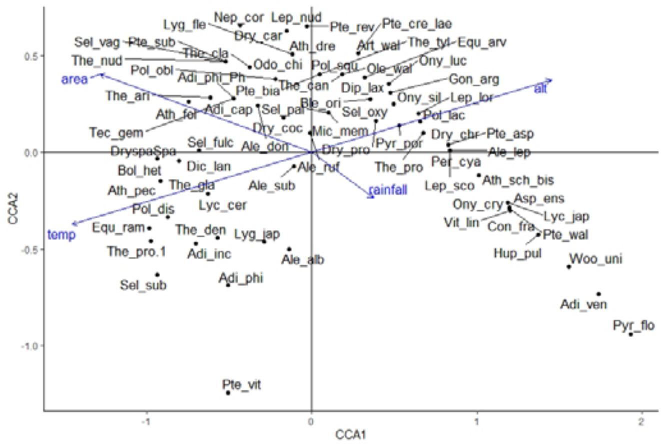

Figure 5: CCA biplot showing relationship among species and environment variables

is the richest family and Polypodiceae lies in second position. Polypodiaceae (polypod ferns) with 4,080 species is the largest family of ferns in world (State of the World Plant [SOTWP], 2017). There are of 8 monogeneric families and 23 genera consist of single species. This fact also indicates the variation in fern diversity.

Most of the fern species are terrestrial which are followed by lithophytes and epiphytes. The Presence of more terrestrial species indicates the favorable condition on land for fern diversity in this area. The species *Oleandra wallichii*, *Drynaria propinqua*, *Lepisorus loriformis*, *Lepisorus nudus* etc. are inhabiting on the bark of perennial trees as *Schima-Castanopsis*, *Castonopsis-Quercus forest of* areas above than 1500 m asl. The presence of more epiphytic specieson higher altitude possibly occurs due to increasing environmental humidity (Acebey et al., 2017). Shrestha & Rajbhandary (2019) also found the most of the epiphytic species on barks of *Quercus*, *Acer* & *Betula* species. The epiphytic ferns cannot survive as the forest become more and more open (Edward et al., 2003), or they lie mainly on ecologically undisturbed forest. Presence of epiphytic fern indicates that some forest of this area provides suitable place for epiphytic ferns.

The fern species shows dome shaped pattern against altitude, area and interpolated temperature and rainfall. Lili et al. (2014) found the hump shaped pattern of fern against altitude, area, mean annual temperature and mean annual precipitation. Bhattarai et al. (2004) also reported unimodal pattern of fern species against altitude in central Nepal with maximum species at 2000 m. The richness of terrestrial fern species exposes the mid elevation maximum (Watkin et al., 2006).The species richness plotted the peak value at 1300 m against altitudeand sharply decreased onwards in this study. This decreased altitude of highest modeled species richness than Bhattarai et al. (2004) may be the matter of scale and hard boundary effect due to low altitudinal range of species or narrow distribution. The random placement of range of species increases the overlap of species towards middle of geographical domain and results hump shaped (Colwell & Lees, 2000). This shows that species richness generally increases with increase of elevation and decrease of temperature and fitness of unimodal pattern of fern species looks stronger with altitude and temperature ($R^2 = 0.95$) than area and rainfall.

Rainfall does not show clear relation with temperature and altitude, but temperature showed opposite relation with altitude. However, these parameters showed effect in distribution pattern of species. No single variables can explain for distribution of plants (McCain & Grytnes, 2010). Altitude determines the climatic condition of any place and is main controlling factor of altitudinal species richness than others Li & Feng, 2015.The actual mechanism controlling the patterns lies in a combination of factors related to biology, the environment and geometric constrains on geographical range (Watkins et al., 2006).The area size can also significantly affect relationship between species richness and elevation and covers the effects of other predictors (Xiang & Hua-yong, 2017). Generally, area decreases with the increase of altitude in the Himalaya region.The increasing area per elevation band up to mid altitudinal range may be played main role to develop the unimodal pattern of species richness.

According to Acebey et al. (2017) the understory is a very important habitat for epiphytic ferns and others depending on the elevational zone. The different microhabitats in the forest understory determine the high diversity of epiphytes (Kromeret al., 2007).The number of epiphytes found in dense and moist forests is comparatively higher than those found on trees in open and dry areas (Rajbhandary, 2016). The species like *Adiantum venustum, Asplenium ensiforme, Dryopteris chrysocoma, Lepisorus loriformis, Lepisorus scolopendrium, Polypodiodes lachnopus, Onychium lucidum, Woodwardia unigemmata* etc. are restricted at Schima-Castonopsis- Quercus forest above than 1500m which covers about 25% of total species.

The study shows that most of the fern species are moist loving and present at north aspect of hills.

Conclusion

On the floristic study of Pteridophyte, 75 species belonging to 18 families and 36 genera were found in Arghakhanchi**.** Peridaceae consisting of 5 genera and 21 species was found the richest family. Most of the species (39) were located as terrestrial. The species richness showed the statistically significant unimodal pattern against altitude and temperature. Most of the fern species were present at moist places of *Schima-Castonopsis-Quercus* forest above than 1500 m. The further detail systematic study of plants including pteridophyte and their ecological status in this district is necessary.

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References

Acebey, A.R., Kromer, T. & Kessler, M. (2017). Species richness and vertical distribution of ferns and lycopods along elevational gradients in Los Tuxtlas Veracruz, Mexico. *Flora*, *235*, 83-91. www.elsevier.com/locate/flora.

Acharya, K.P., Chaudhary, R.P. & Vetaas, O.R. (2010).Medicinal plants of Nepal: Distribution pattern along an elevational gradient and effectiveness of existing protected areas fortheir conservation. *Banko Janakari*, *19* (1), 16-22.

Acharya, K.P., Vetaas, O.R. & Birks, H.J.B. (2011). Orchid species richness along Himalayan elevation gradients. *Journal of Biography*, 38, 1821-1833.

Alternative energy promotion center. (2016). *District Climate and Energy Plan of Arghakhanchi District*, (pp. 84). National Rural and Renewable Energy Program, p.84 Lalitpur, Nepal: Author.

State of the World Plant. (2017). *Naming and counting the world's plant families*, (pp. 5-9). Kew, UK: Royal Botanic Gardens.

Baniya, C.B., Solhøy, T., Gauslaa, Y. & Palmer, M. W. (2010). The elevation gradient of lichen species richness in Nepal. *The Lichenologist*, *42*(1), 83-96.

Bhattarai, K.R. & Vetaas, O.R. 2003. Variation in plant species richness of different life-forms along a subtropical elevation gradient in the Himalayas, East Nepal. *Global Ecology &Biogeography*, *12*(4), 327-340. Doi:10.14046/j.1466-822x.2003.00044x.

Bhattarai, K.R., Oleg, O.R. & Grytnes, J.A. (2004). Fern species richness along a central Himalayan elevational gradient, Nepal. *Journal of Biogeography*, *31*, 389-400.

Central Bureau of Statistics. (2012). *National Population and Housing Census 2011* (National Report). National Planning Commission Secretariat, Kathmandu, Nepal: Author.

Colwell, R.K. & Lees, D.C. 2000. The mid domain effect: geographical constrains on the geography of species richness, *Trends in Ecology & Evolution*, *15*, 70-76.

Department of Hydrology and Meteorology. (2017). *Observed Climate Trend Analysis in the Districts and Physiographic Zones of Nepal (1971-2014*), (pp. 93), Kathmandu, Nepal : Author.

Edward, A.E., Charles, M.M. & Gebhard, B.L. (2003). Studies on epiphytic ferns as potential indicators of forest disturbances, XII Forestry Congress, Quebec City, Canada. Proceedings.

Fraser-Jenkins, C.R., Kandel, D.R. & Pariyar, S. (2015). *Ferns and Fern-allies of Nepal*, Vol.1, (pp. 492), Kathmandu, Nepal : Department of Plant Resources.

Fraser-Jenkins, C.R. & Kandel, D.R. (2019). *Ferns and Fern-allies of Nepal*, Vol. 2, (pp. 492), Kathmandu, Nepal : Department of Plant Resources.

Grau, O., Grytnes, J. A. & Birks, H.J. B. 2007. A Comparison of altitudinal species richness patterns of bryophytes with other plant groups in Nepal, Central Himalaya. *Journal of Biogeography*, *34*, 1907-1915.

Gurung, V.L. (1984). Ferns of Nepal, Majupuria, T.C. (Ed), *Nepal Natures Paradise*, p. 198-211.

Gurung, V.L. (1991). *Ferns the beauty of Nepalese flora*. Kathmandu, Nepal: Sahayogi press Pvt. Ltd.

Hastie, TJ. & Tibshirani, RJ. (1990). *Generalized additive models*. New York/Boca Raton: Chapman & Hall/CRC.

Jeyalatchagan, S.K., Ayyanar, M. & Silambarasan, R. (2020). Pteridophyte species richness along elevation gradients in Kolli Hills of the Eastern Ghats, India. *Journal of Asia-Pacificiodiversity*, *13*(1), 92-106. https://doi.org/10.1016/j.japb.2019.11.008

Kent, R. & Carmel, Y. (2011). Presence-only versus presence–absence data in species composition determinant analyses. *Diversity and Distributions*, *17*, 474–479. DOI:10.1111/j.1472-4642.2011.00755.x 474 http://wileyonlinelibrary.com/journal/ddi

Khare, P.B. (1996). Ferns and fern allies their significance and fantasies. *Applied Botany Abstracts*, *16*(1), 50-61.

Kromer, T., Kessler, M. & Gradstein, S.R. (2007). Vertical stratification on vascular epiphytes in submontane and montane forest of Bolivia Andes. *Plant Ecology*, *189*, 261-278.

Li, M. & Feng, J. (2015). Biogeographical Interpretation of Elevational Patterns of Genus Diversity of Seed Plants in Nepal. *Plos One*, *10*(10), 1-32. https://doi.org/10.1371/journal.pone.0140992

McCain, C.M. & Grytnes, J.A. (2010). Elevational Gradients in Species Richness, *Encyclopedia of Life Sciences*. p 1-10, USA: John Wiley & Sons, Ltd.https://sci-hub.tw/https://doi.org/10.1002/9780470015902.a0022548

Ministry of Agriculture and Co-operatives. (2011). *District Disaster Risk Management Plan*: *Arghakhanchi District*, (pp. 101), Kathmandu, Nepal: Author.

Moran, R.C. (2004). *A natural history of ferns*. Portland: Timber Press.

Nagalingum, N.S. & Cantrill, D.J. (2015). The Albian fern flora of Alexander Island, Antarctica. *Cretaceous Research*, *55*, 303-330. http://dx.doi.org/10.1016/j.cretres.2015.02.005 0195-6671/

Panthi, M.P. & Chaudhary, R.P. (2002). Angiospermic flora of Arghakhanchi district and adjoining areas, west Nepal, *Natural History Museum*, *21*, 7-21.

Rahbek, C. 1997. The elevational gradient of species richness, a uniform pattern? *Ecography,18*, 200-205.

Rajbhandary, S. (2016). Fern and Fern Allies of Nepal. In Jha, P.K., Siwakoti, M. and Rajbhandary, S.(ED.). *Frontiers of Botany* (pp. 124-150). Kathmandu, Nepal: Central Department of Botany, Tribhuvan University.

R Core Team. (2019). *R: A language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing.

Shrestha, H.S. & Rajbhandary,S. (2019). Floristic study of ferns and fern allies along altitudinal gradient from Besishahar to lower Manang, Central Nepal. *Journal of Plant Resources*, *17*(1), 29-34.

Vidyashree Chandrashekar, S.Y., Hemla Naik, B., Jadeyegowda, M., & Revannavar, R. (2018). Diversity of Fern Flora for Ecological Perspective – A Review, *International. Journal of Pure & Applied Bioscience*, *6*(5), 339-345. doi: http://dx.doi.org/10.18782/2320-7051.6750

Watkins, J.E., Cardelus, C., Colwell, R.K. & Moran, R.C. (2006). Species richness and distribution of ferns along an elevational gradient in Costa Rica. *American Journal of Botany*, *93*(1), 73–83.

Xiang X. & Hua-yong, Z. (2017). Area corrected species richness patterns of vascular plants along a tropical elevational gradient, *Journal of Mountain Science*,*14* (40), 694-704. http://link.link.springer.com

Zhang, S., Chen,W. & J. Huang 2015. Orchid species richnessalong elevational and environmental gradients in Yunnan, China. *PLoS ONE, 10*(10), 371. /journal.pone.0142621

Table 1: Name list of Pteridophyte found in Arghakhanchi district

S.N.	Family	Name of species	Habitat	Altitude Range	*Location (® Type of Forest, Altitude)
1	Aspleniaceae	*Asplenium ensiforme* Wall. ex Hook. & Grev.	Epi	1600-1900	Ml(SQ.1610),Go(CS,1890)
2	Athyriaceae	*Athyrium drepanopterum* (Kunze) A. Braun ex Milde	Tere	1000-1700	Bh(SP,1520), Da(PS,1010), Ka(SC,1680)
3	Athyriaceae	*Athyrium foliolosum* T.Moore ex R.Sim	Tere	800-1400	Bh(SP,820),Sa(SP,1400)
4	Athyriaceae	*Athyrium pectinatum* (Wall ex Mett.) T. Moore	Tere	600-1300	Ba(S,620),Th(S,1320);Ra(S,1100)
5	Athyriaceae	*Athyrium schimperi* Moug.ex Fee subsp biserrulatum (Christ)	Tere	1500-1900	Se(SC,1915), Dh(SC,1510)
6	Athyriaceae	*Diplazium laxifrons* Rosenst.	Tere	1100-1900	Ml(SQ,1880); Ro(CS,1090)
7	Blechnaceae	*Blechnum orientale* L.	Tere	1100-1700	Dn(SC,1110), Se(SC,1685)
8	Blechnaceae	*Woodwardia unigemmata* (Makino) Nakai	Tere	1700-2000	Ml(SQ,1730;1990)
9	Dennstaediaceae	*Pteridium revolutum* (Blume) Nakai	Tere	1200-1600	Ml(SQ,1610),Sa(CS,1220)
10	Dryopteridaceae	*Bolbitis heteroclita* (C. Presl) Ching	Tere&Litho	800-1200	Hi(S,820), Ra(S,1190)
11	Dryopteridaceae	*Dryopteris carolihopei* Frasser-Jenk	Tere	1100-1600	La(CS,1080), Gl(CS,1380), Dn(SC,1580)
12	Dryopteridaceae	*Dryopteris chrysocoma* (Christ) C. Chr.	Tere	1300-2000	Se(SC,1985),Sa(CS,1310)
13	Dryopteridaceae	*Dryopteris cochleata* (D. Don) C. Chr.	Tere&Litho	800-1800	Ny(S,820), Ka(SC,1770)
14	Dryopteridaceae	*Dryopteris sparsa* (D.Don) Kuntze subsp. Sparsa	Tere	700-1400	Sa(SC,1410), Bh(SP,1120),Ra(S,710)
15	Dryopteridaceae	*Peranema cyatheoides* D.Don	Tere	1400-1900	Poudi khola(PS,1425), Ga(SQ,1910)
16	Dryopteridaceae	*Polystichum discretum* (D.Don) Sm	Tere	600-1400	Gl(CS,1390),Do(S,610),
17	Dryopteridaceae	*Polystichum obliquum* (D. Don) T. Moore	Tere	900-1700	Ro(CS,910),Ha(CS,1120), Kh(RS,1690)
18	Dryopteridaceae	*Polystichum squarrosum* (D. Don) Fée	Tere&Litho	1000-1800	Kh(RS,1780),Sa(SC,1340),Bh(SP,1020)
19	Equisetaceae	*Equisetum arvense* L.	Tere	1200-1800	Na(SQ,1810), Po(PS,1225)
20	Equisetaceae	*Equisetum ramosissimum* Desf.	Tere	700-1200	Kc(W,885), Bh(SP,1120), Du(W,720)
21	Gleicheniacea	*Dicranopteris lanigera* Fraser-Jenk.	Tere	700-1400	Th(S,1380), Pk(810),Si(690)
22	Lindsaeaceae	*Odontosoria chinensis* (L.) J. Sm.	Tere	900-1600	Bh(SP,930),Da(SP,1595)
23	Lycopodiaceae	*Huperzia pulcherrima* (Wall. ex Hook. & Grev.) T. Sen & U. Sen	Epi	1600-2000	Kh(RS,1630), Ms(SC,2020)
24	Lycopodiaceae	*Lycopodiella cernua* (L.) Pic. Serm.	Tere& Lit	600-1600	Si(SC,625), Na(PS,1582)
25	Lycopodiaceae	*Lycopodium japonicum* Thunb.	Tere	1600-1850	Ka(SC,1590; 1780)
26	Lygodiaceae	*Lygodium flexuosum* (L.) Sw.	Tere	1000-1700	Am(S,960),Bh(SP,1650)
27	Lygodiaceae	*Lygodium japonicum* (Thunb.) Sw.	Tere& Lit	500-1900	Do(S,520), Am(S,950),Wa(S,1340),Ag(PS,1890)
28	Nephrolepidaceae	*Nephrolepis cordifolia* (L.) C. Presl	Tere	1000-1500	Ny(SC,1115), Ma(SC,M,1300),Gl(CS,1480)
29	Oleandraceae	*Oleandra wallichii* (Hook.) C. Presl	Epi	1100-1800	Se(SC,1785), Gl(CS,1120), Ml(SQ,1685)
30	Polypodiaceae	*Arthromeris wallichiana* (Spreng.)Ching	Epi	1300-1700	Ma(SS,1310), Ka(SC,1680)
31	Polypodiaceae	*Drynaria propinqua* (Wall. ex Mett.) Bedd.	Epi	1000-2000	Se(SC,1985), De(SC,1550), Dd(CS,1020)
32	Polypodiaceae	*Goniophlebium argutum* (Wall. ex Hook.) J. Sm. ex Hook.	Epi	1400-1700	Ja(P,1420), Ml(SQ,1710)
33	Polypodiaceae	*Lepisorus loriformis* (Wall. ex Mett.) Ching	Epi	1300-1900	Bh(SP,1290), Al(SC,1520),Na(SC,1880)
34	Polypodiaceae	*Lepisorus nudus* (Hook.) Ching	Epi & Lit	1200-1600	Dn(SC,1800), Dh(SC,1620),Da(SP,1220)
35	Polypodiaceae	*Lepisorus scolopendrium* Mehra& Bir	Epi	1400-1900	Bh(SP,1380),Se(SC,1910)
36	Polypodiaceae	*Microsorum membranaceum* (D. Don) Ching	Tere& Epi	900-1900	Rosa(CS,910),Se(SC,1890), Gl(CS,1498)
38	Polypodiaceae	*Pyrrosia flocculosa* (D. Don) Ching	Epi	1600-2000	Ml(SQ,1610; 2010)
39	Polypodiaceae	*Pyrrosia porosa* (C. Presl) Hovenkamp	Epi	1100-1900	Ka(SC,1780), Dd(CS,1120)
40	Polypodiaceae	*Selliguea oxyloba* (Wall. ex Kunze) Fraser-Jenk.	Epi	1000-2000	Ms(B,1950); Na(PS,1620); Da(CS,980)
41	Pteridaceae	*Adiantum capillus* veneris L.	Lit	800-1700	Bh(SP,780), Da(SC,1680)

42	Pteridaceae	*Adiantum incisum* Forssk.	Lit	500-1600	Am(S,1050), Ra(S,520), Na(SC,1610)
43	Pteridaceae	*Adiantum philippense* L.	Lit	400-1800	Do(S,390),Am(S,990),Na(SQ,1790), Ha(CS,1220)
44	Pteridaceae	*Adiantum philippense* L.subsp. philippense	Lit	800-1600	Bh(SP,810), Na(SC,1580)
45	Pteridaceae	*Adiantum venustum* D.Don	Lit	1700-2100	Ml(SQ,1710;2080)
46	Pteridaceae	*Aleuritopteris albomarginata* C.B. Clarke	Lit	500-2000	Do(S,480);Ar(PS,1770), Gl(CS,1190),Go(SQ,1970)
47	Pteridaceae	*Aleuritopteris doniana* S.K. Wu	Lit	800-1700	La(CS,815), Sa(SC,1450),Di(PS,1705)
48	Pteridaceae	*Aleuritopteris leptolepis* (Fraser-Jenk.) Fraser-Jenk.	Lit	1400-1900	Se(SC,1880), Gl(CS,1415)
49	Pteridaceae	*Aleuritopteris rufa* (D. Don) Ching	Lit	800-1900	Se(SC,1880), Ar(PS,1710),La(CS,790)
50	Pteridaceae	*Aleuritopteris subdimorpha* (C.B. Clarke & Baker) Fraser-Jenk.	Lit	700-1900	Se(SC,1890), Gl(CS,1415), Ha(CS,900),Ba(S,710)
51	Pteridaceae	*Coniogramme fraxinea* (D.Don) Fee ex Diels	Tere	1700-1900	Ml(SQ,1690;1910)
52	Pteridaceae	*Onychium cryptogrammoides* Christ	Tere	1500-2000	Se(SQ,1920), Da(PS,1520)
53	Pteridaceae	*Onychium lucidum* (D. Don) Spreng.	Tere	1300-1800	Se(SC,1780),De(SC,1305)
54	Pteridaceae	*Onychium siliculosum* (Desv.) C. Chr.	Tere& Lit	1200-1900	Bh(S,885), Ml(SQ,1780), Se(SC,1880)
55	Pteridaceae	*Pteris aspericaulis* Wall. ex J. Agardh	Tere	1300-2000	Ms(B,2020), Na(SC,1710) , Bh(SP,1320)
56	Pteridaceae	*Pteris biaurita* L.	Tere& Lit	1000-1800	Se(SC,1780),Am(S,980)
57	Pteridaceae	*Pteris cretica* var. laeta (Wall. ex Ettingsh.) C. Chr. & Tardieu	Tere	1300-1700	Bh(SP,1320),Na(SC,1680)
58	Pteridaceae	*Pteris subquinata* Wall. ex J. Agardh	Tere	900-1500	Th(S,1240), Pk(CS,840), Sa(SC,1480)
59	Pteridaceae	*Pteris vittata* L.	Tere& Lit	200-1900	Se(SQ,1875), Gl(CS,1320), Am(S,1070), Do(S,230)
60	Pteridaceae	*Pteris wallichiana* J. Agardh	Tere	1400-2100	Kh(SQ,2065), Pn(CS,1400)
61	Selaginellaceae	*Selaginella fulcrata* (Buch.-Ham. Ex D.Don) Spring	Tere	700-1500	Kc(W,770),Bh(SC,1205), Gl(CS,1490)
62	Selaginellaceae	*Selaginella pallida* (Hook. &Grev.) Spring	Tere	900-1900	Ro(CS,910),Se(SQ,1880), Gl(CS,1380)
63	Selaginellaceae	*Selaginella subdiaphana* (Wall. Ex Hook & Grev.) Spring	Tere	500-1400	Do(S,505),Bh(SP,1160), Gl(CS,1410)
64	Selaginellaceae	*Selaginella vaginata*	Tere& Lit	900-1500	La(CS,910),Gl(CS,1480)
65	Tectariaceae	*Tectaria gemmifera* (Fée) Alston	Tere& Lit	800-1600	Bh(S,810),Pn(CS,1350), Gl(SC,1580)
66	Thelypteridaceae	*Thelypteris arida* (D.Don) C.V.Mortum	Tere	800-1500	Du(W,780),Gl(CS,1470), Bh(SP,1120)
67	Thelypteridaceae	*Thelypteris cana* (J.Sm.) Ching	Tere	1100-1800	Pa(SS,1325),Th(SS,1120), Ka(SC,1810)
68	Thelypteridaceae	*Thelypteris clarkei* (Bedd.) C.F.Reed	Tere	900-1600	Ra(S,910), Ja(PS,1220), Ml(SC,1610)
69	Thelypteridaceae	*Thelypteris dentata* (Forssk.) E.P. St. John	Tere	500-1700	Do(S,520),Th (SS,1250), Da(SC,1670)
70	Thelypteridaceae	*Thelypteris glanduligera* (Kunze) Ching	Tere	600-1600	Si(SC,650), Sa(SC,1420),Gl(SC,1590)
71	Thelypteridaceae	*Thelypteris nudata* (Roxb.) C.V. Morton	Tere	900-1500	Bh (S,920),De(PS,1260), De(SC,1510)
72	Thelypteridaceae	*Thelypteris procera* (D.Don) Fraser-Jenk	Tere	1200-1900	Se(SQ,1910), Bh(SP,1220)
73	Thelypteridaceae	*Thelypteris prolifera* (Retz.) C.F.Reed	Tere	600-1300	Do(S,580), ,Th(S,1310)
74	Thelypteridaceae	*Thelypteris tylodes* (Kunze) Ching	Tere	1000-1800	Se(SC,1780), Pn (CS,1020)
75	Vittariaceae	*Vittaria linearifolia* Ching	Lit	1300-2000	Dh(SC,1620),Ms(RS,1980), Pn(SP,1290)

Note: Ag-Argha, Al-Alamnagar, Am-Amadanda, Ar-Arichour, Ba-Baseri, Bh-Bhuwandanda, Da-Dahakhola, Dd-Daduwa, Dh-Dhakaband, Di-Diverna, Dn-Dhanchour, Do-Dohote, Du-Durghaphat, Ga-Gargare, Gl-Goldhung, Go-Gokhunga,Ha-Halde, Hi-Hilekhola, Ja-Jalkanda, Ka-Kalikathi, Kc-Khanchikhola, Kh-Khanchi, la-Lamdanda, Ma-Maidan, Ml-Malarani, Ms-Masina, Na-Narapani, Ny-Nayagaun, Pa-Patauti, Pn-Panku, Pk-Pokhadanda, Po-Poudikhola, Ra-Rajiya, Ro-Rosa, Sa-Sanodeva, Se-Sengleng, Si-Sitkhola, Th-Thada & Wa-Wangla

Types of forest: S= Sal, SC=Schima-Castonopsis, SQ= Schima-Quercus; SP=Sal-Pinus; SS=Sal-Schima; P=Pinus; CS=Chiuri-Sal; PS=Pinus-Schima, RS=Rhododendron-Schima; W=Wetland; B=Bushes

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Status, Distribution and Habitat Suitability Mapping of *Cycas pectinata* in Chure Range of Makawanpur, Central Nepal

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Abstract

Cycas pectinata is an evergreen plant with no branches and having a single above-ground trunk. It is listed in the vulnerable category of IUCN and appendices II of CITIES. The study was carried out in the Chure range of Makawanpur district, Central Nepal. Field data were collected to assess status, distribution and habitat variables. Data were analyzed through descriptive statistics and the analytical hierarchy process in integration with Arc GIS 10.2.2 to prepare habitat suitability maps. A totalof 780 individuals of *Cycas* were recorded in scattered form. The major preferred habitat of this species was forest land having elevation range of 400 – 500m mostly on steep terrain with slope range from 25° to 55°, nearer of small gullies and far from human influences. This study revealed that, the Chure range wasa suitable habitat for *Cycas* of which 40.52% area was highly suitable, 56.08% area was moderately suitable and 3.40% area as less suitable. Though it is in danger of extinction, very limited research about its status, distribution, reproduction biology, and habitat suitability have been conducted. This paper aims to determine the status, distribution and habitat suitability of *Cycas* in the Chure range of Makwanpur district.

Keywords: *Cycas,* Scattered distribution, Suitable habitat, Vulnerable

Introduction

Cycas pectinata Buch.-Ham.is a dioecious tall palm-like tree growing up to 2-3 m high, with a single aboveground trunk and crowns formed by large, evergreen and pinnate leaves (Singh & Singh, 2010; Joshi et al., 2017). The male cones of this species are usually large, cylindrically ovoid, and yellowish or orange in maturity while female cones are huge and compactly filled with megasporophylls which are deeply pectinate and densely covered with hairs (Singh & Singh, 2010). Seeds are flattened ovoid, glabrous green when immature and orange-red at maturity (Pant & Osborne, 2002).

The plant is famous due to its multiple uses in ornamental, medicinal, food, and socio-cultural values. Local communities have been usedvarious parts of the plant for several purposes since long back. Sago or stem starch obtained from the stem has been used as an alternative source of food for many centuries in many countries (Jones, 2002). The young, circinate, succulent leaves and young tubers are consumed as vegetables (Joshi et al., 2017). Fertilized ovules or immature seeds are eaten as food and are also used as medicine for gastric problems (Patiri & Borah, 2007) whereas in some areas microsporophylls are also used to cure asthma while megasporophylls are used for treating piles (Das & Dutta, 2007).

The species usually grows at an elevation of 300 m to 1200 m in difficult terrains of Chure hills and lower foothills with medium to a tall forest on deep, often clay-rich and more fertile soils, usuallyin the moist understory in moderate to deep shade in central and eastern Nepal. It prefers tropical climate with wet, humid summers and milder, drier winters. Although often found on limestone substrates, it is by no means restricted to these, and it also occurs on granites and meta-sediments (Lindstrom & Hill, 2007; Nguyen, 2010). The forests of the Chure region in central and eastern Nepal provide important habitat for *Cycas pectinata* where they are found in either the forest of *Shorea robusta*, *Shorea-Schima*, or riverside up to 730m elevation. Bhuju& Joshi (2009) reported 36 plants of *Cycas pectinata* from 15 sites in the Churiya hills of eastern Nepal.

However, due to the continuous decline in a natural population, it is listed in Appendices II of CITES and the conservation status of the species is endangered (Joshi et al., 2017). The species is assigned to the Vulnerable (VU) category by IUCN due to an estimated decline in a habitat of more than 30% over the past 90 years (Nguyen, 2010).

A habitat is a unique place for a certain species to develop and it is often referred to as species-specific concept (Fischer & Lindenmayer, 2007). Mapping typically involves the identification and demarcation of the spatial extent of specific attributes. Geographical Information System (GIS) application has been adopted in ecological modeling as tools for producing the data needed in modeling on different spatial and temporal scales (Wu & Smeins, 2000), as platforms on which models are run and data stored (Hirzel et al., 2001), and as tools for extrapolating the results from point basis to spatial basis (Osborne et al., 2001).

Land-use change is a major factor that modifies the habitat and can determine the distribution of species within an ecosystem (Musiega & Kazadi, 2004). As habitat indicates 'where' a species lives and 'what' is the environment type where a species lives, mapping and monitoring the wildlife habitat are the important aspects in assessing the quality of habitat (Wintleet al., 2005). To avoid the extinction of endangered species, it is important to understand and manage their habitats (Ortigosa et al., 2000). Thus there is a need for information that can assist with locating suitable habitats in fragmented and degraded landscapes to aid the reintroduction of at-risk plant species (Questad et al., 2014). Integration of GIS, Remote Sensing, and Global Positioning System (GPS) technologies have proved to be effective in the assessment of habitat quality (Memarbashi et al., 2017). *Cycas pectinata* is one of the widest spread cycadsthat are now under threat and its populations are declining at pace as compared to any other species of *Cycas* (Singh & Singh, 2014). Though this species is in the vulnerable condition of extinction, very few similar research has been conducted of the species to document its habitat, status, and distribution. This paper aims to determine the status, distribution and habitat suitability mapping of *Cycas pectinata*, in the Chure range of Makwanpur district.

Materials and Methods

Study area

This study was carried out in Chure range of Makawanpur district (Figure 1) which covers 140,297 ha. of the southern part of Makawanpur

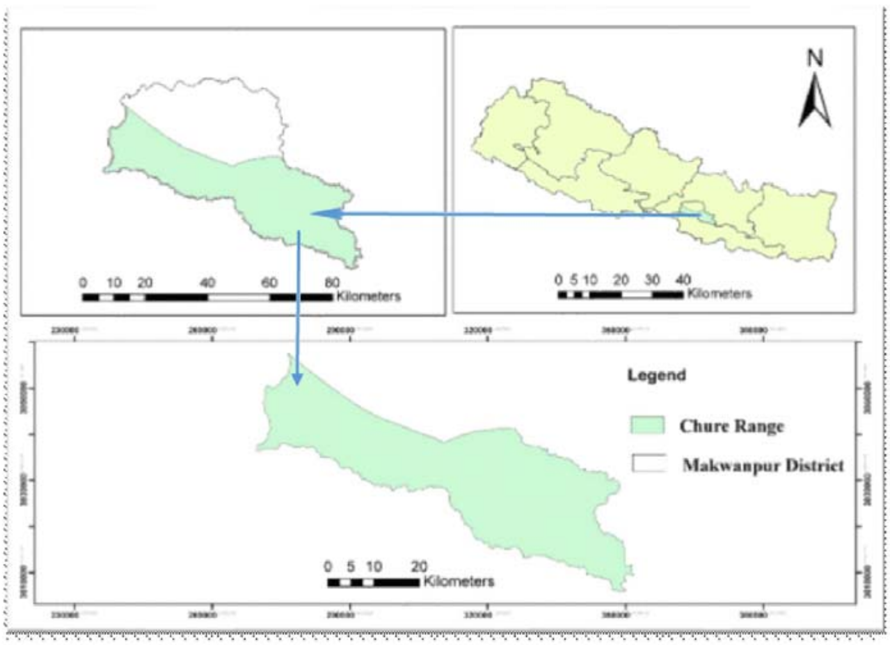

Figure 1. Map of Study Area

district (President Chure-Tarai Madhesh Conservation Development Board [PCTMCDB], 2015) mainly consisting of fluvial sedimentary rocks such as mudstone, shale, sandstone, siltstones, and conglomerates naturally being a very sensitive area (Ghimire, 2016). The climate of the Chureregion ranges from tropical to warm temperate and is characterized by hot and sub-humid summers, intense monsoon rain, and cold dry winters where *Shorea robusta*, *Dalbergia sissoo*, *Acacia catechu*, *Adina cordifolia*, etc. were the major tree species (Department of Forest Research and Survey [DFRS], 2014) whereas Tamang, Chepang, Rai, Brahmin, Chhetri, etc. were the major ethnic groups (PCTMCDB, 2015).

Data collection

A preliminary reconnaissance survey was carried out initially to identify the pocket areas of *Cycas*, identification and ranking of factors which affected the habitat of *Cycas*. Ten focus group discussions and key informant interviews with Divisional Forest Office, Plant Research Centre Office, local peoples and community forest user groups were carried out.Based on information collected from discussions, an extensive survey of the identified potentials area were conducted with local resource persons during March-April, 2019.The relevant habitat parameters like altitude, aspect, slope, associated species, soil type, nearest distance from the water body, road, and settlement were recorded. The GPS coordinates of all *Cycas* plant sighted were recorded for the mapping purpose. Secondary data were sourced from relevant journals, books and web pages.

Data analysis

All together nine variables viz. land use land cover, forest type, aspect, slope, elevation, proximity to the river, soil type, proximity to the road, and proximity to the settlement were perceived as their prominent influences in habitat dynamics of this species. Integration of Analytical Hierarchy Process and Arc GIS 10.2.2 was used for the suitability mapping purpose.

Results and Discussion

Status and distribution of Cycas

All together 780 *Cycas* plants were recorded, out of which 16 were found as planted populations and 764 were found as natural populations.The highest number of *Cycas* were found as indeterminate i.e. male and female cone was not distinguished. The plants were not distributed uniformly rather in a patch or as a cluster and occurred in different micro-habitat with densely populated in each area found.

Habitat suitability based on multiple variables

With the Classified Landsat OLI imagery of 2018, Aster Global DEM, SOTER, and Google Earth, the following nine layers were prepared and ranked as less suitable, moderately suitable, and highly suitable. The suitability classification was done based on focus group discussion, expert opinions, and field data collected during the March- April 2019.

Habitat suitability based on land use land cover (LULC)

Forest was classified asa highly suitable area in which 82% of total *Cycas* were found. Agricultural land use was classified as moderately suitable where 15% of total *Cycas* were found. Other land-use classeswere classified as less suitable where 3% of total *Cycas* were found (Figure 2A).

Habitat suitability based on forest type

Sal forest was classified as the highly suitable forest type in which 82% of total *Cycas* were found. Ravine broad-leaved forest was classified as moderately suitable forest type 16% of total *Cycas*. Other forest types were classified as less suitable forest types in which 2% of total *Cycas* were found (Figure 2B).

Habitat suitability based on the slope and the aspect

Slope class of 25°-55° was classified as highly suitable in which 62% of total *Cycas* were found. Slope class of 0-25 degree was classified as moderately suitable in which 20% of total *Cycas* were found and slope class of 55-90 degree was classified as less suitable in which 18% of total *Cycas* were found (Figure 5). Similarly, Northeast, Southeast, Southwest aspects were classified as highly suitable aspects in which 76% of total *Cycas* were found followed by North, West, and Northwest (Figure 2C & 2D).

Habitat suitability based on elevation

Elevation of 400-500m was classified as ahighly suitable area in which 80% of total *Cycas* were found. Elevation below 400m was classified as moderately suitable in which 12 % of total *Cycas*

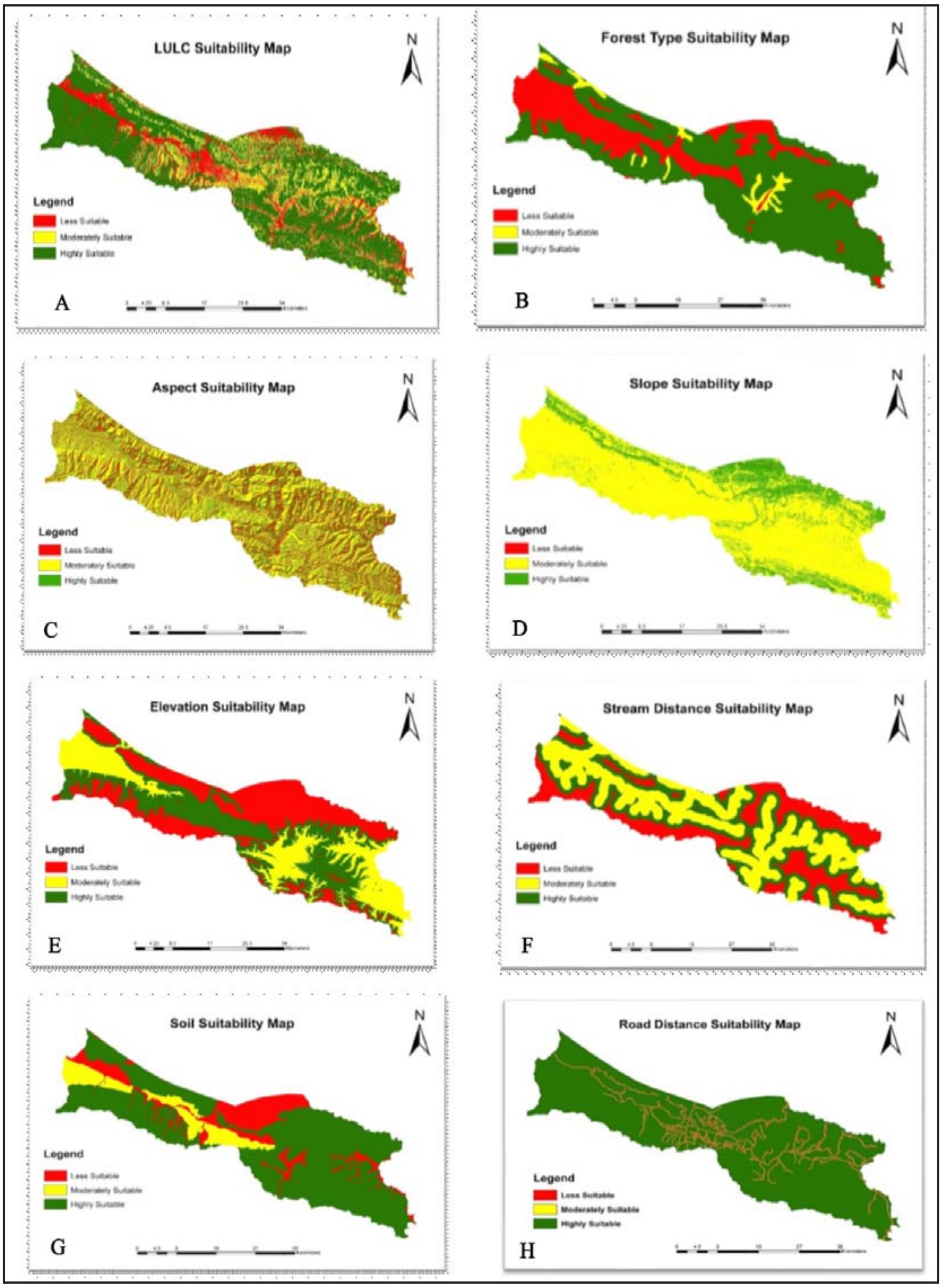

Figure 2: Habitat suitability map: A. LULC suitability map; B. Forest type suitability map; C. Aspect suitability map; D. Slope suitability map; E. Elevation suitability map; F. Stream distance suitability map

were found. Elevation above 500m was classified as the less suitable area in which 8% of total *Cycas* was found (Figure 2E).

Habitat suitability based on proximity to the river

The area which is present in between 1000 to 2000 meter distance from the river was classified as the highly suitable area in which 63% of total *Cycas* were found. The area which is present below 1000m distance from the river was classified as the moderately suitable area in which 28% of total *Cycas* were found. The area which is present above 2000m distance from the river was classified as the low suitable area in which 9% of total *Cycas* were found (Figure 2F).

Habitat suitability based on soil type

Eroded Soil i.e. Dystric Regosols (RGd) was classified as highly suitable soil in which 98.33% of total *Cycas* were found, Well-drained Soil i.e. Haplic Phaeozems (PHh) was classified as moderately suitable soil in which 1.54% of total *Cycas* were found and other soil types were classified as less suitable soil in which 0.13% of total *Cycas* were found (Figure 3A).

Habitat suitability based on proximity to road and settlement

The area which is present above 100m distance from both road and settlement was classified as highly suitable to flourish the species. About 75% of the total population was found about 100m far from the settlement. The population declined largely on proximity of the road and settlement (Figure 3B and 3C) thus indicating the impact of unplanned road construction and settlement in this and other similar threatened species.

Habitat suitability based on combined variables

The final suitability map was prepared by overlaying layers of nine factors using a weighted overlay procedure according to their weight obtained from AHP. The Red, Yellow, and Green color indicated the less, moderately, and highly suitable habitat for *Cycas* respectively (Figure 3). Out of 1, 40,297-hectare area of Chure, 40.52% falls under the highly

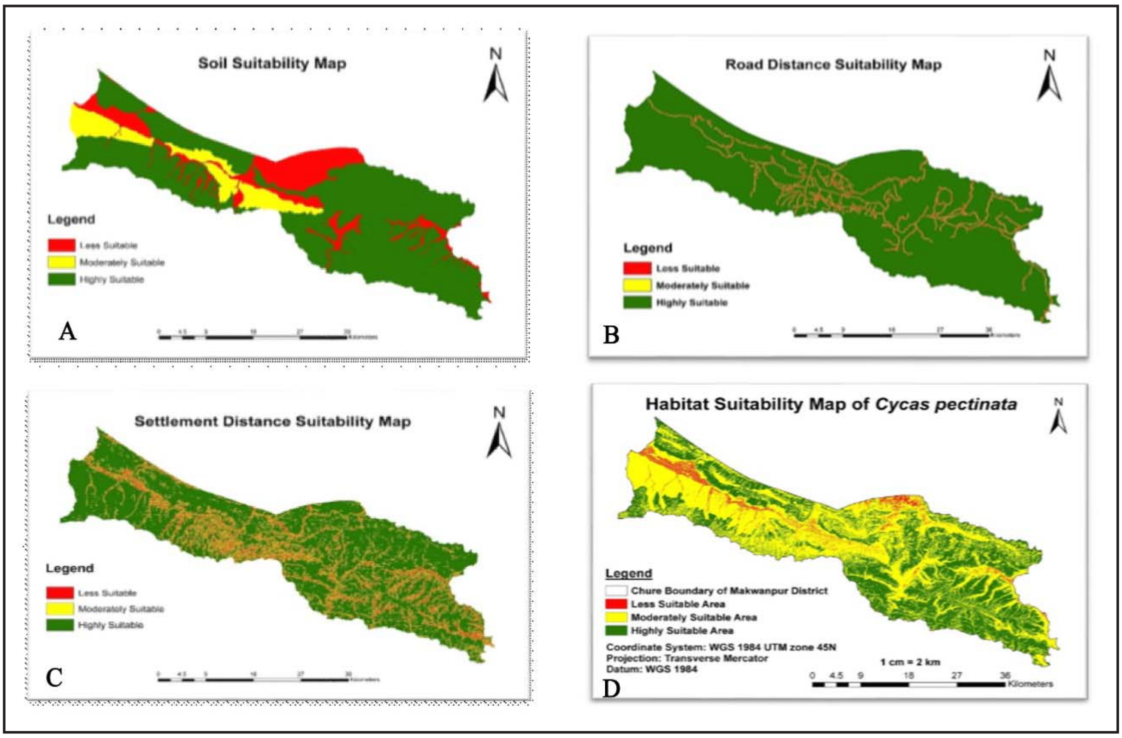

Figure 3: Habitat suitability map; A - Soil suitability map; B - Road distance suitability map; C - Settlement distance suitability map; D - Combined variables suitability map

suitable area, 56.08% falls under moderately suitable area and 3.40% falls under less suitable area (Figure 3D).

Field verification of habitat suitability map

After making the final suitability map, the map was analyzed through field verification. Out of total *Cycas* sighted during field verification, 38.20% were found on the highly suitable area, 61.80% were found on the moderately suitable area and no *Cycas* were found on the less suitable areas (Figure 12). Similar to the finding of this study, Bhuju & Joshi (2009) recorded 36 plants of *Cycas pectinata* from 15 sites in the Chure hills of eastern Nepal along with the altitudinal range up to 750m as an understory of *Shorea robusta, Shorea-Schima* forest.

Conclusion

The distribution of the species was observed in patches or clustersin different micro-habitat with densely populated in each area found. All together 780 number of *Cycas* were recorded. Out of which 27 were male, 35 were female and the rest of them being indeterminate. The number of reproductive plants was not proportional to the number of individual plants. The *Cycas* were distributed more in the elevation range from 400 m to 500 m in the Northeast, Southeast and Southwest aspect. Thesewere found mostly on steep terrain with slope range from 25° to 55° nearer of small gullies that are medium to near distance from perennial river sources. The habitat which wasfar from human influence was preferred most. Based on this study it can be concluded that about 40.52% of the total Chure area of Makawanpur is highly suitable, 56.08% moderately suitable and 3.40% to be less suitable habitat for *Cycas*.

Recommendations

- Limited research and information gap on population status, range of distribution, reproduction biology, phenology, and threatsto the species are causing them to be less prioritized so extensive survey and scientific exploration are necessary for proper documentation and conservation.
- The identified potential suitable areas for *Cycas* should be developed as a pocket area and conservation of these areas is of utmost importance.
- Care should be given to conserving the habitat of *Cycas* during infrastructure development.
- As *Cycas* is listed in CITIES Appendices II, a conservation action plan should be formulated and implemented to conserve this historic species.
- A community-based *Cycas* conservation program should be encouraged in the potential areas of *Cycas* habitat.

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References

Bhuju, D. R. & Joshi, G. P. (2009). Records of *Cytheaspinulosa* Wallich ex Hooker (Cytheaceae) & *Cycaspectinata* Griff. (Cycadaceae) from the Churiya Hills of Eastern Nepal. *Nepal Journal of Science and Technology*.*10*, 69-72.

Das, A. K. & Dutta, B. K. (2007). *Cycaspectinata*- a wonderful native plant remedy for piles and asthma in Southern Assam, N. E. India. *Ethnobotany*. *19*, 140-141.

Department of Forest Research and Survey. (2014). *Churia Forests of Nepal*. Forest Resource

Assessment Nepal Project. Kathmandu, Nepal: Author.

Fischer, J. & Lindenmayer, D. B. (2007). Landscape modification and habitat fragmentation: a synthesis. *Global ecology and biogeography*, *16*(3), 265-280.

Ghimire, P. (2016). Chure Conservation Area: Lessons for the Management and Use of natural and Biological Resources in Nepal. *South Asia Watch on Trade, Economics & Environment* (SAWTEE).

Hirzel, A. H., Helfer, V. & Metral, F. (2001). Assessing habitat-suitability models with a virtual species. *Ecological Modelling*, *145*(2-3), 111-121.

Jones, D. L. (2002). *Cycads of the World*. Washington DC, USA: Smithsonian Institution Press.

Joshi, N., Dhakal, K. S. & Saud, D. S. (2017). *Checklist of CITES Listed Flora of Nepal*. Kathmandu Nepal: Department of Plant Resources.

Lindstrom, A. J. & Hill, K. D. (2007). The genus *Cycas* (Cycadaceae) in India. *Telopea*, *11*(4), 463-488.

Memarbashi, E., Passel, S., Witlox, F., Mohajeri, F., Barati, A., & Azadi, H. (2017). Land-Use Suitability in Northeast Iran: Application of AHP-GIS Hybrid Model. ISPRS *International Journal of Geo-Information*, *6*(12), 396.

Musiega, D. E. & Kazadi, S. N. (2004). Simulating the East African Wildebeest migration patterns using GIS and remote sensing. *African Journal of Ecology*, *42*(4), 355-362.

Nguyen, H. T. (2010). *Cycaspectinata*. The IUCN Red List of Threatened Species.e.T T42062A10617695 http://dx.doi.org/10.2305/IUCN.UK.2010.RLTS.T42062A10617695.en accessed on March 15, 2019.

Ortigosa, G. R., De Leo, G. A., & Gatto, M. (2000). VVF: integrating modelling and GIS in a software tool for habitat suitability assessment. *Environmental Modelling & Software*, *15*(1), 1-12.

Osborne, P. E., Alonso, J. C., & Bryant, R. G. (2001). Modelling landscape scale habitat use using GIS and remote sensing: a case study with great bustards. *Journal of applied ecology*, *38*(2), 458-471.

Pant, D. D., & Osborne, R. (2002). *Introduction to gymnosperms, Cycas, and Cycadales*. Birbal Sahni Institute of Palaeobotany.

Pariti, B. & Borah, A. (2007). *Wild edible plants of Assam*. Assam: Director Forest Communication, Forest Department.

Osborne, P. E., Alonso, J. C., & Bryant, R. G. (2001). Modelling landscape scale habitat use using GIS and remote sensing: a case study with great bustards. *Journal of applied ecology*, *38*(2), 458-471.

Pant, D. D., & Osborne, R. (2002). *Introduction to gymnosperms, Cycas, and Cycadales*. Birbal Sahni Institute of Palaeobotany.

President Chure-Tarai Madhesh Conservation Development Board. (2014). Chure Conservation Area (An atlas of 36 districts).

Questad, E. J., Kellner, J. R., Kinney, K., Cordell, S., Asner, G. P., Thaxton, J., & Evans, S. A. (2014). Mapping habitat suitability for at risk plant species and its implications for restoration and reintroduction. *Ecological Applications*, *24*(2), 385-395.

Singh, K. J., & Singh, R. (2014). Population assessment and distribution of *Cycaspectinata* Buchanan-Hamilton in Northeast India. *Pleione*, *8*(1), 17-25.

Singh, R., & Singh, K. J. (2010). Status and survey of *Cycaspectinata* Buchanon-Hamilton in Sikkim and West Bengal. *Pleione*, *4*(2), 235-239.

Wintle, B. A., Elith, J., & Potts, J. M. (2005). Fauna habitat modelling and mapping: a review and case study in the Lower Hunter Central Coast region of NSW. *Austral Ecology*, *30*(7), 719-738.

Wu, X. B., & Smeins, F. E. (2000). Multiple-scale habitat modeling approach for rare plant conservation. *Landscape and Urban Planning*, *51*(1), 11-28.

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Population Structure of *Juniperus indica* Bertol. along Elevation Gradient in Manang, Trans-Himalayas Nepal

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Abstract

Elevation gradients are complex involving different co-varying factors that influence plant population structure. Along the elevation gradient, subalpine forests are highly vulnerable to natural variation in climate as well as are also under high anthropogenic pressure. The present study aims to study habitat characteristics and regeneration status based on population structure of *Juniperus indica*, a common and highly useful species, along elevation gradient in Manang, north-central Nepal. The distribution range was divided into lower- (3350-3580 m), mid- (3650-3880 m) and higher- (3950-4250 m) elevation bands, where populations were sampled in a total of 54 plots (18 plots per band) of 10 m x 10 m size. In each plot, we recorded aspect, slope and associated species, and number of individuals of studied species classified into seedling, juvenile and adult; and trunk diameter of adult individuals. Altogether, 88 plant species were identified. Plots in the lower- and mid-elevation bands mostly comprised woody shrubs, whereas herbaceous species dominated the higher-elevation band. Mid-elevation band tended to show highest density of seedlings and juveniles, but adult density was high in the plots at lower-elevation band. *J. indica* exhibited almost similar population structure in three bands, with high contribution of juveniles than seedlings and adults. Density-diameter curve was reverse J-shaped, indicating continuous regeneration. Population density is influenced differently by the variations in elevation. *J. indica* in Manang exhibits successful regeneration despite harsh ecological conditions and anthropogenic pressure.

Keywords: Density-diagram curve, Habitat characteristics, Population density, Regeneration

Introduction

Elevation gradients are complex involving many different co-varying factors like topography, soil and climate (Austin et al., 1996). Elevation strongly influences length of growing season and the availability of soil moisture and nutrients (Namgail et al., 2012). Plant species growing along the elevation gradient show considerable variations in structure of their populations and in traits related to their life history and growth. Population structure reflects biological and ecological characteristics of plants which are used to determine regeneration profile (Teketay, 1996). Continuous regeneration is necessary for the long-term persistence of a species population (Thakuri, 2010). Population density of seedlings and juveniles are considered as regeneration potential of a species (Bharali et al., 2012). Abundance of established seedlings and juveniles affects the future composition of forests (Thakuri, 2010). The inclusion of seedlings and juveniles in plant population structures would provide better information about the status of the species at early stage of regeneration. Germination of seeds and establishment of seedlings and juveniles are related to the availability of space and moisture conditions and to adaptation to particular light regimes (Ramakrishnan et al., 1982).

Population dynamics of plant species can be described by demographic properties such as recruitment, mortality and growth. The balance among these properties regulates the dynamics and the structure of a population (Bharali et al., 2012). Plants generally grow and survive in a limited range of the environmental conditions, for example, temperature and light availability and variation in these factors play important roles in shaping the age/size structure and regeneration at different elevations (Block & Treter, 2001; Duan et al., 2009).

Subalpine forest represents the uppermost forest ecosystems along the elevation gradient. They are highly vulnerable to natural variation in climate (Sano et al., 2005) and are also under high anthropogenic pressure (Sharma et al., 2009). Ecological study of subalpine forests in the Nepal Himalaya is very scanty, though some initiatives have been taken in recent time (e.g., Ghimire & Lekhak, 2007; Shrestha et al., 2007; Ghimire et al., 2008; Suwal, 2010; Gaire et al., 2013).

Juniperus indica is an important component of sub-alpine forest of Manang district, northcentral Nepal (Ghimire et al., 2008). The local community and traditional Tibetan practitioner utilize maximum parts of *Juniperus* species, for example, fruits, leaves, stem and barks in traditional medicine to cure kidney, skin and lymph disorders, fever, cough and cold, sores, wounds, and paralysis of limbs (Bhattarai et al., 2006; Ghimire et al., 2008); leaves to burnt for incense by Buddhists. The plant is also used for fencing purpose (Bhattarai et al., 2006). Dried leaves are sold for incense locally. Essential oil obtained from steam distillation of fresh leaves is exported for its use in medicines and cosmetics (Ghimire et al., 2008). Leaves are harvested throughout the year while fruits during July to August.

Studies on Himalayan junipers are confined to essential oil variation in leaf (e.g., Adams & Chaudhary, 1996; Adams et al., 1998), taxonomic determination (e.g., Adams et al., 2009), ethnobotany (e.g., Bhattarai et al., 2006), vegetation analysis (Ghimire et al., 2008), and variation in leaf biomass and fruit outputs (Chapagain et al., 2017). But there are inadequate studies on habitat characteristics, population structure and regeneration potential of *J. indica* which are on high anthropogenic pressure (e.g., destructive practices, such as over-harvesting of leaves for incense and slash-burning to harvest its wood) as well as harsh climatic conditions.

The present study aims to explore habitat characteristics of *J. indica* and predict its regeneration status based on population structure of *J. indica* along elevation gradient in Manang, north-central Nepal, to understand the influences of environmental factors on forest regeneration (Wang et al., 2004). For this, we sampled *J. indica* along an elevation gradient and studied variation in its population ecology. First, the habitat characteristics conditioning differences in plant composition along the elevation gradient were analyzed. Second, variation in population structure and regeneration potential were assessed among populations distributed along the elevation gradient.

Materials and Methods

Study species

Study species *J. indica* is native to high-altitude Himalaya, ranging from Kashmir, India to western Yunnan, China. It is abundant throughout Nepal ranging from 3300 m to 4500 m asl (Press et al., 2000). The plant originates on open and rocky alpine slopes in drier areas. The plant occurs as dwarf woody-shrub at higher elevations above 4,200 m asl and as trees growing at elevational range of 3300-4000 m asl (Ghimire et al., 2008). The leaves are dark grey-green, dimorphic. Mature plants have mostly scale-like leaves which are decussate or sometimes in whorls of 3, closely appressed, 1-3 mm long; while young plants have mostly needle-like leaves, which are borne in whorls of 3 and are 5-8 mm long. Needle-like leaves are also found on shaded shoots of matured plants. The plant is dioecious with male (pollen) and female (seed) cones on individual plants. The pollen cones are sub-globose or ovoid, 2-3 mm long; seed cones are ovoid, berry-like, 6-10 mm long, glossy black when ripe, and contain a single seed. The cones are seen in April to May that mature in October to December. The seeds are mostly dispersed by birds after eating the cones (Ghimire et al., 2008; Chapagain et al., 2017).

Study area

The study area is in Manang District in the north-central part of Nepal (latitude 28.650715 to 28. 675786°N and longitude 84.050218 to 84.056930°E) within altitude range of 3350 to 4250 m asl (Figure 1). The northern part of Manang Valley receives very low annual monsoonal precipitation of ca. 450 mm, whereas the precipitation at southern region (Chame, Manang, at 2680 m asl) remains >1,000 mm (Miehe et al., 2001; Baniya et al., 2009). Similarly, the average annual temperature rests around 6.2°C in the northern trans-Himalayan valley and 11.0°C in the southern region in Manang. There is decreasing moisture from east to west in the upper Manang Valley, and the south-facing slopes are much drier than those facing north (Bhattarai et al, 2004; Ghimire et al., 2008).

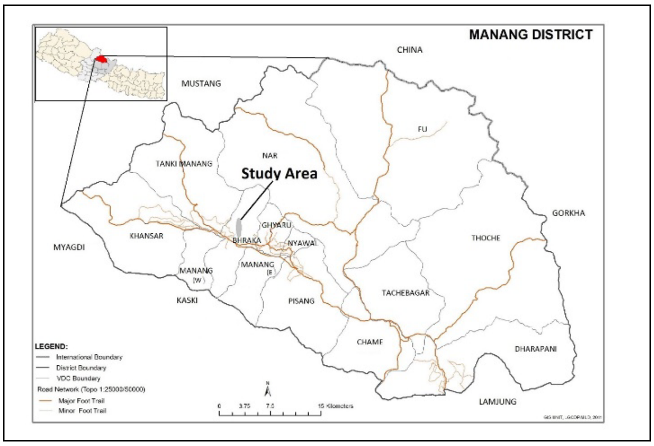

Figure 1. Map of the study area.

Sampling methods

Sampling of *Juniperus indica* population was made in September 2011 in north-eastern part of Manang Valley, using a systematic sampling approach (Chapagain et al., 2017). The study was conducted from Bhraka Village (3350 m asl) of Manang to Ice Lake (4250 m asl) classifying the entire of the distribution range into three elevation bands, based on the harvesting level by local community, lower-elevation band being the nearest and maximum harvest to higher-elevation band being the farthest and least harvest. Lower-elevation band ranged from 3350-3580 m asl, mid-elevation band ranged from 3650-3880 m asl and higher-elevation band ranged from 3950-4250 m asl. In each elevation band, 3 horizontal transects were laid down at *ca*. 75-100 m elevation intervals. In each transect, 6 plots of size 10 m× 10 m were randomly sampled at *ca*. 50-100 m length intervals to record the *J. indica* populations, resulting 54 plots in total. In each sampling plot, individuals of *J. indica* of different size classes were manually counted and recorded separately (Chapagain et al, 2017).

Size classes were recognized related to the growth stage following Chapagain et al. (2017) that was developed from Schemske et al. (1994). The size classes of individuals of *J. indica* were broadly defined according to plant height and/or trunk diameter. Plant height less than 0.1 m and/or trunk diameter less than 1 cm were classified as seedlings. Plant height ranging from 0.1 m to 1.0 m and/or trunk diameter less than 1 cm were classified as juveniles and plant height usually more than 1 m and/or trunk diameter more than 1 cm and also bearing reproductive structure were classified as adult. Adult individuals were recorded for their trunk diameter by using measuring tape. Trunk diameter of adult individuals more than 1-3 m tall was recorded at 25 cm aboveground, while the trunk diameter of adult individuals more than 3 m tall was recorded at 137 cm aboveground.

Latitude, longitude and altitude of each plot were recorded with the help of GPS device. Aspect and slope of each plot were recorded using compass and clinometer respectively. Each 100 m^2 plot was further divided into 4 subplots of 5 m × 5 m size. In each subplot, presence/absence of plant species associated with *J. indica* was recorded. Vouchers of plant species encountered in sampling plots were collected. The vouchers were identified using taxonomic reference (e.g., Polunin & Stainton, 1984; Stainton, 1988) and comparing with specimens housed at Tribhuvan University Central Herbarium (TUCH) and National Herbarium and Plant Laboratories (KATH), Nepal. Herbarium specimens were deposited in TUCH. Nomenclature follows Press et al. (2000).

Data analysis

Habitat characteristics of *J. indica* were evaluated in terms of variation in physical/ topographic variables, and by analyzing patterns of associated species diversity and composition along the elevation gradient. The value of aspect, slope and latitude were combined to calculate potential annual direct incident radiation (PADIR, MJ cm–2 yr–1) by using the formula given by McCune & Keon (2002). Aspect has been folded about the north-south line (rescaling 0-3600 to 0-1800, such that NE = NW, E = W, etc) before calculating PADIR using the following formula: folded aspect = 180 – |Aspect – 180| (McCune & Keon 2002). The formula for calculating PADIR is given by: PADIR = –1.467+1.582×COS(G3) × COS(H3)–1.5×COS(I3) ×SIN(H3) × SIN(G3) - 0.262×SIN(G3) ×SIN(H3) + 0.607×SIN(I3) × SIN(H3); where latitude, slope, and folded aspect are in columns G, H, and I, respectively, all in radians in Microsoft Excel. It gives a relative value (ranging from 0.03-1.11) of how much solar radiation a particular spot receives. Bio-physical variables recorded in *J. indica* growing sites in three elevation bands were compared using one-way ANOVA when the data met assumptions of parametric test (i.e. normal distribution and homogeneity of variance). Bio-physical variables that did not meet the assumption of parametric test even after transformation were treated with non-parametric Kruskal-Wallis tests.

Richness of associated species was calculated as the total number of such species present per plot. Presence-absence data from all the four subplots were combined to calculate abundance of each associated species per plot, in an ordinal scale from 0 (absent from all four subplots) to 4 (presence in all subplots). The abundance data of 88 associate species including *J. indica* from all 54 plots were used to calculate their frequency in lower-, mid- and higher-elevation bands, and overall frequency.

Density and population structure (the relative proportions of seedling, juvenile and adult to total density) of *J. indica* were analyzed for each plot and each elevation band (Bharali et al., 2012). Regeneration potential of *J. indica* was evaluated based on the density of seedlings and juveniles (Shankar, 2001). It was evaluated as the sum of seedling density and juvenile density divided by the density of adults (Endels et al., 2004). In addition, density-diameter (d-d) curve was also developed for individuals with trunk diameter >1 cm to further assess regeneration patterns and population structure of adults. Variation in population density among elevation bands was compared using one-way ANOVA. Liner mixed model (LMM) (McCulloch and Searle, 2000; Verbeke & Molenberghs, 2000) was used to study the effects of elevation on population structure (proportion of life stages - seedlings, juveniles and adults) and regeneration potential. Elevation band and transect nested within elevation band were used as fixed factor and study plot was used as random variable in the model. PADIR and aspect (folded about the north-south line) were used separately in the model as cofactor to account for the effect of insolation and heat load (McCune & Keon, 2002) respectively. LMM procedure fits models a lot of general than those of the generalized linear model (GLM) procedure. LLM also encompasses all models within the variance elements procedure. The major capabilities that differentiate LMM from GLM are that LMM handles correlated and unbalanced data and unequal variances (McCulloch & Searle, 2000). LMM also handles more complex situations in which experimental units are nested in a hierarchy. LMM analyzed the proportions of population structure and regeneration pattern of *J. indica* and also gave significant results for adult proportion and rejuvenation when transects were nested within elevation band. The density-diameter (d-d) curve for adult *J. indica* was analyzed from all study plots to observe the regeneration pattern.

Results and Discussion

Habitat characteristics of J. indica

Juniperus indica was recorded from open dry, rocky and sandy habitats in SE- to SW-facing slopes in upper Manang Valley, with main vegetation type being *Rosa-Berberis-Juniperus* shrubland, subalpine and alpine grasslands, and open forests (mainly of *Pinus wallichiana* at lower elevation), similar to Junicost (2010) and Ueckert (2013), as *Juniperus* mostly prefer limestone and found on dry rocky habitats. The plots at three elevation bands differed in all biophysical variables studied (Table 1). *J. indica* occurred on gentle SW-facing slopes towards higher elevations receiving high solar radiation (Table 1), similar to study of Bhattarai et al. (2006) and Rawat & Everson (2012). Being the root system highly developed and xerophytic nature, the plant has an advantage to establish in dry and rocky substrates. The species is therefore proposed to be particularly suited for afforestation program under xeric ecological conditions of trans-Himalaya (Rawat & Everson, 2012).

Altogether, 88 vascular plant species (representing 64 genera and 37 families), associated with *J. indica*, were identified from the study area, 33 species in lower-elevation band, 49 in mid-elevation band and 72 in higher-elevation band (Table 4). Asteraceae was the dominant family in the study area comprising 16 species within 10 genera, followed by Rosaceae, Gentianaceae, Ranunculaceae, Fabaceae, Scrophulariaceae and Cyperaceae. *Anaphalis*, *Artemisia*, *Carex*, *Juniperus* and *Pedicularis* were the largest genera, each comprising 3 species (Table 5). Subedi (2016) reported 94 species (37 families and 63 genera) associated with population of *J. squamata* in the same study area. Ghimire et al. (2008) reported 19 associated species in *J. indica* forest, representing 14 genera and 11 families within the similar elevation range from 3300 to 4000 m asl. Richness and abundance of associate species were significantly high at higher-elevation plots, the values of which decreased linearly towards lower elevation (Table 1). Vegetation within a landscape is greatly affected by differences in the microclimate, aspect and altitude. The main source of geographic variation in the plant species composition of juniper communities was due to climate and soil texture (Reinoso et al., 2003) and aspect (Bennie et al., 2006).

Frequency of *J. indica* occurrence was estimated to be 68.06%, 61.11%, and 47.22% in lower-, mid- and higher-elevation bands, respectively with an overall frequency of 58.80%. Among the species associated from all sampling plots, *Tanacetum dolichophyllum* (54.17%), *Juniperus squamata* (43.06%), *Rosa sericea* (41.20%), *Berberis aristata* (38.43%), *Carex species* (37.50%), *Tanacetum* sp. (33.80%) and *Cotoneaster microphyllus* (33.33%) exhibited overall high frequency of occurrence (Table 4). The three elevation bands differed, to some extent, in composition of vascular plant species. Plots in the lower- and mid-elevation bands mostly comprised woody shrubs, whereas herbaceous species dominated the plots in the higher-elevation bands. *Carex* spp., *Rosa sericea*, *Tanacetum dolichophyllum*, *Berberis aristata* and *Rhododendron anthopogon* were the most frequent species (with frequency of occurrence >30%) in lower-elevation band. Similarly, *Rosa sericea*, *Cotoneaster microphyllus*, *Tanacetum dolichophyllum*, *Juniperus squamata*, *Berberis aristata*, *Potentilla fructicosa* and *Bistorta macrophylla* were the most frequent species (with frequency >30%) in mid-elevation band, and *Tanacetum dolichophyllum*, *Juniperus squamata*, *Tanacetum* sp., *Lonicera hypoleuca*, *Potentilla peduncularis*, *Rhododendron lepidotum*, *Conyza* sp., *Spiraea canescens*, *Berberis aristata*, *Carex* spp., *Delphinium brunonianum*, *Ajuga lupulina* and *Kobresia gammiei* were the most frequent species (with frequency >30%) in higher-elevation band (Table 4)

Density and size distribution of J. indica

Density of seedling, juvenile and adult of *J. indica* in the entire study area were found to be 4.89 ± 0.67, 6.59 ± 0.95 and 2.26 ± 0.28 (mean±SE) individuals per 100 m^2 plot. The overall density, combining all three size classes was 13.74±1.47 individuals per

Table 1: Bio-physical variables (mean ± SE) recorded in *J. indica* growing sites in three elevation bands (low, mid, high) in Ice Lake area, upper Manang. For elevation, range values are given in the parentheses.

Variables	Low	Mid	High	p
Elevation (m)	3472.83 ± 20.11 (3351-3585)	3761.56 ± 18.70 (3655-3885)	4064.50 ± 20.72 (3947-4197)	<0.001
Aspect (0)	114.06 ± 10.34	136.39 ± 8.52	98.00 ± 8.89	0.021
Slope (0)	46.67 ± 3.96	24.72 ± 2.96	20.28 ± 3.17	<0.001
PADIR (MJ cm–2 yr–1)	0.84 ± 0.04	1.03 ± 0.03	1.00 ± 0.02	0.001
Associate species richness*	23.94 ± 1.54	33.00 ± 2.63	60.44 ± 2.88	<0.001
Associate species abundance	11.56 ± 0.48	15.78 ± 1.42	27.39 ± 1.32	<0.001

Note: Number of vascular plant species associated with *J. indica* per plot. *p* values based on Kruskal-Wallis tests or on one-way ANOVA.

100 m^2 plot or 1374 individuals per ha. Mid-elevation band tended to show high density of seedlings and juveniles, but the results were statistically insignificant (Table 2). On contrary, adult density was high in plots at lower-elevation band.

The total density in the present study is almost three times higher than the value (404 individuals per ha) obtained by Ghimire et al. (2008) in Manang, but almost three times less than the value obtained by Chhetri & Gupta (2007) in Mustang (4250 individuals per ha), Rai (2013) in Langtang region (3500 individuals per ha) and Subedi (2016) in the same study area for *J. squamata* (4850 individuals per ha). However, the elevation trend of population density was almost identical with that of previous reports. In the present study, total density and density of seedlings and juveniles of *J. indica* were found to be high at mid elevation (3650-3880 m asl), but the density of adult trees was high at lower elevation (3350-3580 m). Ghimire et al. (2008) also recorded the total density of *J. indica* to be highest at 3500-3800 m asl (516.66 individuals per ha), followed by 3800-4000 m asl (375 individuals per ha) and 3300-3500 m asl (320 individuals per ha). Subedi (2016) observed density in low, mid and high elevation bands to be 700, 2,422 and 1,728 individuals per hectare respectively in the same study area for *J. squamata*. The variation in population size of seedlings, juveniles and adults at different elevations may be the results of climatic (especially rainfall and temperature) and edaphic factors (availability of soil water) that are critically important for the successful recruitment, establishment, survival and reproduction of plants (Bharali et al., 2012). There is sharp decline in temperature with the rise of elevation (temperature lapse rate for the western Himalayas is estimated to be 0.6-0.74°C per 100 m elevation raise for various months of the year (Jain et al., 2008). The pattern of decrease in density with elevation can vary with species as biotic interactions, mainly competition, also play a role in growth rate (McPherson & Wright, 1989; Ghimire et al., 2010).

The proportions of seedling, juvenile and adult of *J. indica* in all 54 plots were 0.356, 0.480 and 0.164 respectively. In three elevation bands, *J. indica* exhibited almost similar population structure, with high contribution of juveniles than seedlings and adults (Figure 2). However, proportion of adult was significantly high in lower-elevation band (LMM $F2, 44$ = 3.425, p = 0.041, Figure 2, Table 3), whereas proportions of seedling and juvenile tended to be high in mid- and higher-elevation bands (but the results were statistically insignificant, Figure 2 and Table 3). But significantly higher value of rejuvenation (expressed as the sum of seedling and juvenile density divided by the density of the adults) at mid- and higher-elevation bands compared to lower-elevation (LMM $F2, 44$ = 3.280, p = 0.047, Figure 2, Table 3) signifies potentially higher regeneration of *J. indica* towards mid- and higher elevations.

LMM analyses also gave significant results for adult proportion and rejuvenation when transects were nested within elevation band (Table 3), indicating that these parameters spatially varied within the same elevation band. However, in either of the case, the effect of aspect or PADIR (both used separately as covariate in the model) were not significant, indicating SE and SW gradient or incident radiation did not affect population structure. Among the remaining predictor variables considered in this study, richness and abundance of associate species also did not show significant relationship with either

Table 2: Density (number of individuals per 10 × 10 m plot) of *J. indica* recorded in three elevation bands in Ice Lake area, upper Manang valley. Data shown are mean ± SE.

	Low	Mid	High	F2,53	p
Seedling	5.39 ± 1.35	6.22 ± 1.39	3.06 ± 0.39	1.484	0.236
Juvenile	7.06 ± 1.63	8.39 ± 2.20	4.33 ± 0.64	0.582	0.563
Adult	3.17 ± 0.52	1.50 ± 0.27	2.11 ± 0.57	2.536	0.089
Total	15.61 ± 2.32	16.11 ± 3.45	9.50 ± 1.15	1.894	0.161

Note: *F* and *p* values based on one-way ANOVA.

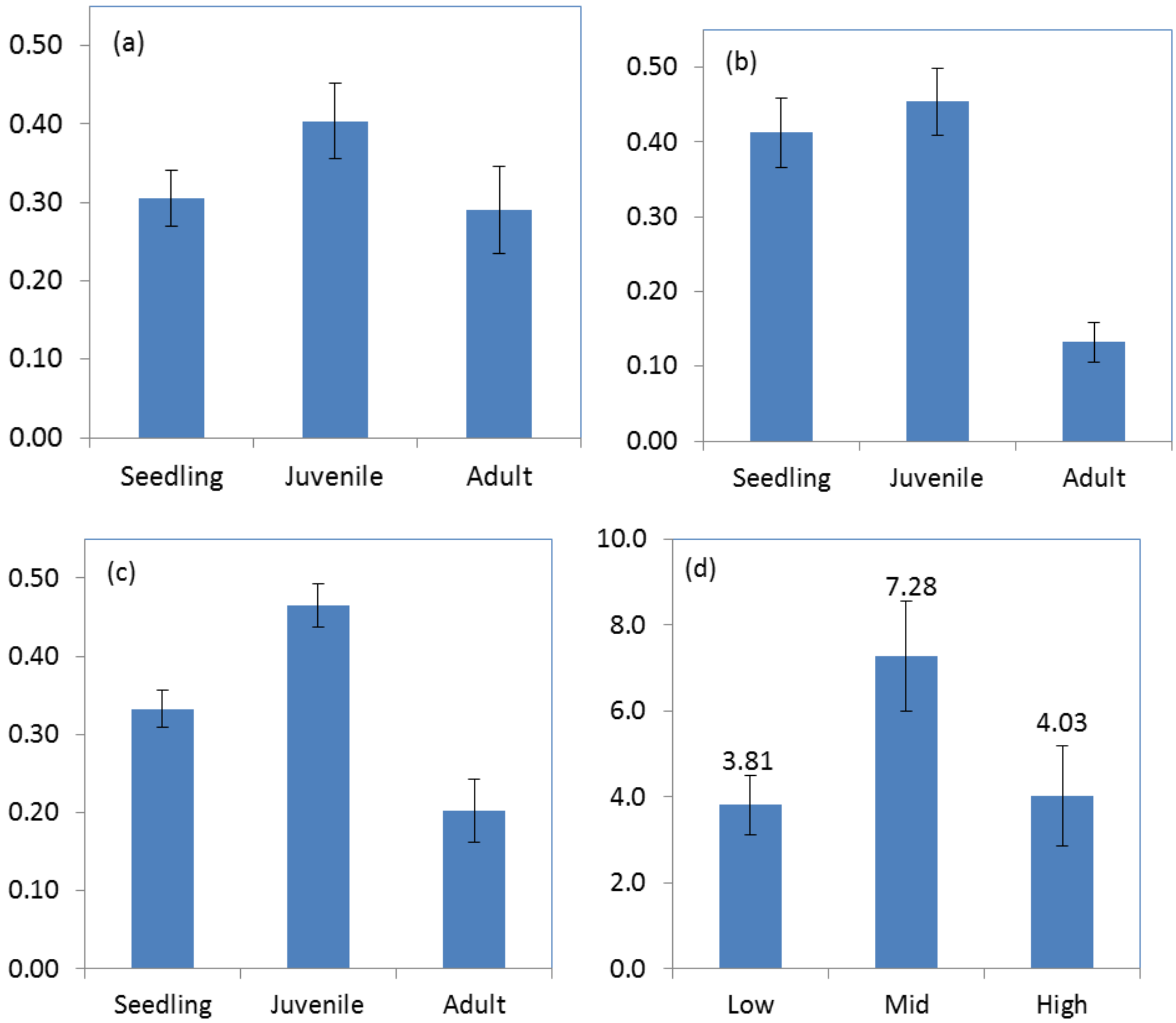

Figure 2: Proportions of Population structure [proportions (mean ± SE) of seedling, juvenile and adult] and regeneration pattern of *J. indica* at three elevation bands in Ice Lake area, upper Manang valley: (a-c) Population structure at three elevation bands (a – lower-elevation, b – mid-elevation, and c – higher-elevation), (d) rejuvenation expressed as the sum of seedling and juvenile density divided by the density of adults.

Table 3: Results of liner mixed model (LMM) showing the effects of elevation band and transect (nested within elevation band) as fixed factor and aspect (folded about the north-south line) as cofactor on size class proportions and rejuvenation of *J. indica*. Position of plots was used as random variable in the LMM.

	Seedling proportion				Juvenile proportion			
Source of variation	Ndf	Ddf	F	P	Ndf	Ddf	F	P
Elevation band	2	44	1.423	0.252	2	44	0.632	0.536
Transect (elevation band)	6	44	0.711	0.643	6	44	0.792	0.581
Aspect	1	44	1.526	0.223	1	44	0.297	0.589
	Adult proportion				Rejuvination			
Source of variation	Ndf	Ddf	F	P	Ndf	Ddf	F	P
Elevation band	2	44	3.425	0.041	2	44	3.280	0.047
Transect (elevation band)	6	44	2.678	0.027	6	44	5.037	0.001
Aspect	1	44	3.290	0.077	1	44	.471	0.496

Note: Ndf = numerator df; Ddf = denominator df density of *J. indica* in three size classes or their proportions.

The future community structure and regeneration status of plant species can be predicted from the relative proportion of seedlings, juveniles and adults in the total population (Bharali et al., 2012). The highest contribution of juveniles may be the result of maximum seedlings grown to juveniles. Seedlings and juveniles together constituted about 84.35% of the total population of *J. indica*. This shows good regeneration potential of *J. indica* in the study area.

Some of the previous studies have reported that regeneration potential of *Juniperus* species is generally low (Juan et al., 2003; Otto et al., 2010). The factors considered for explaining low regeneration in *Juniperus* spp. are low production of viable seeds (Juan et al., 2003; Junicost, 2010; Otto et al., 2010), disturbance, increased competition, and absence of suitable dispersal vectors (McPherson & Wright, 1989), and in some dioecious species, low amount of pollen that reaches female individuals resulting in less number of fruits set (Juan et al., 2003). In contrast, our study indicates successful regeneration of *J. indica* in Manang, central Nepal despite harsh ecological conditions. *J. indica* exhibits dioecious or monoecious habit (Adams, 2014). Even in dioecious form, the distribution of male and female adults in most populations was random with no evidence of sex clustering resulting in high reproductive success by high amount of pollen reaching female individuals to produce high number of fruits set and this might be the reason why *J. indica* showed high regeneration potentiality in Manang.

However, regeneration of *J. indica* was comparatively low in lower elevation band, despite higher seed output than in mid and higher elevations. This might be due to several reasons. Firstly, this can be linked to disturbance, as the lower elevation band of the study area is close to settlement area where it received greater human pressure especially from livestock grazing. Secondly, *J. indica* in lower elevation band is associated with *Pinus wallichiana* where dominance of pine needles deposition of later species might have suppressed recruitment and successful establishment of *Juniper* seedlings. Reinoso et al. (2003), for example, observed that maritime juniper woodlands of Spain were affected by pine plantations, where deposition of pine needles reduced recruitment of juniper seedlings and increased their mortality. Junicost (2010) reported that as pines are growing much faster than junipers, they produce massive numbers of easily germinable seeds and modify the microenvironment against other species by deposition of pine needles. Junipers have low germination capacity [e.g., <5% in maritime juniper (Reinoso et al., 2003)] are difficult to propagate from seed and are slow growing which, when coupled with human disturbance and competitive stresses, can make establishment difficult (Forestry commission, 2003).

Increasing proportions of younger individuals of *J. indica* in mid and high elevation band show its capacity to tolerate harsh ecological conditions of higher elevations. Wide elevation amplitude (3300-4500 m within Nepal) together with its capacity to tolerate harsh conditions may enable *J. indica* to be quite successful species to adapt ecological changes. There is evidence that increasing carbon dioxide concentrations in the atmosphere during the last century may be benefitting junipers as they utilize the elevated carbon dioxide concentrations in C3 photosynthetic pathway increasing their distribution and abundance (Mayeux et al., 1991).

Density-diameter (d-d) curve

The density-diameter (d-d) curve for adult *J. indica* from all study plots was nearly reverse *J*-shaped (Figure 3a), where the density of *J. indica* was successively reduced with the increase of trunk diameter. In the lower-elevation band, smaller- and large-sized adult individuals were more or less equally present except for size class >4-6, which had highest density value (Figure 3b); whereas in the mid- and higher-elevation bands, the large-sized adults(>8 cm in mid- and >6 cm in higher-elevation band) were completely absent (Figure 3c,d).

The nearly reverse J-shaped d-d curve for *J. indica*, indicates its continuous regeneration (Vetaas, 2000). Similar reverse J-shaped patterns of population

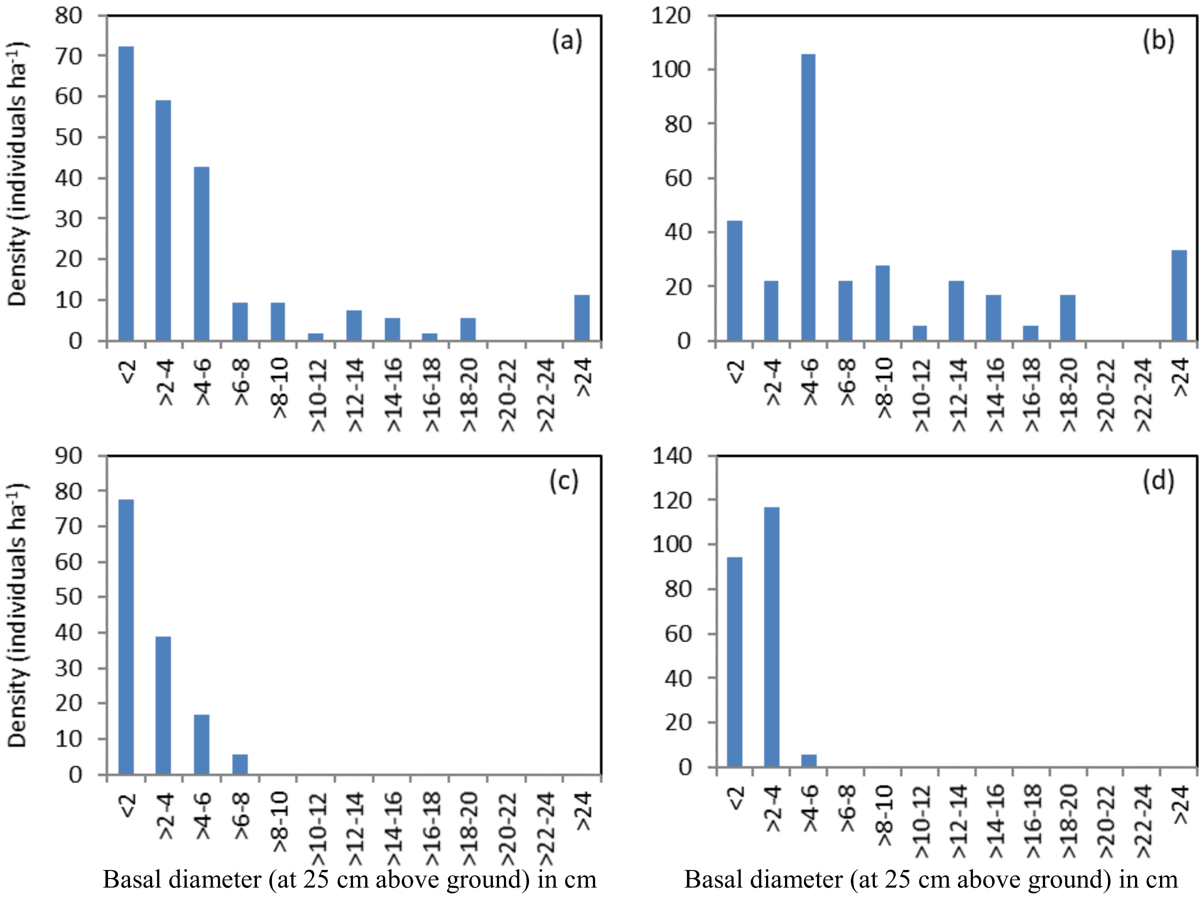

Figure 3: Density-diameter curve for adult *J. indica* in (a) the overall study plots, (b) lower-elevation band, (c) mid-elevation band, and (d) higher-elevation band. Basal diameter (at 25 cm above ground) in cm Basal diameter (at 25 cm above ground) in cm.

structure have been reported for different high-altitude tree species of Manang, central Nepal, such as *Abies spectabilis* (Ghimire & Lekhak 2007), *Betula utilis* (Shrestha et al., 2007), and *Pinus wallichiana* (Ghimire et al., 2010).

Conclusion

Juniperus indica preferred dry and rocky habitats in SE- to SW-facing slopes, along with *Rosa-Berberis-Juniper* shrubland, subalpine and alpine grasslands, and open forests. Above 3800 m on the southern aspect, the forest was comprised of only bushy *J. indica*. Altogether, 88 plant species, associated with *J. indica*, were identified. Lower- and mid-elevation bands mostly comprised woody shrubs, whereas herbaceous species dominated higher-elevation band. Population density of *J. indica* is influenced differently by the variations in elevation. Mid-elevation band tended to show highest density of seedlings and juveniles, but adult density was high in the plots at lower-elevation band. *J. indica* exhibited almost similar population structure in three bands, with high contribution of juveniles than seedlings and adults. However, proportion of adult was high in lower-elevation, whereas proportions of seedling and juvenile tended to be high in mid- and higher-elevations. Density-diameter curve for adult *J. indica* was reverse *J*-shaped, indicating continuous regeneration.

It is concluded that *J. indica* in Manang exhibits higher regeneration at mid- and higher elevation bands indicating plants ability to tolerate adverse environmental conditions as well as a tendency for expansion of its distribution niche towards cooler habitat of high elevation despite high anthropogenic pressure.

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Conflict of Interest

There is no any conflict of interest among the authors.

References

Adams, R. P. & Chaudhary, R. P. (1996). Leaf essential oil of *Juniperus indica* Bertol from Nepal. *Journal of Essential Oil Research*, *8,* 677–680.

Adams, R. P., Thapa, R. K., Agrawal, S. G., Kapahi, B. K., Srivastava, T. N. & Chaudhary, R. P. (1998). The leaf essential oil of *Juniperus recurva* Buch. Ham. ex D. Don from India and Nepal compared with *J. recurva* var. *squamata* (D. Don) Parl. *Journal of Essential Oil Research*, *10*, 21–24.

Adams, R.P. (2014). *Junipers of the World: The Genus Juniperus*. Fourth Edition. Bloomington, USA: Trafford Publishing Co.

Adams, R.P., Chaudhary, R.P, Pandey, R.N. & Singh, L. (2009). *Juniperus recurva* var. *uncinata*, the hooked branchlet juniper, a new variety from Nepal. *Phytologia*, *91*(3), 361-366.

Austin, M. P., Pauses, J. G. & Nicholls, A. O. (1996). Pattern of tree species richness in relation to environment in south-eastern New South Wales Australia. *Australian Journal of Ecology, 21*, 154-164.

Baniya, C.B., Solhøy, T. & Vetaas, O.R. (2009). Temporal changes in species diversity and composition in abandoned fields in a trans-Himalayan landscape, Nepal. *Plant Ecology, 201*, 383-399.

Bennie, J., Hill, M.O., Baxter, R. & Huntley, B. (2006). Influence of slope and aspect on long-term vegetationchange in British chalk grasslands. *Journal of Ecology, 94,* 355-368.

Bharali, S., Paul, A. Khan, M.L. & Singha, L.B. (2012). Impact of altitude on population structure and regeneration status of two *Rhododendron* species in a temperate broad leaved forest of Arunachal Pradesh, India. *International Journal of Ecosystem, 2* (1), 19-27.

Bhattarai K.R., Vetaas O.R. & Grytnes J.A. (2004). Relationship between plant species richness and biomass in an arid sub-alpine grassland of the central Himalayas, Nepal. *Folia Geobotanica, 39*, 57-71.

Bhattarai, S., Chaudhary, R.P. & Taylor, R.S.L. (2006). Ethnobotany of wild Junipers (*Juniperus* species) in Manang district, central Nepal. *Scientific World, 4* (4), 109-112.

Block, J. & Treter, U. (2001). The limiting factors at the upper and lower forest limits in themountain-woodland steppe of northwest Mongolia Joachim Block and Uwe Treter. In Kaennel D.M. and Braker O.U. (Eds.), *the International Conference on Tree Rings and People*, Davos. Proceedings.

Chapagain, A., Chaudhary, R.P. & Ghimire, S.K. (2017). Variation in leaf biomass and fruit output of *Juniperus indica* along an elevation gradient in north-central Nepal. *Banko Jankari (A Journal for Forestry Information of Nepal), 27* (1), 3-10. doi: http://dx.doi.org/10.3126/banko.v27i1.18543

Chhetri, H.B. & Gupta, V.N.P. (2007). A survey of non-timber forest products (NTFPS) in upper mustang. *Scientific World, 5* (5), 89-94.

Duan, R.Y., Wang, X.A., Tu, Y.B., Huang, M.Y., Wang, C., Zhu, Z.H. & Guo, H. (2009). Recruitment pattern of tree populations along an altitudinal gradient: *Larix chinensis* in Qinling mountains (China). *Polish Journal of Ecology, 57* (3), 451–459.

Endels, P., Adriaens, D., Verheyen & Herm, M.

(2004). Population structure and adult performance of forest herbs in three contrasting habitats. *Ecography*, *27*, 225-241.

Forestry commission. (2003). Growing Juniper: Propagation and Establishment Practices. Retrieved from http://www.forestry.gov.uk/pdf/FCIN050.pdf/$FILE/FCIN050.pdf. (assessed 11Nov. 2016).

Gaire, N.P., Koirala, M., Bhuju, D.R. & Borgaonkar, H.P. (2013). Treeline dynamics with climate change at Central Nepal Himalaya. *Climate Past Discussion, 9*, 5941–5976.

Ghimire, B.K. & Lekhak, H.D. (2007). Regeneration of *Abies spectabilis* (D. Don) Mirb. in subalpine forest of Upper Manang, north-central Nepal. In (Eds.) R. P. Chaudhary, T. H. Aase, O.R. Veetas and B. P. Subedi, *Local Effects of Global Changes in the Himalayas: Manang, Nepal* (pp. 139-149). Nepal: Tribhuvan University and Norway: University of Bergen.

Ghimire, B.K., Lekhak, H.D., Chaudhary, R.P. & Vetaas, O.R. (2008). Vegetation analysis along an altitudinal gradient of *Juniperus indica* forest in southern Manang Valley, Nepal. *International Journal of Ecology and Development*, *9*, 20-29.

Ghimire, S. K., Sapkota I. B., Oli, B. R. & Parajuli, R. R. (2008). *Non-Timber Forest Products of Nepal Himalaya: Database of Some Important Species found in the Mountain Protected Areas and Surrounding Regions.*, Kathmandu, Nepal: WWF Nepal Program.

Ghimire, B.K., Mainali, K.P., Lekhak, H.D., Chaudhary, R.P. & Ghimeray, A.K. (2010). Regeneration of *Pinus wallichiana* A.B. Jackson in a trans-Himalayan dry valley of north-central Nepal. *Himalayan Journal of Sciences, 6* (8), 19–26.

Jain, S.K., Goswami, A. & Saraf, A.K. (2008). Determination of land surface temperature and its lapse rate in the Satluj River basin using NOAA data. *International Journal of Remote Sensing, 29* (11), 3091–3103.

Juan, R., Pastor J., Fernandez, I. & Diosdado, J.C. (2003). Relationships between cone traits and seed viability in *Juniperus oxycedrus* L. subsp. *macrocarpa* (Sm.) Ball (Cupressaceae). *Acta Biologica Cracoviensia Series Botanica, 45* (2), 69–78.

Junicost. (2010). *Composition and structure of Juniperus macrocarpa subpopulations*. Retrieved from http://www.junicoast.gr/documents/0000/0048/LIFE_07_NAT_GR_000296_Deliverable_A.3rev1.pdf

Mayeux, H.S., Johnson, H.B. & Polley, H.W. (1991). Global change and vegetation dynamics. In L.F. James, J.O. Evans, M.H. Ralphs and R.D. Child (Eds.), *Noxious Range Weeds*. Boulder, Colorado: Westview Press.

McCulloch, C.E. & Searle, S.R. (2000). *Generalized, Linear, and Mixed Models*, USA: John Wiley and Sons.

McCune, B. & Keon, D. (2002). Equations for potential annual direct incident radiation and heat load. *Journal of Vegetation Science, 13*, 603-606.

McPherson, G.R. & Wright, H.A. (1989). Direct effects of competition on individual juniper plants: a field study. *Journal of Applied Ecology, 26*, 979-988.

Miehe, G. Winiger, M., Böhner, J. & Zhang, Y. (2001). The climate diagram map of High Asia: purpose and concepts. *Erdkunde*, *55*, 94–95.

Namgail, T., Rawat, G.S., Mishra, C., Wieren, S.E. & Herbert, H.T.P. (2012). Biomass and diversity of dry alpine plant communities along altitudinal gradients in the Himalayas. *Journal of Plant Research*, *125*, 93–101.

Otto, R., Krüsi, B.O., Delgado, J.D., Fernández-Palacios, J. M., García-Del-Rey, E. & Arévalo. J.R. (2010). Regeneration niche of the Canarian juniper: the role of adults, shrubs and environmental conditions. *Annals of Forest Science*, *67*(7), 709.

Polunin, O. & Stainton, A. (1984). *Flowers of the Himalayas*. New Delhi, India: Oxford Press.

Press, J.R., Shrestha, K.K. & Sutton, D.A. (2000). *Annotated Checklist of the Flowering Plants of*

Nepal. London: Natural History Museum.

Rai, R. (2013). *Documentation, Promotion and Awareness of Vital Medicinal and Aromatic Plant Species (MAPs) of Langtang Region, Nepal*. The Rufford Small Grants Foundation. Retrieved from http://www.rufford.org/files/10943-1%20Final%20Report.doc

Ramakrishnan P.S., Sukla P.R. and Boojh R. (1982). Growth strategies of trees and their application to forest management. *Current Science, 51*, 448-455.

Rawat, Y.S. & Everson, C.S. (2012). Ecological status and uses of juniper species in the cold desert environment of the Lahaul valley, North-western Himalaya, India. *Journal of Mountain Science, 9*(5), 676-686.

Reinoso, M. & Carlos, J. (2003). *Juniperus oxycedrus* ssp. *macrocarpa* in SW Spain: ecology and conservation problems. *Journal of Coastal Conservation, 9*, 113-122.

Sano, M., Furuta, F., Kobayashi, O. & Sweda, T. (2005). Temperature variations since the mid-18th century for western Nepal, as reconstructed from tree-ring width and density of *Abies spectabilis*. *Dendrochronologia, 23*, 83-92.

Schemske, D.W., Husband, B.C., Ruckelshaus, M.H., Goodwillie, C., Parker, I.M. and Bishop, J.G. (1994). Evaluating approaches to the conservation of rare and endangered plants. *Ecology, 75*, 584-606.

Shankar, U. (2001). A case study of high tree diversity in a sal (*Shorea robusta*) dominated lowland forest of Eastern Himalaya: floristic composition, regeneration and conservation. *Current Science, 81*(7), 776–786.

Sharma, C.M., Suyal, S., Gairola, S. & Ghildiyal, S.K. (2009). Species richness and diversity along an altitudinal gradient in moist temperature forest of Garhwal Himalaya. *Journal of American Science, 5*, 119-128.

Shrestha, B.B., Ghimire, B.K., Lekhak, H.D. & Jha, P.K. (2007). Regeneration of Treeline birch (*Betula utilis* D.Don) forest in a trans-Himalayan dry valley in Central Nepal. *Mountain Research and Development, 27*, 259-267.

Stainton, A. (1988). *Flowers of the Himalayas – a Supplement*. New Delhi, India: Oxford press.

Subedi, P. U. (2016). *Population Structure and Plant Performance of Juniperus squamata Buch.-Ham. ex D. Don along an Elevation Gradient in Manang, Nepal*. (MSc Thesis), Tribhuvan University, Nepal.

Suwal, M. (2010). *Tree Species Line Advance of* Abies spectabilis *in Manaslu Conservation Area, Nepal Himalaya*. (MSc Thesis), Tribhuvan University, Nepal.

Teketay, D. (1996). *Seed ecology and regeneration in dry Afromontane forests of Ethiopia*. (PhD Thesis), Swedish University of Agricultural Sciences, Umea.

Thakuri, P.S. (2010). *Plant community structure and regenerarion of Quercus semecarpifolia Sm. forests in disturbed and undisturbed areas*. (MSc Thesis), Tribhuvan University, Nepal.

Ueckert, D.N. (2013). *Biology and ecology of redberry juniper*. Texas A&M AgriLife Research and Extension Center.

Verbeke, G. & Molenberghs, G. (2000). *Linear Mixed Models for Longitudinal Data*. USA: Springer.

Vetaas, O.R. (2000). The effect of environmental factors on the regeneration of *Quercus semecarpifolia* Sm. in central Himalaya, Nepal. *Plant Ecology, 146*, 137- 144.

Wang, T., Liang, Y.H., Ren, B., Yu, D., Ni, J. and Ma, K.P. (2004). Age structure of *Picea schrenkiana* forest along an altitudinal gradient in the central Tianshan Mountains, northwestern China. *Forest Ecology and Management, 196* (2-3), 267–274.

Table 4: Frequency of occurrence (%) of vascular plant species associated with *Juniperus indica* in different elevation bands

S.N.	Plant species	Family	Frequency of occurrence (%) in different elevation bands			
			Lower-elevation band	Mid-elevation band	Higher-elevation band	Overall
1.	*Ajuga lupulina* Maxim.	Lamiaceae	0.00	1.39	33.33	11.57
2.	*Allium sikkimense* Baker	Amaryllidaceae	0.00	0.00	2.78	0.93
3.	*Anaphalis triplinervis* (Sims) C. B. Clarke	Asteraceae	0.00	0.00	1.39	0.46
4.	*Anaphalis xylorhiza* Sch. Bip. ex Hook. f.	Asteraceae	0.00	26.39	27.78	18.06
5.	*Anaphalis contorta* (D. Don) Hook. f.	Asteraceae	0.00	0.00	13.89	4.63
6.	*Androsace muscoidea* Duby	Primulaceae	0.00	0.00	11.11	3.70
7.	*Androsace tapete* Maxima.	Primulaceae	0.00	0.00	1.39	0.46
8.	*Anemone rupicola* Cambess.	Ranunculaceae	0.00	0.00	12.50	4.17
9.	*Arabidopsis himalaica* (Edgew.) O.E. Schulz	Brassicaceae	0.00	1.39	13.89	5.09
10.	*Arisaema jacquemontii* Blume	Araceae	0.00	0.00	9.72	3.24
11.	*Artemisia* sp. 1	Asteraceae	16.67	0.00	1.39	6.02
12.	*Artemisia* sp. 2	Asteraceae	0.00	4.17	19.44	7.87
13.	*Artemisia subdigitata* Mattf.	Asteraceae	0.00	12.50	5.56	6.02
14.	*Asparagus filicinus* Buch.-Ham. ex D.Don	Asparagaceae	9.72	0.00	0.00	3.24
15.	*Aster himalaicus* C.B. Clarke	Asteraceae	11.11	25.00	1.39	12.50
16.	*Aster albescens* (DC.) Hand.- Mazz	Asteraceae	20.83	26.39	16.67	21.30
17.	*Astragalus multiceps* Wall. ex Benth.	Fabaceae	1.39	9.72	11.11	7.41
18.	*Astragalus candolleanus* Royle ex Benth.	Fabaceae	0.00	12.50	19.44	10.65
19.	*Berberis aristata DC.*	Berberidaceae	38.89	38.89	37.50	38.43
20.	*Betula utilis* D.Don	Betulaceae	0.00	0.00	4.17	1.39
21.	*Bistorta macrophylla* (D. Don) Sojak	Polygonaceae	0.00	33.33	1.39	11.57
22.	*Bistorta affinis* (D. Don) Greene	Polygonaceae	8.33	0.00	11.11	6.48
23.	*Carex* sp. 1	Cyperaceae	65.28	18.06	29.17	37.50
24.	*Carex* sp. 2	Cyperaceae	56.94	16.67	29.17	34.26
25.	*Carex* sp. 3	Cyperaceae	27.78	18.06	37.50	27.78
26.	*Chesneya nubigena* (D.Don) Ali	Fabaceae	0.00	0.00	4.17	1.39
27.	*Cremanthodium ellisii* (Hook.f.) Kitam.ex Kitam.& Gould	Asteraceae	0.00	0.00	5.56	1.85
28.	*Cicerbita macrorhiza* var. *saxatilis* (Edgew.) P.Brauv	Asteraceae	0.00	2.78	15.28	6.02
29.	*Clematis buchananiana DC.*	Ranunculaceae	6.94	0.00	0.00	2.31
30.	*Coelogyne sp.*	Orchidaceae	4.17	0.00	0.00	1.39
31.	*Conyza sp.*	Asteraceae	0.00	4.17	38.89	14.35
32.	*Cortia depressa* (D.Don) C.Norman	Apiaceae	0.00	0.00	20.83	6.94
33.	*Corydalis juncea* Wall.	Fumariaceae	5.56	16.67	22.22	14.81
34.	*Cotoneaster microphyllus* Wall. ex. Lindl.	Rosaceae	27.78	52.78	19.44	33.33
35.	*Cotoneaster affinis* Lindl.	Rosaceae	0.00	6.94	0.00	2.31
36.	*Cyananthus microphyllus* Edgew.	Campanulaceae	0.00	0.00	2.78	0.93
37.	*Delphinium brunonianum* Royle	Ranunculaceae	0.00	15.28	37.50	17.59
38.	*Ephedra gerardiana* Wall.ex Stapf	Ephedraceae	0.00	0.00	27.78	9.26
39.	*Equisetum sp.*	Equisetaceae	11.11	0.00	0.00	3.70
40.	*Euphorbia himalayensis* Klotzsch	Euphorbiacea	0.00	5.56	6.94	4.17
41.	*Euphorbia stracheyi* Boiss.	Euphorbiacea	0.00	0.00	11.11	3.70
42.	*Fragaria nubicola* Lindl. Ex Lacaita	Rosaceae	0.00	2.78	11.11	4.63
43.	*Galium aparine L.*	Rubiaceae	0.00	0.00	2.78	0.93
44.	*Gentiana robusta* King ex. Hook.	Gentianaceae	0.00	0.00	29.17	9.72
45.	*Gentiana depressa* D.Don	Gentianaceae	0.00	15.28	9.72	8.33
46.	*Gentianella pedunculata* (D.Don) H.Smith	Gentianaceae	0.00	2.78	27.78	10.19

S.N.	Plant species	Family	Frequency of occurrence (%) in different elevation bands			
			Lower-elevation band	Mid-elevation band	Higher-elevation band	Overall
47.	*Gentianella paludosa* (Hook.) H. Sm.	Gentianaceae	0.00	5.56	6.94	4.17
48.	*Kobresia gammiei* C.B. Clarke	Cyperaceae	0.00	13.89	31.94	15.28
49.	*Heracleum obtusifolium* Wall. ex. DC.	Umbeliferae	5.56	0.00	4.17	3.24
50.	*Hippophae tibetana* Schlecht.	Elaeagnaceae	0.00	5.56	0.00	1.85
51.	*Iris kemaonensis* Wallich ex. Royle	Iridaceae	0.00	0.00	12.50	4.17
52.	*Juniperus squamata* Buch-Ham ex. D.Don	Cupressaceae	22.22	40.28	66.67	43.06
53.	*Juniperus communis* L.	Cupressaceae	23.61	29.17	0.00	17.59
54.	*Salix calyculata* Hook.f. ex Andersson	Salicaceae	1.39	0.00	0.00	0.46
55.	*Leontopodium stracheyi* (Hook.f.) C.B. Clarke ex Hemsl.	Asteraceae	16.67	20.83	23.61	20.37
56.	*Ligustrum confusum* Decne.	Oleaceae	0.00	0.00	8.33	2.78
57.	*Lonicera hypoleuca* Decne.	Caprifoliaceae	0.00	5.56	58.33	21.30
58.	*Lonicera minutifolia* Kitam.	Caprifoliaceae	0.00	0.00	19.44	6.48
59.	*Morina nepalensis* D.Don	Morinaceae	20.83	0.00	0.00	6.94
60.	*Oxytropis williamsii* Vass.	Fabaceae	8.33	0.00	0.00	2.78
61.	*Pedicularis pectinata* Wall. Ex. Benth.	Scrophulariaceae	0.00	0.00	19.44	6.48
62.	*Pedicularis rhinanthoides* Schrenk.	Scrophulariaceae	11.11	0.00	0.00	3.70
63.	*Pedicularis flexuosa* Hook. f.	Scrophulariaceae	0.00	13.89	25.00	12.96
64.	*Pinus wallichiana* A.B. Jackson	Pinaceae	11.11	0.00	0.00	3.70
65.	*Pleurospermum apiolens* C.B. Clarke	Apiaceae	2.78	15.28	22.22	13.43
66.	*Polygonatum hookeri* Baker	Convallariaceae	0.00	8.33	23.61	10.65
67.	*Polygonatum cirrhifolium* (Wall.) Royle	Convallariaceae	0.00	5.56	13.89	6.48
68.	*Potentilla fructicosa* Lindl. ex Lehm.	Rosaceae	19.44	34.72	15.28	23.15
69.	*Potentilla peduncularis* D.Don	Rosaceae	0.00	6.94	41.67	16.20
70.	*Primula primulina* (Spreng.) H. Hara	Primulaceae	0.00	18.06	15.28	11.11
71.	*Rhododendron anthopogon* D.Don	Ericaeae	31.94	19.44	0.00	17.13
72.	*Rhododendron lepidotum* Wall. ex D. Don	Ericaeae	0.00	11.11	41.67	17.59
73.	*Rosa sericea* Lindl.	Rosaceae	41.67	63.89	18.06	41.20
74.	*Rumex nepalensis* Spreng.	Polygonaceae	0.00	0.00	22.22	7.41
75.	*Saussurea nepalensis* Spreng.	Asteraceae	0.00	5.56	15.28	6.94
76.	*Saxifraga andersonii* Engl.	Saxifragaceae	0.00	9.72	2.78	4.17
77.	*Spiraea canescens* D.Don	Rosaceae	0.00	0.00	38.89	12.96
78.	*Swertia cuneata* D.Don	Gentianaceae	0.00	0.00	27.78	9.26
79.	*Swertia chirayita* Karsten	Gentianaceae	0.00	0.00	23.61	7.87
80.	*Tanacetum dolichophyllum* Kitam.	Asteraceae	41.67	47.22	73.61	54.17
81.	*Taraxacum eriopodum* DC.	Asteraceae	1.39	4.17	0.00	1.85
82.	*Tanacetum sp.* (local name Khamsang)	Asteraceae	11.11	25.00	65.28	33.80
83.	*Thalictrum cultratum* Wall.	Ranunculaceae	0.00	1.39	27.78	9.72
84.	*Thalictrum alpinum* L.	Ranunculaceae	11.11	22.22	0.00	11.11
85.	*Thymas linearis* Benth.	Lamiaceae	0.00	0.00	12.50	4.17
86.	*Trifolium sp.*	Fabaceae	0.00	5.56	12.50	6.02
87.	*Verbascum thapsus* L.	Scrophulariaceae	4.17	12.50	8.33	8.33
88.	*Viola biflora* L.	Violaceae	0.00	0.00	6.94	2.31

Table 5: Families with number of genera and species recorded from the study area

S.N.	Families	No. of Genera	No. of species
1	Asteraceae	10	16
2	Rosaceae	5	7
3	Gentianaceae	3	6
4	Ranunculaceae	4	5
5	Fabaceae	4	5
6	Scrophulariaceae	2	4
7	Cyperaceae	2	4
8	Primulaceae	2	3
9	Polygonaceae	2	3
10	Cupressaceae	1	3
11	Apiaceae and Lamiaceae	Two each (4)	One each (4)
12	Caprifoliaceae, Convallariaceae, Ericaeae and Euphorbiaceae	One each (4)	Two each (8)
13	Alliaceae, Araceae, Asparagaceae, Berberidaceae, Betulaceae, Brassicaceae, Campanulaceae, Elaeagnaceae, Ephedraceae, Equisetaceae, Fumariaceae, Iridaceae, Morinaceae, Oleaceae, Orchidaceae, Pinaceae, Rubiaceae, Salicaceae, Saxifragaceae, Umbeliferae and Violaceae	21(One each)	21(One each)
Total	37 Families	64 Genera	89 Species

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Effect of *Psidium guajava* L. on Biofilm Forming Multidrug Resistant Extended Spectrum Beta Lactamase (ESBL) Producing *Pseudomonas aeruginosa*

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Abstract

Psidium guajava L. (Guava) commonly known for its food and nutritional values. The medicinal properties of leave of *Psidium guajava* is well known in traditional system of medicine.The relative efficacy of guava leave ethanolic extract and guava leave tea and antibiotic standards was tested against pathogenic *Pseudomonas aeruginosa*. This study was carried out at Microbiology laboratory of KIST medical college and teaching hospital, Lalitpur, a tertiary care hospital from 15th June 2017 to 15th Dec 2017. The isolates were tested for antibiotic susceptibility by Kirby-Bauer disk diffusion method. Biofilm formation was detected by microtitre culture plate method and ESBL production by combine disk methods. Antimicrobial activity of guava leaves were determined by well diffusion method. Ethanol extract of fresh guava leaves exhibited higher antibacterial activity than dry and fresh leaves tea, but significantly less than the standard antibiotics. In this study, 83.67% of biofilm producer, 65.30% of MDR and 6.12% of ESBL producing *P. aeruginosa* were isolated. The leaves extract of guava have shown effective result against *P. aeruginosa* and could serve as good source of antibacterial agents. Guava leaves extract can be an economical ternative to antibiotics. However, active compound of this extract need to be purified and pharmacologically tested before its application.

Keywords: Antibacterial activity, Antimicrobial susceptibility testing, Guava

Introduction

Infectious diseases are the major cause of death in the developing countries and account upto 50% of it. The global burdens of infectious disease are reduced by using antimicrobial agents. However, emergence and spread of resistant pathogen has diminished the effectiveness of the antibiotics (Bisht & Agrawal 2016). *Pseudomonas aeruginosa* is a Gram-negative aerobic and facultative anaerobicbacilli of Pseudomonadaceae family. *P. aeruginosa* is causative agent of various infections such as: urinary tract infections, respiratory infections, otitis media, skin and soft tissue infections, bone and joint infections; and bacteremia. Besides, it can also cause serious systemic infections particularly in people with compromised immune systems including patients of burn suffer, cystic fibrosis, cancer and AIDS (Neopane et al., 2017). It has emerged as one of the leading causes of nosocomial infections. *P. aeruginosa*is the sixth most frequently isolated nosocomial pathogen, causing 7.3% of all hospital acquired infection in the US (Weiner et al., 2016). Infections due to *P. aeruginosa*are difficult to eradicate due to their intrinsic resistance as well as their ability to acquire resistance to different antibiotics. The resistance is due to over expression of efflux pump, acquisition of extended spectrum â-Lactamases (ESBLs) and Metallo-â-Lactamases (MBLs), target site or outer membrane modification, porinmutations and plasmid enzymaticmodification (Heydari & Eftekhar, 2015). *P. aeruginosa*can form biofilms, which exponentially increase antibiotic resistance. The three exo-polysaccharides that mainly contribute to the biofilm formation in this bacteria are; alginate, Psl (Polysaccharides Synthesis Locus), and Pel (Pellicle). Alginate confers additional protection against antimicrobials and the immune system while Psl and Pel contribute to aggregation and adherence (Nithyalakshmi et al., 2015). Hence, dealing with multi-drug resistant strain of these bacteria is challenging.

Various types of plants contain natural preservatives which have antimicrobial and antioxidant property.

Guava leave (*Psidium gaujava* L), a phyto-therapeutic plant used in folk medicine, is believed to have active components whichcan be used to treat and cure various diseases (Mailoa et al., 2014). Different parts of the plant have been used in traditional medicine against ailmentslike malaria, gastroenteritis, vomiting, diarrhoea, dysentery, wounds, ulcers, toothache, coughs, sore throat, inflamed gums, etc. Besides, it has also been used for the controlling of diabetes, hypertension and obesity (Biswas et al., 2013).

In Nepal, study has shown the emergence of multidrug resistant (MDR) isolates of *P. aeruginosa* associated with nosocomial infections. The occurrence of biofilm and MDR bacteria in a hospital setting possess a therapeutic problem, as well as a serious concern for infection control management. Therefore minimizing the use of antibiotics and where possible substituting with antimicrobial compound from other source can be an alternative to subside the growing antibiotic resistant problem. Guava leaf tea and extract from it with antimicrobial activity can be a choice. This study was therefore undertaken to evaluate the antimicrobial activity of guava leaves on *P. aeruginosa* isolated from various clinical samples.

Materials and Methods

P. aeruginosa isolated from different clinical samples such as pus/wound, blood, sputum and urine wereidentified by standard microbiological techniques. The isolates were further tested for antibiotic susceptibility by Kirby-Bauer disk diffusion method on Mueller Hinton agar as per CLSI guidelines. ESBL production was screened using two disks, ceftazidime (30µg) and cefotaxime (30 µg) according to the CLSI guidelines. An inhibition zone of d≤22 mm for ceftazidime and d≤ 27 mm for cefotaxime indicated as ESBL producing strain which was further confirmed bycombination disc method.

Detection of biofilm production

Biofilm production was detected by microtitre culture plate method (TCP), a quantitative test as described by Christensen et al.(1995). Organisms isolated from fresh agar plates were inoculated in 10 mL of trypticase soy broth with 1% glucose and were incubated at 37ÚC for 24 h. The cultures were then diluted 1:100 with fresh medium. Sterile 96 flat bottomed polystyrene tissue culture plates were filled with 200 µL of the diluted cultures in each well. The control organisms used was *P. aeruginosa* ATCC 27853. For negative controls wells contained sterile broth without inoculum. The plates were incubated at 37°C for 24 hr. After incubation, contents of each wells were removed by gentle tapping. The wells were washed with 0.2 ml of phosphate buffer saline (pH 7.2) for four times to remove free floating bacteria. Biofilm formed by bacteria in the wells were fixed by 2% sodium acetate and stained by crystal violet (0.1%). Excess stain was removed by using deionized water and plates were kept for drying. Optical density (OD) of stained biofilm was obtained by using micro ELISA autoreaderat wavelength 570 nm. The experiment was performed in triplicate with three repetition. Interpretation of biofilm production was done according to the criteria explained by Stepanovic et al. (2000).

Preparation of guava tea and ethanol extract

Guava leaf samples were randomly collected from Kathmandu Valley. The leaves were washed with sterile distilled water. Extract first (fresh leaf tea) was prepared (10 leaves were taken in conical flask containing 200 ml of distilled water and boiled for 10-20 minutes). Extract second (dry leaf tea) was prepared from freeze dried leaves of the same weight and numbers. Similarly tea was prepared by same procedure. Extract third and fourth (ethanol extract of fresh and dried leaves respectively) were prepared by using absolute ethanol as a solvent. The leave pieces were added to solvent. The mixtures were made in sterile flask wrapped in aluminum foil to avoid evaporation and exposed to light for 3 days at room temperature. The flasks were placed on a platform shaker at 70 rpm. After 3 days of soaking in solvent, the mixtures were transferred to tubes and centrifuged for 10 min at 4,000 rpm at 25°C. The supernatant was collected and stored at 4°C until use.

Determination of antimicrobial activity of guava leave tea against P. aeruginosa

Antimicrobial susceptibility testing was done using agar diffusionmethod in triplicate according to the National Committee for Clinical Laboratory Standards guidelines. The plant extracts were tested on Mueller Hinton Agar (MHA) plates against *P. aeruginosa* to detect the antibacterial activity. Prior to streaking the plates with bacteria, 5mm diameter wells were punched into the medium using a sterile borer. All plates were inoculated with the broth culture of isolated *P. aeruginosa* which has been previously adjusted to a 0.5 McFarland standard solution.A sterile cotton swab was dipped into the bacterial suspension, rotated several times, and pressed firmly to the inside wall of the tube above the fluid level removing excess inoculum. The surface of the agar plate was streaked over the entire sterile agar surface rotating the plate to ensuring even distribution of inoculum. The plates are allowed stand for 5minto absorb the excess moisture content. Exactly 100µl aliquots of each test extract and leave tea were dispensed into each well of MHA plates swabbed with bacteria. For positive control an antibiotic tobramicin was placed at the centre of the plate.While ethanol was usedfor negative control. *P. aeruginosa* ATCC 27853 was used for positive control organism.

Results and Discussion

A total of 3000 specimens were processed and bacterial growth was observed in 25% samples and *P. aeruginosa* was isolated from 7% of the total sample (Figure 1)

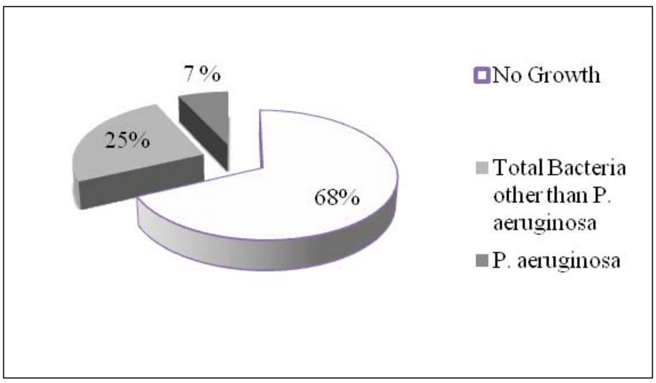

Figure1: Growth pattern of bacterial isolates in clinical samples

Prevalence of MDR P. aeruginosa

Among the total *P. aeruginosa* isolates 65.3% (*n*=32) were MDR (Figure 2).

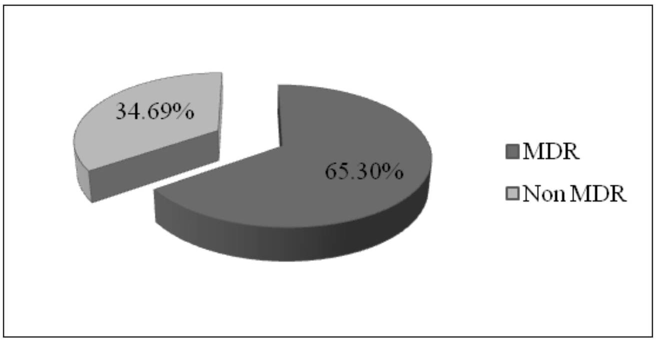

Figure 2: Percentage of MDR strain from the total isolates of *P. aeruginosa*

Antibiotic susceptibility pattern of P. aeruginosa isolates

Most of the isolates were sensitive towards colistin and resistant towards cefepime antibiotics (Table 1).

Table 1: Antibiotic susceptibility pattern of *P. aeruginosa*

Antibiotic	Sensitive%	Intermediate%	Resistant%
Piperacillin	31(63.3)	-	18 (36.7)
Piperacillin/Tazobactam	27 (55.1)	3 (6.1)	19 (38.8)
Ceftriaxone	20 (40.8)	7 (14.3)	22(46.9)
Ceftazidime	22(44.9)	4(8.2)	23(46.9)
Cefepime	16(32.7)	-	33(67.3)
Aztreonam	29(59.2)	6(12.2)	14(28.6)
Gentamicin	29(59.2)	-	20(40.8)
Amikacin	24(49.0)	1(2.0)	24(49.0)
Imipenum	19(38.8)	1(2.0)	29(59.2)
Meropenem	24(49.0)	2(4.1)	23(46.9)
Ciprofloxacin	30(61.0)	-	19(38.8)
Colistin	46(93.9)	-	3(6.1)
Polymyxin-B	32(65.3)	5(10.2)	12(24.5)
Cefotaxime	20(40.8)	3(6.1)	26(53.1)
Tobramycin	29(59.2)	1(2.0)	19(38.8)

Biofilm production detection in P. aeruginosa

Out of 49 isolates of *P. aeruginosa* 35(71.40%)were strong biofilm producer, while 6(12.24%) were weak producers and 8(16.32%) were non-biofilm producers (Figure 3).

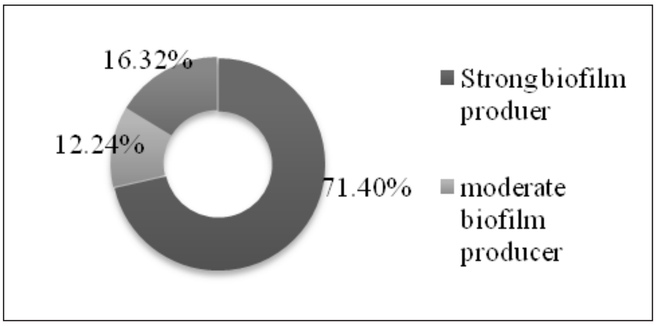

Figure 3: Percentage of biofilm producing *P. aeruginosa*

ESBL production in P. aeruginosa

Among the total *P. aeruginosa* isolates, 3 (6.12%) were found to be ESBL producer. Significant numbers of biofilm producers are MDR isolates ($p<0.05$) (Table 2).

Table 2: Comparison between MDR and biofilm production

MDR	Biofilm producer		Total (%)	p-value
	Positive (%)	Negative (%)		
Positive	31 (96.8)	1 (3.2)	32 (100.0)	0.02
Negative	10 (58.82)	7 (41.18)	17 (100.0)	(<0.05)
Total	**41 (83.67)**	**8 (16.33)**	**49 (100.0)**	

Antimicrobial activity of guava leave

Guava leaf tea (fresh and dry leave tea) showed its antimicrobial activity against all types of *P. aeruginosa* isolates. However, compared to the antimicrobial activity of standard antibiotictobramicin, the zone of inhibition shown by guava leavewas less (Figure 4). The inhibition zone given by fresh leave for both guava leaf tea and ethanol extract were higher compared to the dry leave (Figure 4 and 5).

P. aeruginosa is an important opportunistic nosocomial pathogen of great important due to its

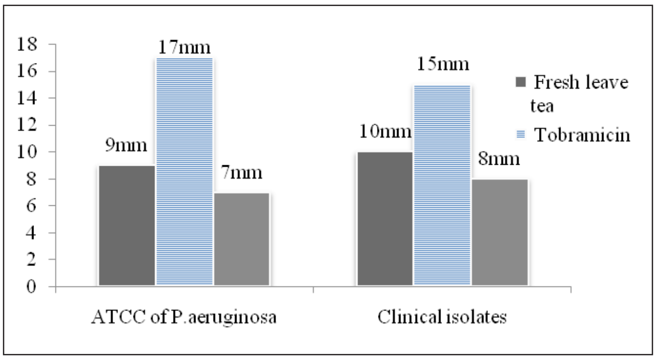

Figure 4: Antimicrobial activity of guava leave tea and tobramicin against *P.aeruginosa*

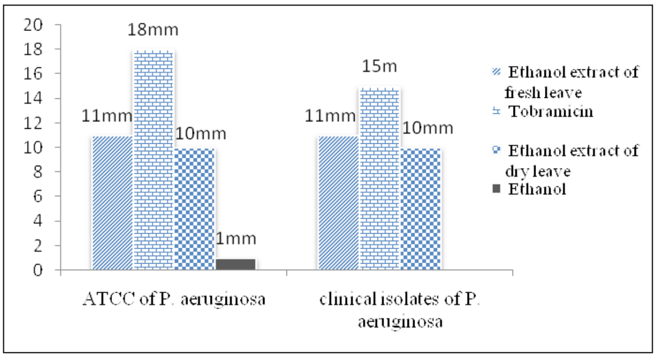

Figure 5: Antimicrobial activity of ethanol extract of guava leaves and tobramicin against *P.aeruginosa*

resistance to multiple antibiotics (Goel et al., 2013). In this study, prevalence of *P. aeruginosa* from different clinical samples was found to be 27.22% which is less compared to Ali et al., (2015) and Goel et al., (2013) who reported 39% and 37.7% respectively from Pakistan and India.

High prevalence of MDR *P. aeruginosa* (65.3%) was reported in the study of Fatima et al., (2012) where MDR isolates accounted for 73.9%. This findingis also be corroborated with MDR *P. aeruginosa* in Southeast Asia, where 71% reported during 2007-2009 (Suwantarat & Carrol 2016). A high prevalence of MDR *P. aeruginosa* seen in hospital acquired infections wasdue to selective pressure exerted byover usage of broad spectrum antibiotics. However, the emergence of MDR is related to the empirical use of antibiotics rather than the rational use of broad-spectrum antibiotics before the sample collection.

Prevalence of ESBL producing *P. aeruginosa* was 6.12%. Among the ESBL producer 66.67% were MDR and 6.25% MDR were ESBL producer, which issimilar to a study by Stepanovic et al. (2000). But different to Shaikh et al. (2015) who reported 25.13% isolates of *P. aeruginosa* were observed as ESBL positive among the 187 samples. The ESBL producing *P. aeruginosa* isolates exhibited co-resistance against most of the antibiotics tested.

In this study, 83.67% isolates of *P. aeruginosa* were biofilm producer. Maita and Boonbumrung (2014) reported 79.4% biofilm producer in Thailand. A little higher 89.3% was reported by Sharma and Chaudhary (2015) but lower (48.8%) by

Tamaraiselvi et al. (2015). The biofilm production was found to be independent of the antibiotic susceptibility profile of the bacteria. When the degree of adhesion of the biofilm is high, the penetration of the antimicrobial compound into its structure is reduced resulting in the increased resistance of the bacteria.

Antibacterial activity of ethanol extract of fresh leave showed higher activity (11mm) followed by ethanol extract of dry leaves and least activity by dry guava tea solution (7mm). In Philippines, Cruzada et al. (2014) did quantification of biofilm in microtiter plates for overview of testing conditions and practical recommendations for assessment of biofilm production by Staphylococci. Different concentration of guava extract exhibited antibacterial strengths against both, *E. coli* and *P. aeruginosa* bacteria but was significantly less than the standard antibiotics. Biswas et al. (2013) has reported that *P. guajava* has antibacterial effect against both Gram-negative and Gram-positive bacteria. It was due to presence of alkaloids, flavonoids, tannins, saponins, glycosides and terpenoids in the leaves extracts of *P. guajava* (Savoia, 2012). These phytochemicals have in vitroinhibitory activity against some clinical bacterial isolates. In Brazil, Sanches et al. (2005) reported that the aqueous extracts of *P. guajava* leaves, roots and stem bark were active against the Gram positive bacteria but not against Gramnegative species. This can be due to the outer membrane of Gram negative bacteria which act as barrier to penetration of numerous antibiotic molecules. Besides, the enzymes present in the periplasmic space have ability to break down foreign molecules.

Conclusion

P. aeruginosa is a pathogen of interest in most of the hospital acquired infection. Increase in the drug resistant *P. aeruginosa* is a great challenge in treatment of infections caused by it.Since crude ethanol extract of guava leave and its tea was able to inhibit *P. aeruginosa.* The compound present in guava leave extract and its tea contain some antimicrobial compounds which can effectively control pathogenic bacteria.It has potential for use in therapy against infections caused by pathogens. Thus can be recommendedadditional test of pure extracts along with further pharmacological evaluation is needed.

Acknowledgments

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References

Ali, Z., Mumtaz, N., Naz, S.A., Jabeen, N. & Shafique, M. (2015). Multi-drug Resistant *Pseudomonas aeruginosa*.A threat of nosocomial infections in tertiary care hospitals.*J Pak Med*.,*65*, 10-21.

Bisht & Agrawal. (2016). Antimicrobial and phytochemical analysis of leaf extract of medicinal plants. *Asian J Pharm Clic*., *9*, 131-136.

Biswas, B., Rogers, K., McLaughlin, F., Daniels D & Yadav, A. (2013). Antimicrobial activities of leaf extracts of guava (*Psidiumguajava* L.) on two Gram-negative and Gram-positive bacteria. *Int J of Microbio*., ***7***, 122-129.

Christensen, G.D., Simpson, W.A. & Younger, J.A. (1995). Adherence of coagulase negative Staphylococci to plastic tissue cultures: a quantitative model for the adherence of Staphylococci to medical devices. *J Clin Microbiol*., *22*, 996-1006.

Cruzada, C. J.O., Co, K.J. , Chua, T.M., Chuabio, V.A., Cruz, K.M., De Castro, D.A., Chua, J.S. & Chua, J.E. (2014). Evaluation of *Psidiumguajava* (Guava) leaf extract against Gram-negative bacteria in planktonic and biofilm lifestyles. *Annual Research & Review in Biology*, *4*, 4370-4380.

Fatima, A., Naqvi, S.B., Kali SA, Perveen S and Jabeen S (2012). Antimicrobial susceptibility pattern of clinical isolates of *Pseudomonas aeruginosa* isolated from patients of lower respiratory tract infections. *Springer Plus 1,* 70.

Goel V, Sumati A, Hogade, S.A. & Karadesai, S.G. (2013). Prevalence of extended spectrum β - latamases, AmpC beta lactamase, and metallo-β - lactamase producing *Pseudomonas aeruginosa* and *Acinetobacterbaumanni* in an intensive care unit in a tertiary care hospital. *J SciSco*., *40*, 28-31.

Heydari, S. & Eftekhar, F. (2015). Biofilm formation and â-Lactamase production in burn isolates of *Pseudomonas aeruginosa*. Jundishapur. *J Microbio*., *4,*111-123.

Mailoa, M.N., Mahendradatta, M., Laga, M. & Djide, N. (2014). Antimicrobial activities of tannins extract from guava leaves (*Psidiumgaujava* L) on pathogens microbial. *Int J of Sci and Tech Res*., *1*, 236-241.

Maita, P. & Boonbumrung, K.(2014). Association between biofilm formation of *pseudomonas aeruginosa* clinical isolates versus antibiotics resistant and genes involved with biofilm. *J Chem and Pharma Res*., 6, 1022-1028.

Mendiratta, D.K., Deotale, V. & Narang, P. (2005). Metallo beta lactamase producing *Pseudomonas aeruginosa* in a hospital from rural area. *Indian J. Med. Res.*,701-703.

Neopane, P., Nepal, H.P., Gautam, R., Paudel, R., Ansari, S. & Shrestha, S. (2017). Is there correlation of biofilm formation with multidrug resistance and esbl production in *Pseudomonas aeruginosa*. *European J. of Biomed*, *4*, 366-372.

Nithyalakshmi, J., Akila, K., Mohanakrishnan, K. & Sumathi, G. (2015). Evaluation of resistance profile of *Pseudomonas aeruginosa* with reference to biofilm production – An emerging challenge. *J. of Den and Med Sci*.,14, 13-18.

Sanches, R.N., Garcia, A.D., Schiavini, S.M., Nakamura, C.V. & Filho, D.B. (2005). An evaluation of antibacterial activities of *P. guajava* (L). *An Int J*., *48*,429-436.

Savoia, D. (2012). Plant-derived antimicrobial compounds. *Future Microbio*.,*7*, 979-990.

Shaikh, S., Fatima, J., Shakil, S., Mohd, S., Rizyi, D. & Amijakamal, M. (2015). Prevalence of multidrug resistant and extended spectrum beta-lactamase producing *Pseudomonas aeruginosa* in a tertiary care hospital. *Saudi J of Bio Sci*.,*22*,62-64.

Sharma, I. & Chaudhary, D. (2015). Detection of Pel A Gene in *P.aeruginosa* from clinical samples using polymerase chain reaction with reference to biofilm production in N E India.*Ind J Res*., *4*, 119-121.

Stepanovic, S., Vukovi, D. & Hola, V. (2000). Quantification of biofilm in microtiter plates: overview of testing conditions and practical recommendations for assessment of biofilm production by Staphylococci. *APMIS, 115*, 891-899.

Suwantarat, N. & Carroll, K.C. (2016). Epidemiology and molecular characterization of multi-drug resistant Gram negative bacteria in Southeast Asia. *Antimicrob Res and Infect Cont*., *5*, 1-8.

Tamaraiselvi, S., Swarma, SR., Madhavan, R. & Gomathi, S. (2015). Biofilm production and multi drug resistant among the isolates from acute and chronic wound infection. *Int J Pharma Bio Sci*.,*6*, 1197-1203.

Weiner, LM., Webb, AK., Limbago, B., Dudeck, MA., Patel, J., Kallen, AJ., Edwards, JR. & Sievert, DM. (2016). Antimicrobial resistant pathogens associated with healthcare-associated infections: summary of data reported to the national healthcare safety network at the Centres for Disease Control and Prevention, 2011-2014. *Infect Cont and HospEpid*., *4*, 1-14.

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Antimicrobial and Phytochemical Studies of Methanolic Bark Extract of *Psidium guajava* L. and *Punica granatum* L.

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Abstract

Psidium gaujava L. and *Punica granatum* L., commonly known as guava and pomegranate respectively are popular for their edible fruits. In addition, they also have medicinal value. Local people use their bark and leaves to cure many ailments. The aim of this study was to determine the antimicrobial and phytochemical properties of bark samples of guava and collected from Tinpatan-3, Bagthala, Sindhuli, Nepal during March, 2017. Methanolic extracts of the collected bark samples were evaluated for their antimicrobial activities against 12 microorganisms by agar well diffusion method. Pomegranate bark extract was found most active against methicillin-resistant *Staphylococcus aureus* (MRSA) and *Proteus vulgaris* showing zone of inhibitions (ZOIs) 25mm and 22mm respectively, and minimum microbicidal concentrations (MMCs) 6.25 $mg.ml^{-1}$ and 0.78125 mg.ml-1 respectively. Guava bark extract was found most active against *Proteus vulgaris* with ZOI value 18mm and MMC value 1.5625 mg.ml-1. Phytochemical screening of guava bark extract confirmed the presence of alkaloids, flavonoids, reducing sugar and steroids, and absence of volatile oils, terpenoids, tannins, saponins and proteins. Pomegranate bark extract showed presence of alkaloids, terpenoids, flavonoids, tannins, saponnins, reducing sugar and steroids, and absence of volatile oils and proteins.

Keywords: Extraction, Medicinal plants. Microorganism, Minimum inhibitory concentration (MIC), Zone of inhibition (ZOI)

Introduction

Medicinal plants are in use to treat different diseases since ancient time. Increasing resistance pattern and associated side effects of antibiotics have evolved the importance of medicinal plants to be used as an antibacterial agent (Sajjad et al., 2015). An antimicrobial is the physical or chemical agent that kills or inhibits the growth of micro-organisms such as bacteria, fungi, protozoa. Medicinal plants constitute several bioactive compounds that show antimicrobial and antioxidant activities (Kundal, 2013). Phyto constituents employed by plants to protect themselves against pathogenic insects, bacteria, fungi or protozoa have found applications in human medicine (Nascimento et al., 2000). Plant diversity serves the humankind as renewable natural resources for a variety of biologically active chemicals. These chemicals bear a variety of properties viz antibacterial, antifungal, antiviral, anthelmintic, anticancer, sedative, laxative, cardiotonic, diuretic and others (Parajuli et al., 1998). Medicinal plants represent a rich source of antimicrobial agents (Abi Beaulah et al., 2011).

Psidium guajava L., commonly known as guava, belongs to Myrtaceae family. It is locally called ambaa in Nepali and is popular for its edible fruits. The plant is about 4-6m tall with peeling, reddish brown bark on young branches. Leaves are very short petioled, ovate or oblong, 7-10cm long, veins prominent, coriacious, old leaves are reddish brown. Flowers are white, peduncled and axillary. Fruits are globose or pear-shaped, yellow and many seeded. *P. guajava* plant is native of Brazil and it is cultivated in Nepal. (Department of Plant Resources [DPR], 2016).

Punica granatum L., commonly known as pomegranate, belongs to Lythraceae family. It is a deciduous spiny tree of 5-10m tall. Its leaves are simple, glossy, opposite, oblong or obovate, 2-6cm long, narrowed to a short petiole. Flowers are bright red, rarely white with thick fleshy petals. Fruits are ovoid, crowned with persistent calyx, outer surface woody, smooth, brownish red, 5-8cm in diameter and containing much red juice around the seed. Seeds are white, 1-2cm long. Flowering and fruiting time is May to June. (DPR, 2016).

Rokaya et al. (2014) found that bark of 152 plant species were used to cure gastrointestinal disorder in Nepal. Pomegranate and guava are popular not only for their edible fruits, but also for their stem, bark and leaf which are used for various purposes by the different communities in remote areas of Nepal. Traditional practices of local people in Nepal shows that stem bark decoction of guava is used to treat fever, diarrhea and dysentery, and leaf juice is taken to treat bowels, cuts, wounds and ulcers. Leaf bud is chewed to treat headache (Malla & Chhetri, 2009; Acharya, R., 2012; Thapa, S., 2012). Stem bark of pomegranate is also used to get relief during diarrhea and dysentery. This is due to medicinal properties found in the bark and leaf of this plant. Antimicrobial studies support and provide scientific basis for the traditional medical practices of local communities. In this study methanolic bark extracts of pomegranate and guava were screened for phytochemical and antimicrobial properties.

Materials and Methods

Collection and processing of samples

The bark samples of guava and pomegranate were collected in March, 2017 from Tinpatan-4, Bagthala, Sindhuli, Nepal. The bark samples were collected from the branch in strips of 3 inches randomly along the length of the tree taking precautions to avoid girdling. The collected samples were washed thoroughly, chopped into small pieces, dried in hot air oven at 60ºC for 24 hours and ground into powder.

Extraction of plant materials

Powder of bark of pomegranate (35.5gm) and guava (15.2gm) were loaded for the Soxhlet extraction with methanol for 72 hours till the colorless solvent appeared in the siphon to obtain crude methanol extract of respective plants. After complete extraction, solvent, i.e. methanol, was evaporated with the help of rotary vacuum evaporator using the water bath below 65ºC. Solvent was completely evaporated and condensed solvent was collected in the separate round bottom flask (Eloff, 1998; Tiwari et al., 2011).

Percentage yield of the extract was calculated by using the following formula:

$$\text{Percentage yield} = \frac{\text{Initial weight of the sample} - \text{final weight of the sample}}{\text{Initial weight of the sample}} \times 100\%$$

Antimicrobial activity

Test organisms: *Bacillus subtilis* (Ehrenberg) Cohn, *Enterococcus faecalis* (Andrewes & Horder) Schleifer & Kilpper-Balz, *Staphylococcus aureus* Rosenbach, methicillin-resistant *S. aureus* (MRSA), *Escherichia coli* (Migula) Castellani & Chalmers, *Klebsiella pneumoniae* (Schroeter) Trevisan, *Proteus vulgaris* Hauser, *Pseudomonas aeruginosa* (Schröter) Migula, *Salmonella enterica* subsp. *enterica* (ex Kauffmann & Edwards) Le Minor & Popoffserovar. Typhii, *Shigella dysenteriae* (Shiga) Castellani & Chalmers, *Candida albicans* (C.P. Robin) Berkhout, *Saccharomyces cerevisiae* Meyen ex E.C. Hansenwere used as test organisms.

Preparation of the working solutions: Sterilized screw-capped tubes were calibrated and marked for 10ml. About 1 gm of extract was transferred into the calibrated tube. Methanol (solvent used for extraction) was added to the tube making the final volume to 10ml. Mixture was then homogenized by vortexing.

Preparation of standard culture inoculums: Required numbers of 18-24 hours old colonies of test organisms were inoculated aseptically to separate sterilized vials containing 5 ml of sterilized nutrient broth and were homogenized by vortexing. The inoculum, so prepared, was compared with turbidity of 0.5 McFarland Nephelometer standard recommended by World Health Organization [WHO] (1991) for antimicrobial susceptibility test.

Screening and evaluation of antimicrobial activity: The extract samples were screened for antimicrobial activity using agar well diffusion methods as described by Perez et al. (1990).

A sterile swab was used to evenly distribute inoculums over Muller-Hinton Agar (MHA) for bacteria and Muller-Hinton Agar with Glucose and Methylene Blue (MHA, GMB) for fungi. The plate was rotated through an angle of 60º after each swabbing. The swabbing was done three times. The inoculated plates were allowed to dry for maximum 15 minutes. Four wells, each of 6 mm diameter, were created in the inoculated plates using a sterile cork borer (three wells for test samples and one well for the solvent as negative control). Micropipettes were used to dispense 50µl of the test solution of the extract samples and solvent as negative control into each of the four wells. The plates were left in the upright condition with lids closed for half an hour so that the test solutions diffused into the media. The inoculated plates were then incubated in inverted position at suitable temperature (35±2°C for bacteria and 25±2°C for fungi). After proper incubation (18-24 hours for bacteria, 24-48 hrs for fungi) the plates were examined for zone of inhibition (ZOI) around the well which is suggested by clear area with no growth of organisms. Diameter of each ZOI was measured using digital vernier caliper to the nearest whole millimeter (Rana et al., 2017).

Determination of Minimum Inhibitory Concentration (MIC) and Minimum Microbicidal Concentration (MMC): MIC was determined by observing the visible growth of the test microorganism in two-fold serial diluted antimicrobial substances in broth culture medium while MBC was determined by sub culturing the MIC cultures on suitable agar plates (Forbes et al., 2007).

The crude extract of medicinal plants, which showed zone of inhibition (ZOI), were subjected to two-fold serial dilution method to determine the MIC and further MMC. A set of 12 screw-capped vials, each containing 1 ml Muller Hinton Broth (MHB) for bacterium or 1 ml Sabouraud Dextrose Broth (SDB) for fungus, were prepared. The vials were then numbered from 0 to 11. MHB/SDB was discarded from vial no. 0 and 1 ml test solution was added. Then, 1 ml of test solution was added to vial no. 1 and was homogenized by vortexing. From it, 1 ml content was transferred aseptically to vial no. 2 followed by homogenization. This process was repeated till two-fold serial dilution was done up to vial no. 10. Finally, 1 ml of the content was discarded from vial no. 10 after homogenization. Hence, all the vials contain equal volume i.e. 1 ml with gradually decreasing concentration. Now with the help of micropipette, 20µl of inoculums (a 1:100 dilution of a suspension of turbidity equal volume to McFarland Standard 0.5 supposed to have organism 1.5×10^6 CFU.ml^{-1}) was added to all vials except vial no. 0. All the tubes were incubated at 37±2ºC for 18-24 hours for bacteria and 25±2ºC for 24-48 hours for fungi. The tubes were then observed for turbidity and MIC of an extract was determined as the lowest concentration of antimicrobial agent in the two-fold dilution series which the inhibited the growth of the test organisms (i.e. the lowest concentration in two-fold dilution series without turbidity). The vials were sub-cultured on nutrient agar plates and incubated at 35±2ºC for 18-24 hrs (for bacteria) or potato dextrose agar plates at 25±2ºC for 24-48 hrs (for fungi). Then, plates were examined for the growth of microorganisms. The tubes with minimum concentration of extract in which the growth was completely checked was noted as the MBC of the plant extract (Grumachhan, 2018; Gurmachhan et al., 2019).

Phytochemical screening

The phytochemical screenings of the plant extracts were carried out according to the standard protocol (Ciulei, 1982; Harborne, 1998). The barks of the plants were dried and extracted with methanol. Different phytochemicals in the extracts were identified by color reactions with different reagents.

Results and Discussion

Among the bark samples subjected to Soxhlet extraction with methanol, guava bark showed higher extract yield of 22%, while that of pomegranate showed comparatively lower yield of 19.7% (Table 1).

Antimicrobial activities of bark extracts were tested against twelve test organisms, amongst which pomegranate bark extract showed antimicrobial activities against *Bacillus subtilis* (Ehrenberg) Cohn, *Enterococcus faecalis* (Andrewes & Horder) Schleifer & Kilpper-Balz, *Staphylococcus aureus* Rosenbach, methicillin resistant *S. aureus* (MRSA), *Proteus vulgaris* Hauser and *Candida albicans* (C.P. Robin) Berkhout. Similarly, guava bark extract showed antimicrobial activities against *B. subtilis*, *E. faecalis*, *S. aureus*, MRSA and *P. vulgaris*. The ZOI values of methanolic bark extract of guava bark extract were found to be 14 mm, 18 mm, 13 mm, 18 mm and 18 mm against *B. subtilis*, *E. faecalis*,

Table 1: Percentage yields of bark extracts

S.N.	Plants	Parts used	Sample weight (gm)	Total thimble weight with sample			Percentage yield
				Before extraction (gm)	After extraction (gm)	Weight of extract (gm)	
1	*Psidium guajava* L.	Bark	35.5	39.4	31.6	7.8	22.0
2	*Punica granatum* L.	Bark	15.2	19.1	16.1	3.0	19.7

Table 2: Zones of inhibition (ZOIs) of bark extracts (mm)

S.N.	Name of microorganisms	Zones of inhibition (ZOIs) (mm)	
		***Psidium guajava* L.**	***Punica granatum* L.**
1	*Bacillus subtilis* (Ehrenberg) Cohn	14	18
2	*Enterococcus faecalis*(Andrewes & Horder) Schleifer & Kilpper-Balz	18	21
3	*Staphylococcus aureus* Rosenbach	13	16
4	*Methicillin Resistant S. aureus* (MRSA)	18	25
5	*Escherichia coli* (Migula) Castellani & Chalmers	0	0
6	*Klebsiella pneumoniae* (Schroeter) Trevisan	0	0
7	*Pseudomonas aeruginosa* (Schröter) Migula	0	0
8	*Proteus vulgaris* Hauser	18	22
9	*Salmonella enterica subsp. enterica* (ex Kauffmann & Edwards) Le Minor & Popoff serovar. *Typhi*	0	0
10	*Shigella dysenteriae*(Shiga) Castellani & Chalmers	0	0
11	*Candida albicans* (C.P. Robin) Berkhout	0	14
12	*Saccharomyces cerevisiae* Meyen ex E.C. Hansen	0	0

Table 3: Minimum microbicidal concentration of methanolic bark extract of plants

S.N.	Plants	Minimum Microbicidal Concentration ($mg.ml^{-1}$)					
		Bacillus subtilis	*Enterococcus faecalis*	*Staphylococcus aureus*	Methicillin Resistant *S. aureus* (MRSA)	*Proteus vulgaris*	*Candida albicans*
1	*Psidium guajava* L.	>50	6.25	6.25	12.5	1.5625	ND
2	*Punica granatum* L.	>50	6.25	1.5625	6.25	0.78125	12.5

ND= not done due to 0 ZOI

Table 4: Results of phytochemical screening of bark extracts

Plants	Volatile oils	Alkaloids	Terpenoids	Flavonoids	Tannins	Saponins	Glycosides	Reducing sugar	Steroids	Protein
Psidium guajava L.	-	+	-	+	-	-	±	+	+	-
Punica granatum L.	-	+	+	+	+	+	±	+	+	-

Note: + indicate Presence, - indicate absence, ± indicate may or may not

S.aureus, MRSA and *P. vulgaris* respectively. Similarly, ZOI values of pomegranate bark extract were found to be 18 mm, 21 mm, 16 mm, 25 mm, 22 mm and 14 mm against *B. subtilis, E. faecalis, S.aureus, MRSA, P. vulgaris* and *C. albicans* respectively. Methanolic bark extract of pomegranate formed comparatively larger ZOIs against Methicillin Resistant *S. aureus* (MRSA) and *P. vulgaris* while that of guava did so against *P. vulgaris* and *S. aureus*. Among all the ZOIs formed by the two extracts, the largest ZOI observed was of 25 mm diameter which was formed by methanolic bark extract of pomegranate against MRSA (Table 2).

Minimum microbicidal concentration (MMC) values of guava bark extract were found 1.5625 $mg.ml^{-1}$, 6.25 $mg.ml^{-1}$, 6.25 $mg.ml^{-1}$ and 12.5 $mg.ml^{-1}$ against *P. vulgaris, E. faecalis, S. aureus* and MRSA respectively. Similarly, MMC values of pomegranate bark extract were found 0.78125 $mg.ml^{-1}$, 1.5625 $mg.ml^{-1}$, 6.25 $mg.ml^{-1}$, 6.25 $mg.ml^{-1}$ and 12.5 $mg.ml^{-1}$ against *P. vulgaris, S.aureus, E. faecalis*, MRSA and *C.albicans* respectively. MMC value of sample extract was found >50 $mg.ml^{-1}$ against *B. subtilis*. MMC value 0.78125 $mg.ml^{-1}$ of pomegranate bark extract against *P. vulgaris* was found as the lowest MMC value. (Table 3). Hamid et al. (2015) found the ZOI values of methanolic extract of bark of pomegranate 23.3 mm, 22 mm, 22 mm, 22 mm and 20.4 mm against *K. pneumonia, E. coli, P. aeruginosa, S. aureus* and *E. faecalis* respectively. They reported minimum inhibitory concentration (MIC) values of 6.10 $mg.ml^{-1}$, 6.25 $mg.ml^{-1}$, 6.0 $mg.ml^{-1}$, 6.2 $mg.ml^{-1}$ and 6.8 $mg.ml^{-1}$ against *K. pneumonia, E. coli, P. aeruginosa, S. aureus and E. faecalis* respectively. Kuber et al. (2013) found the ZOI values 1.8 cm, 1.1 cm, 2 cm and 1.5 cm at 10 $mg.ml^{-1}$ concentration of guava root bark extract against *S. aureus, E. coli, B. subtilis* and *P. vulgaris* respectively.

B. subtilis sometimes causes food poisoning. *E. faecalis* causes endocarditis & septicemia, urinary tract infection, meningitis and other infections in humans. *S. aureus* causes skin infection, boils, conjunctivitis, secondary infections, pneumonia, acute endocarditis toxic shock syndrome and food poisoning (Collee et al., 1996). MRSA is responsible for several difficult to treat infections in human pathogens. MRSA is common in hospitals, prisons and nursing homes where the people with open wounds, invasive devices such as catheters. In human, S. aureus is part of the normal microbiota present in the upper respiratory tract, on skin and in the gut mucosa. *S. aureus* along with similar species that can colonize and act symbiotically but can cause disease if they begin to take over the tissues they have colonized or invade other tissues, have been called pathobionts. After 72 hours MRSA can take hold in human tissues and eventually become resistant to treatment. *P. vulgaris* occasionally causes urinary tract infections, wound infection and abscesses and form cases of otitis media, meningitis, septicemia (Cheesbrough, 2000).

Phytochemical screening of methanol extract of guava bark showed the presence of alkaloids, flavonoids, reducing sugar, steroids and absence of volatile oils, terpenoids, tannins, saponins, and protein. Similarly, the methanol extract of pomegranate bark indicated the presence of alkaloids, terpenoids, flavonoids, tannins, saponins, reducing sugar, steroids and absence of volatile oils and protein. The study could not confirm the presence or absence of glycosides (Table 4). Phytochemical constituents such as alkaloids, flavonoids, tannins, phenols, saponins, and several other aromatic compounds are secondary metabolites of plants that serve as defenses against many microorganisms, insects and other herbivores (Bonjar et al., 2004; Shihabudeen et al., 2010). Sajjad (2015) found presence of glycosides, tannins, anthraquinones, carbohydrates, amino acid, alkaloids, steroids, flavonoids and absence of saponins in crude methanolic peel extract of pomegranate. Growther and Sukritha (2018) found the ethanol extract of bark of guava more active than methanol extract, minimum bactericidal concentration (MBC) of ethanol extract of bark being 16$gm.ml^{-1}$ with presence of active phytochemicals alkaloids, steroids, flavonoids, saponins, tannins. They concluded that the observed antimicrobial activity was due to the phytochemicals present in the bark samples of the plant.

Conclusion

Methanol extract of pomegranate stem bark exhibited a potent antimicrobial activity against *S.aureus*, *P. vulgaris* and MRSA while that of guava stem bark against the *P. vulgaris*. From this study, it can be concluded that stem barks of pomegranate and guava contain important phytochemicals which are responsible for antimicrobial activity. Identification and isolation of such active phytochemicals from extract play crucial role in the development of new biologically active products.

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References

Abi Beaulah, G., Mohamed Sadiq, A., & Jaya Santhi, R.(2011). Antioxidant and antimicrobial activity of *Achyranthes aspera*: an *in vitro* study. *Annualsof Biological Research*,*2*(5), 662-670.

Acharya, R. (2012). Ethonobotanical study of medicinal plants of Resunga Hill used by Magar community of Badagaun VDC, Gulmi district, Nepal. *Scientific World*, *10*(10), 54-65.

Bonjar, G.H.S., Nik, A.K.,& Aghighi S. (2004). Antibacterial and antifungal survey in plants used in indigenous herbal-medicine of south east regions of Iran. *J Biol Sci*,*4*, 405-412.

Cheesbrough, M. (2000). *District Laboratory Practices in Tropical Countries* (Part 2, pp 124 130). Cambridge, UK: CambridgeUniversity Press.

Ciulei, I. (1982). *Methodology for Analysis of Vegetable Drugs: Practical Manual on the Industrial Utilisation of Medicinal and Aromatic Plants* (pp 1-62). Bucharest, Romania: Author.

Collee, J.G., Duguid, J.P., Fraser, A.G., Marmion, B.P., & Simmonas, A. (1996). *Laboratory Strategy in the Diagnosis of Infective Syndrome*. In J.G. Collee, B.P. Marmion BP, A.G. Fraser and A. Simmons (Eds),*Mackie and McCartney Practical Medicinal Microbiology* (14th ed., pp 245-413). New York: Churchill Livingstone.

Department of Plant Resources. (2016). *Medicinal Plants of Nepal* (2nd ed.). Kathmandu, Nepal: Author.

Eloff, J. N.(1998). Which extract should be used for the screening and isolation of antimicrobial components from plants? *Journal of Ethonopharmacology, 60*, 1-8.

Forbes, B., Sahm, D., & Weissfeld, A. (2007). *Bailey & Scott's Diagnostic Microbiology* (12thed.). Missouri, USA: Mosby Elseviver.

Gurmachhan, C.M., Tandukar, U., Shrestha, N., Lakhey, P.B., & Pokhrel, C.P. (2019). Antimicrobial and phytochemical studies of bark extract of *Berberis asiatica* Roxb. ex DC. and *Myrica esculenta* Buch.-Ham ex D. Don.*Journal of Plant Resources*,*17*, 139-146.

Gurmachhan, C. M., (2018), Antimicrobial and Phytochemical Studies of Bark of Some Medicinal Plants. M. Sc. Thesis, Central Department of Botany, T.U., Kirtipur.

Growther, L., & Sukirtha, K. (2018). Phytochemical analysis and antimicrobial properties of *Psidium gaujava* leaves and bark extract. *Asian Journal of Pharmacy and Pharmacology*, *4*(3), 318-323.

Harborne, J.B. (1998). *Phytochemical Methods*: *A Guide to Modern Techniques of Plant Analysis* (3rd ed.). London, U.K.: Chapman & Hall.

Hamid, K., Nora, N. B., Abderaouf, B., Boumediane, M., & Abdella, M. (2015). Phytochemical and antimicrobial screening of *Punica granatum* L. bark of south-west Algeria. *Journal of Chemical and Pharmceutical Research*, *7*(8), 880-884.

Kuber, B.R., Lakhshmi, M.R., Deepika, E., & Yamuna, P. (2013). Phytochemical screening, *invitro*antibacterial and antioxidant activity of *Psidium gaujava* root bark. *Int. J. Curr. Microbiol. App. Sci.*, *2*(10), 238-248.

Kundal, J., Purohit, R., Singh, R., & Purohit, M.C. (2013). Antimicrobial and antioxidant activities of bark extract of *Myrica esculenta*. *Journal of Applicable Chemistry*, *2*(5), 1141-1146.

Malla, B., & Chhetri, R.B. (2009). Indigenous knowledge on ethnobotanical plants of Kavrepalanchok district. *Kathmandu University Journal of Science, Engineering and Technology*, *5*(II), 96-109.

Nascimento, G.G.F., Locatelli, J., Freitas, P.C., & Silva, G.L. (2000). Antibacterial activity of plant extracts and photochemical on antibiotic resistant bacteria. *Brazillian Journal of Microbiology*, *31*(4), 247-256.

Parajuli, D.P., Gyawali, A.R., & Shrestha, B.M. (1998). *A Manual of the Important Non-timber Forest Products in Nepal*. Pokhara, Nepal: Training and Manpower Development in C.F.M..

Perez, C., Pauli, M., & Bazerque, P. (1990). An antibiotic assay by agar-well diffusion method. *Acta biologiaeet medecine experimentalis*, *15*, 113-115.

Rana, M., Lakhey, P.B., Bhatta, T.D., Khadgi, S., Paudel, K., Adhikari, A.K., Bhattrai, M.R.,& Upadhay, U. (2017). GCMS Quantitative analysis and antimicrobial activity of essential oils of *Cinnamomum tamala* (buch.-Ham.). Nees and Eberm. (Tejpat) leaves collected from different parts of Makawanpur district, Nepal. *Journal of Plant Resources*, *15*(1), 73-80

Rokaya, M.B., Uprety, Y., Poudel, R., &Timsina, B., (2014). Traditional uses of medicinal plants in gastrointestinal disorders in Nepal. *Journal of Ethnopharmacology*, *158*, 221–229.

Sajjad, W., Sohail, M., Ali, B., Haq, A., Din, G. U., Hayat, M., Khan, I., Ahmad, M.,& Khan, S. (2015). Antibacterial activity of *Punica granatum* Peel extract. *Mycopath*,*13*(2), 105-111.

Shihabudeen, N.S., Priscilla, H., & Thirumuragan, K. (2010). Antimicrobial activity and phytochemical analysis of selected Indian folk medicinal plants. *International Journal of Pharma Sciences and Research*, *1*(10), 430-434.

Thapa, S. (2012). Medico-ethnobotany of Magar community in Saliya VDC of Parbat District, Central Nepal. *Our Nature*, *10*, 176-196.

Tiwari, P., Kumar, B., Kaur, M., Kaur, G., & Kaur, H. (2011). Phytochemical screening and extraction: a review. *Internationale pharmaceutica sciencia*, *1*(1), 98-106.

World Health Organization. (1991). *Basic Laboratory Procedure in Clinical Bacteriology*. Geneva, Switzerland: Author.

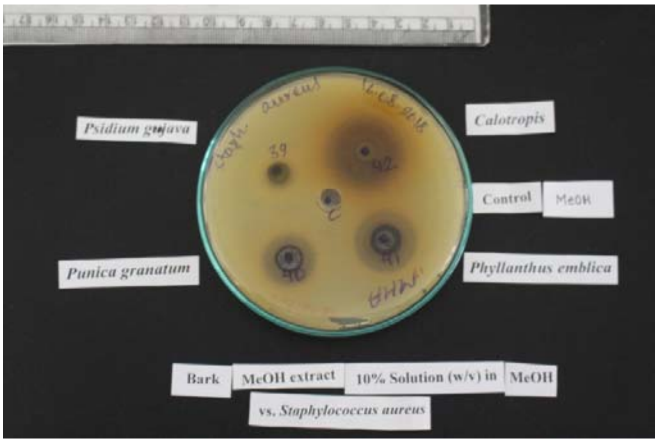

Plate 1: ZOIs of methanol extracts of *Psidium guajava* L. and *Punica granatum* L. barks against *Staphylococcus aureus* Rosenbach

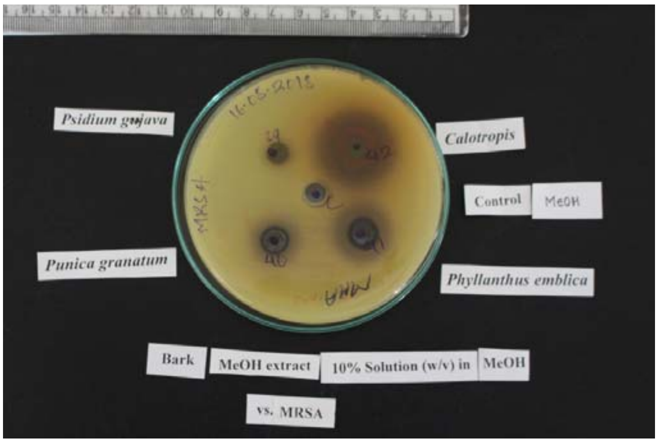

Plate 2: ZOIs of methanol extracts of *Psidium guajava* L. and *Punica granatum* L. barks against against MRSA

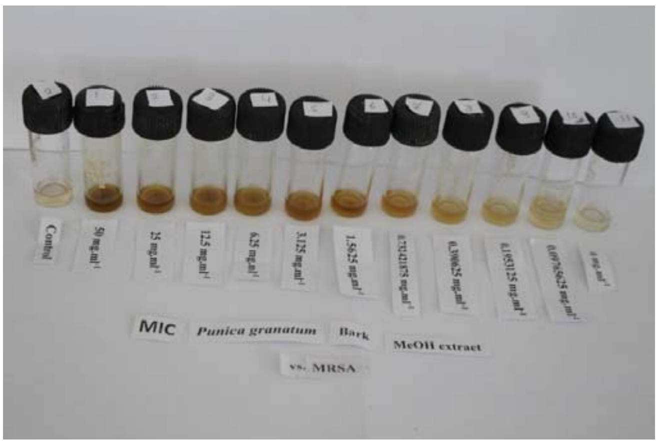

Plate 3: Determination of minimum inhibitory concentration (MIC) of methanolic bark extract of *Punica granatum* L.against MRSA

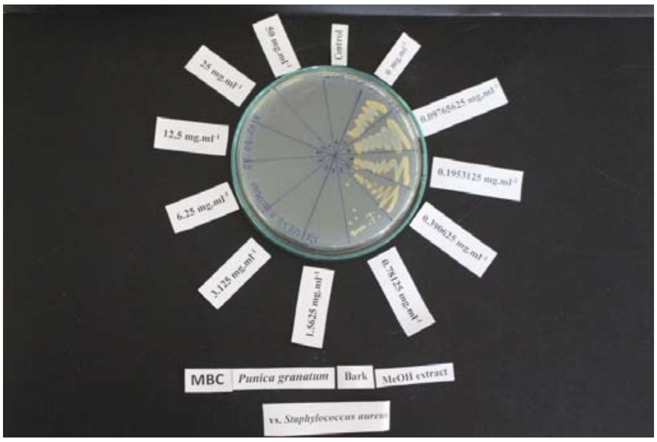

Plate 4: Determination of MBC of methanolic bark extract of *Punica granatum* L. against *Staphylococcus aureus* Rosenbach

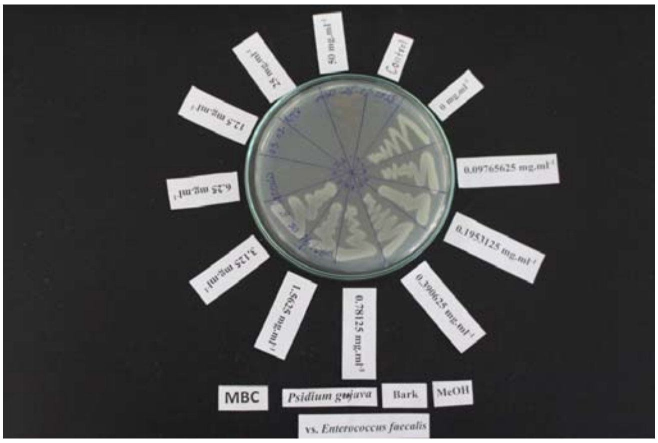

Plate 5: Determination of MBC of methanolic bark extract of *Psidiumgaujava* against *Enterococcus faecalis* (Andrewes & Horder) Schleifer & Kilpper-Balz

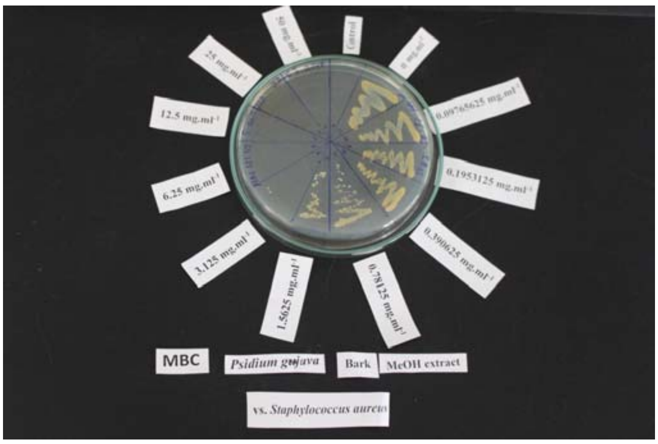

Plate 6: Determination of methanolic bark extract of *Psidium gaujava* L. against *Staphylococcus aureus* Rosenbach

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Antibacterial Activity of Lemongrass on Gram Positive and Gram Negative Bacteria of Human Pathogens

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Abstract

All leaves were thoroughly washed with sterile distilled water and dried in shade until all moisture evaporated. About 200 gram of leaves were grinded with 500 ml water for aqueous juices extraction for triplicate times. Similarly, essential oil was isolated triplicate times from leaves by 3 hour hydrodistillation of 200 gram of leaves in 500 ml of water. The essential oil and aqueous juices extraction was separated and residues obtained were mixed with equal volume of methanol for obtaining methanol extraction of juices and oil. Then, juices extraction and oil extraction were mixed diluted to 0.5%, 1%, 2.5%, 5%, and 10% with DMSO. Agar well diffusion method was done for studied antimicrobial activity for the pure bacterial cultures of *Escherichia coli, Staphylococcus aureus*, *klebsialla pneumonia*, *Salmonella* Typhi, *Pseudomonas aurogenosa* and for quality control *Staphylococcus aureus* (ATCC: 25923) and *Escherichia coli* (ATCC: 25922). A cork borer of 7 mm diameter selected for 70μl pouring in each well of different concentration along positive control (oflaxacin) and negative control (Dimethyl sulfoxide) then incubated at 37°C in an incubator for 24 hrs to 48 hrs in aerobic condition. After incubation, clear zone of inhibition were observed around the wells. Oil extraction of lemon grass was found to be highly effective for gram positive bacteria *S. Aureus* (up to 42 mm- Methanol) and gram negative bacteria *S. Typhi* (upto 36 mm methanol). It was concluded that oil extraction of leaves of lemongrass were found to be highly effective for treatment of human pathogens.

Keywords: Antimicrobial activity,*Cymbopogon citratus*, Extraction, Well diffusion method

Introduction

Plant essential oils and extracts have been used for many thousands of years, in food preservation, pharmaceuticals, alternative medicine and natural therapies. It is continuously used as traditional treatment (Burt, 2004). Among different medicinal plants, *Cymbopogon citratus* is one which is commonly known as Lemongrass. *Cymbopogon citratus* (Lemongrass) belongs to Poaceae family which is a perennial plant with long, thin leaves one of the largely cultivated medicinal plants for its essential oils in parts of tropical and subtropical areas of Asia, Africa and America (Chanthal et al., 2012). Lemon is regarded as one of the grass which is commonly available in Nepal and abroad.

It is widely used in different conditions of pain and discomfort. The oil obtained from the grass has diverse medicinal value. It also produces semi-synthetic Vitamin A that reduces the risk of Xerophthalmia and Night blindness. The grass has great benefits to mankind as it revitalizes the body and mind, helps with infections and act as muscle and skin toner (Srivastava et al., 2013). The leaves of lemongrass present lemony characteristic flavor due to its main content, citral which present great importance to the industry. There were a number of studies carried out to prove the anti-oxidant, anti-microbial and anti-fungal activities of lemongrass (Nikos & Costas, 2007; Oloyede et al., 2010). It is commonly used in folk medicine for treatment of nervous and gastrointestinal disturbances. However, it is also used as antispasmodic, analgesic, anti-inflammatory, anti-pyretic, diuretic and sedative (Dubey et al., 2011).

Essential oils are a concentrated hydrophobic liquids containing volatile chemical compounds from plants materials and do not only originate from flowers, but from herbs, trees and various other plant material. It is estimated that the global number of plants is of the order of 300,000 and about 10% of these contains essential oils and could be used as a source for their production (Husnu & Gerhard, 2010). The common methods to extract essential oil from medicinal plant, including for lemongrass *(Cymbopogon citratus)*, are hydrodistillation (HD), steam distillation, steam and water distillation, maceration, empyreumatic (or destructive) distillation and expression (Ashgari et al., 2010).

Lemongrass has phytoconstituents such as tannins, flavanoids, alkaloids and various essential oils in this herb. Secondary active metabolites of a number of components have also been implicated in the varied pharmacological effects of this plant. Lemongrass possesses various antimicrobial properties. The extracts of lemongrass leaves (dried) with cold, hot and different solvents like ethanol and methanol were screened for its antimicrobial activity against various bacteria like *Bacillus vallismortis, Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa, Vibrio cholerae*. Lemongrass extracts possess a great antimicrobial activity against the antibiotic resistant microorganisms (Isam et al., 2009).

Due to the development of resistance in pathogenic microorganisms to antibiotics used in modern medical science, there is a growing attention towards plant extracts as a source of new antimicrobial drug discoveries. As such investigations on the composition, activity, as well as validation of the use of extracts obtained from medicinal plant is important. The emergence of drug resistance in human and animal pathogenic bacteria, as well as undesirable side effects of certain antibiotics, has triggered immense interest in the search for new antimicrobial alternatives of plant origin. The purpose of this study is to identify the antibacterial activity of the different extract of leaves against the growth of bacteria.

Materials and Methods

Plant material collection

For the study, only leaf extract was used. So, the leaves of the mature plants of lemongrass were collected aseptically from ground of Tribhuvan University, Kirtipur, in clean plastic bag and transported to Laboratory. Then further extraction procedure followed according to Ezekwesili et al. (2004) and Mishra & Mishra (2011).

Preparation of leaf extract

All leaves were thoroughly washed with tap water and then rinsed with sterile distilled water. Then all leaves were dried in shade until all moisture evaporated.

Aqueous extraction of leaves (Juices extraction)

About 200 grams of leaves were ground with 500 ml water and kept 8 hrs at ambient temperature. Then whole mixture was filtered using a cheese cloth and obtained extracts was centrifuged. After centrifugation, supernatant was labeled it and used for antibacterial activity assay. Residue obtained from filtration was used for methanol extraction.

Extraction of essential oil

Essential oil was isolated from leaves by 3 hrs hydrodistillation of 200 grams of leaves which were placed in 500 ml of water. The essential oil was separated, dried over anhydrous sodium sulphate and stored at -20°C until used for other tests.

Alcoholic extraction

Both types of residues obtained after aqueous extraction of juices and essential oil extraction both were also separately treated with equal volume of methanol and kept for 24 hrs at ambient conditions. Then the mixtures were filtered using cheese cloth and extracts were obtained. In half pure extraction (crude) of lemongrass of juices and oil, no dilution were done and also used for study antimicrobial activity.

Dilution of juices and oil extraction

The juices extraction and oil extraction were diluted to 0.5%, 1%, 2.5%, 5%, and 10% with dimethyl sulphoxide (DMSO). Similarly, methanol extraction were diluted by DMSO as similar percentage.

Preparation of microbial cultures

Pure bacterial cultures were used for the study and maintained on nutrient agar medium by subculturing of *Escherichia coli*, *Staphylococcus* species, *Klebsialla pneumonia*, *Salmonella typhi* and *Pseudomonas aurogenosa*. ATCC culture of bacteria with *Staphylococcus aureus* (ATCC: 25923) and *Escherichia coli* (ATCC: 25922) were used for quality control for study of antibacterial activity of bacteria. All bacteria were obtained from Med-Micro Research Laboratory, Kathmandu, Nepal.

Antimicrobial activity

Agar well diffusion method was used to determine the antimicrobial activity of leaves extract in vitro. This method was done triplet times and average mean was taken as accurate mean for zone of inhibition of plants extracts. The pure colonies (3-4 colonies) of bacteria were transferred to nutrient broth and incubated for 4 hrs. then their turbidity was compare with 0.5 McFarland turbidity standards. After adjusting the inoculums, a sterile cotton swab was dipped into the inoculums and rotated against the wall of the tube above the liquid to remove excess inoculums. Then the swab was lawn culture on MHA plate by the entire surface rotated by approximately 60º between streaks to ensure even distribution.

The inoculated plate was allowed to stand for at least 3 minutes but no longer than 15 min, before making wells for different compounds to be tested. A hollow tube or cork borer of 7 mm diameter was sterile and press above the inoculated agar plates. It was removed immediately by making a well in the plate; likewise, other wells on each plate were made, one each for positive control (oflaxacin), negative control (DMSO) and for five respective concentrations (0.5%, 1%, 2.5%, 5%, and 10%).

Micropipettes were used to place 70µl of the solution of respective dilution in each well. Plates were incubated at 37ºC in an incubator for 24 hrs to 48 hrs in aerobic condition. After incubation clear zone of inhibition were observed around the wells. Diameters of those inhibition zones were measured and compare it with ATCC bacteria with *Staphylococcus aureus* (ATCC: 25923) and *Escherichia coli* (ATCC: 25922).

Results and Discussion

In this study, the antibacterial activity of lemongrass extracts was determined against five human pathogenic bacteria by the well diffusion method. Aqueous (juices) and methanol extracts of leaves were test for antibacterial activity. The different human pathogenic bacteria (5) were *S. aureus*, *E. coli*, *K. pneumoniae*, *S.* Typhi and *P. aeruginosa*. On the other hand, ATCC culture of *E. coli* (ATCC: 25922) and *S. aureus* (ATCC: 25923) were used for quality control. The well containing ofloxacin antibiotic was positive control which showed inhibiting activity for all the five bacteria. On the other hand, the blank well containing DMSO as negative control which did not show any inhibiting activity for all bacteria included for antibacterial activity.

From the present study it is clear that lemongrass juices and oil possess anantibacterial activity against gram positive and gram negative bacteria. In juices extraction, direct lemon grass juices of low dilution (0.5%) only showed antibacterial activity for ATCC gram positive bacteria *S. aureus* (ATCC: 25923). The effective of lower dilution of extraction is due to easily diffusion of lower concentration of extraction in media for antibacterial activity. On the other hand, methanol extraction of lemon grass juices of all dilution showed antibacterial activity for ATCC gram positive bacteria *S. aureus* (ATCC: 25923). However, result obtained by Nyamath & Karthikeyan (2018) in which cold water extraction (juice) of lemongrass was found to be effective than hot water, methanol and ethanol extraction which result is interesting in the traditional method of treating a bacterial infection.The results obtained two of these methods differ due to many factors between assays (Janssen et al., 1987; Hili et al., 1997). However, direct lemongrass juices and methanol juices extraction did not show antibacterial activity for human pathogenic bacteria. The results obtained support the general indication that gram positive organisms are more sensitive to lemon grass juices than gram negative bacteria.

The effective antibacterial activity was found in oil extraction of lemongrass leaves than juices extraction. Among different dilution of oil extraction of lemongrass leaves, both pure extraction and all dilution of extraction showed the highest zone of inhibition on gram positive than gram negative bacteria. So, gram positive bacteria were found to be more sensitive to the oil extraction than gram negative bacteria except *P. aeruginosa* and *K. Pneumoniae*. Similar observations were made by Onawunmi & Ongulana (1986) and Cimanga et al., (2002), *P. aeruginosa* and *K. Pneumoniae* were found resistant at all the concentration of lemongrass oil including pure (crude). Similar results were reported by Onawunmi et al. (1984), Alam et al. (1994), Marta et al. (2004), Pereira et al. (2004) and Naik et al. (2010), *P. aeruginosa* was found to be resistant to all dilution of oil extraction of lemongrass. This might be due to the multidrug resistant *P. aeruginosa* and *K. Pneumoniae* which was found to be non-effective with juices and oil extracts of all dilution of lemongrass.

Among two methods of extraction, methanol extraction was found to be more effective than aqueous oil extraction of lemon grass. The highest zone of inhibition was observed in *S. aureus* (ATCC: 25923) and *S. aureus* from methanol oil extraction than aqueous oil extraction of lemongrass. Among gram negative bacteria, antibiotic resistant bacteria like *S.* Typhi, showed highest antibacterial activity than *E. coli.* The results obtained by each of these methods differ due to many like microbial growth, exposure of microorganisms to the oil, the solubility of oil or oil components and the use and quality of an emulsifier etc. mentioned by Naik et al. (2010).

According to Isam et al. (2009) showed lemongrass extracts possess a great antimicrobial activity against the antibiotic resistant microorganisms. Lovet et al. (2010) mentioned that the presence of phytochemical and bioactive ingredients in lemon grass have been attributed to its antimicrobial potentials. So, it is widely used in different conditions of pain and discomfort (Srivastava et al., 2013). The lemongrass oil obtained from the grass has diverse medicinal value. The leaves are used in the treatment of cough, fever, depression, nervous disorder and skin irritations. So, essential oil is one of the important components of lemongrass extracts and its applications will be helpful in treating different infections.

Table 1: Antibacterial activity of juices of aqueous and methanol extraction of lemongrass leaves

Extraction	Juices extract zone of inhibition (mm) with different dilution						
	Bacteria	**0.5%**	**1%**	**2.5%**	**5%**	**10%**	**Pure juices**
Aqueous extraction	*S. aureus*(ATCC: 25923)	13	0	0	0	0	0
	S. aureus	0	0	0	0	0	0
	E. coli (ATCC: 25922)	0	0	0	0	0	0
	E. coli	0	0	0	0	0	0
	K. pneumoniae	0	0	0	0	0	0
	S. Typhi	0	0	0	0	0	0
	P. aeroginosa	0	0	0	0	0	0
Methanol extraction	*S. aureus*(ATCC: 25923)	8	9	11	12	14	0
	S. aureus	0	0	0	0	0	0
	E. coli (ATCC: 25922)	0	0	0	0	0	0
	E. coli	0	0	0	0	0	0
	K. pneumoniae	0	0	0	0	0	0
	S. Typhi	0	0	0	0	0	0
	P. aeroginosa	0	0	0	0	0	0

Table 2: Antibacterial activity of oil of aqueous and methanol extraction of lemongrass leaves

Extraction	Bacteria	Essential oil extracts zone of inhibition (mm) with different dilution 0.5%	1%	2.5%	5%	10%	Pure oil
	S. aureus(ATCC*:* 25923)	8	10	28	30	36	20
	S. aureus	10	15	20	35	42	30
	E. coli (ATCC: 25922)	0	0	0	0	0	0
	E. coli	0	0	0	0	0	0
	K. pneumoniae	0	0	0	0	0	0
Aqueous extraction	*S.* Typhi	12	15	20	36	38	24
	P. aeroginosa	0	0	0	0	0	0
	S. aureus(ATCC*:* 25923)	30	32	30	40	42	30
	S. aureus	20	24	26	36	40	25
	E. coli (ATCC: 25922)	8	8	10	10	11	0
	E. coli	8	0	0	0	0	0
	K. pneumoniae	0	0	0	0	0	0
Methanol extraction	*S.* Typhi	18	20	25	30	36	38
	P. aeroginosa	0	0	0	0	0	0

Plate 1: Hydrodistillation of 200 gram of lemongrass leaves in 500 ml of water

Plate 2: Extraction of oil of lemongrass

Plate3: Antimicrobial activity of *Staplylococcus aureus* (ATCC: 25923) on lemongrass with pure (crude), 10%, 5%, 2.5%, 1% and 0.5% dilution

Conclusion

From this research work, it can be concluded that oil extraction of Lemongrass was found to be highly effective for gram positive bacteria *S. aureus* and gram negative bacteria *S.* Typhi. So, oil extraction of lemongrass showed antibacterial activity for human pathogens and suggested that use of Lemongrass oil would be helpful in the treatment of infections caused by different bacteria.

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References

Alam, K., Agua, T., Maven, H., Taie R., Rao, K.S & Burrows, I., et al. (1994). Preliminary screening of seaweeds, sea grass and lemongrass oil from Papua New Guinea for antimicrobial and antifungal activity. *Inter J Pharmacognosy*, *32*(4), 396-399.

Ashgari, J.C. Touli, J. & Mazaheritehrani, M. (2010). "Microwave-assisted hydrodistillation of essential oils from *Echinophoraplatyloba DC." Journal of Medicinal Plants Research, 6*(28), *4475-4480.*

Burt, S. (2004). Essential oils: their antibacterial properties and potential applications in foods - a review. *International journal of food microbiology*, 94(3), 223-253.

Chanthal, S. Prachakoli, S. & Ruangviriyachai, C. (2012). "Influence of extraction methodologies on the analysis of five major volatile aromatic compounds of citronella grass and lemongrass grown in Thailand. *Journal of AOAC International*, 95, 763-772.

Cimanga, K., Tona, L. & Apers, S., et al. (2002). Correlation between chemical composition and antibacterial activity of essential oils of some aromatic medicinal plants growing in the Democratic Republic of Congo. *J Ethanopharmacol*, 79(2), 213-220.

Dubey, R.Dubey, K. Sridhar, C. & Jayaveera, K.N. (2011). Human Vaginal Pathogen Inhibition Studies on Aqueous, Methanolic and Saponins Extracts of Stem Barks of *Ziziphusmauritiana*. *International Journal of Pharmaceutical Sciences and Research, 2*(3), 659-663.

Ezekwesili, C.N., Obiora, K.A. & Ugwu, O.P. (2004). Evaluation of anti-diarrhoeal property of crude aqueous extract of *Ocimumgratissium* L. (Laniatae) in rats. *Biokemistri*, *16*(2), 122-132.

Hili P, Evans CS & Veniss RG. (1997). Antimicrobial action of essential oils: the effect of Dimethylesulphaoxide on the activity of Cinnamon oil. Let Appl Microbiol, *24*, 269-75.

Husnu, K.C.B. & Gerhard, B. (2010). *Handbook of Essential Oils: Science, Technology, and Applications*, UK: Taylor and Francis Group.

Isam, S. Sundus, H. & Hussain, A. (2009). Study the antimicrobial activity of Lemon grass leaf extracts: Ministry of science& Technology.

Janssen AM, Scheffer JJC & Baerheim SA (1987). Antimicrobial activity of essential oils: a 1976-1986 literature review. Aspects of the test methods. *Plt Med*, *53*, 395-8.

Lovet T. K., Uhunmwangho E.J., & Sylvester C. I., (2018). Assessment of Hot Water and Ethanolic Leaf Extracts of *Cymbopogon Citrates* Stapf (Lemon Grass) against Selected Bacteria Pathogens, *Annals of Microbiology and Infectious Diseases*, *1*(3), 04-08.

Marta War, O.N, Majra, R. R. J., Gaston, G S. & Celia, L.R. (2004). Antimicrobial activity of the essential oil and cream of Cymbopogoncitratus (DC.) stapf. Revcubana. *Plt Med*, *2*, 44-47.

Mishra, P. & Mishra, S. (2011). Study of antibacterial activity of *Ocimum Sanctum* extract against gram positive and gram negative, *Am. J. Food Technol*. *6*(4), 336-336.

Naik, M.I., Fomba, B.A., Jayakumar, E. & Bhat, J.A. (2010). Antibacterial activity of lemongrass (Cymbopogon citrates) oil against some selected pathogenic bacteria. *Asian Pacific Journal of Tropical Medicine, 3*,535-538.

Nikos, G.T. & Costas, D.E. (2007). Antifungal activity of Lemongrass (*Cympopogoncitratus L.*) essential oil against key postharvest pathogens. *Innovative Food Science Emerging Technology*, *8*, 253-255.

Nyamath, S. & Karthikeyan B., (2018). *In vitro* antibacterial activity of Lemongrass (*Cymbopogoncitratus*) leaves extract by agar well method. *Journal of Pharmacognosy and Phytochemistry, 7*(3), 1185-1188.

Oloyede, G. K., u, I.A. & Shodia, A.F. (2010). Chemical Composition and Cytotoxicity of the essential oils of Crinum ornatum (Ait) Bury. *African Journal of Pure and Applied Chemistry*, *4*(3), 35-37.

Onawunmi, G.O. & Ogunlana, E.O. (1986). A study of the antibacterial activity of the essential oil of Lemongrass (*Cymbopogon citrates*). *Inter J Crud Drug Res, 24*(2), 64-68.

Onawunmi, G.O., Yesiak, W.A.B., & Ongulana, E.O (1984). Antibacterial constituent in the essential oil of *Cymbopogoncitratus*. *J Ethanopharmocol,* 12(3), 279-286.

Pereira, R.S., Sumita, T.C. & Furlan, M.R., et al (2004). Antibacterial activity of essential oils on microorganisms isolated from urinary tract infections. *Revista de SaudePublica, 38*(2), 326-328.

Srivastava,V., Dubey, S. & Mishra, A. (2013). A review of lemongrass; Agricultural and Medicinal aspect. *International Research Journal of Pharmacy*, *4*(8), 42-44.

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Asymbiotic Seed Germination and Seedling Development of a Medicinally Important Epiphytic Orchid, *Dendrobium crepidatum* Lindl. & Paxton

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Abstract

Dendrobium crepidatum Lindl. & Paxton is an important medicinal orchid with high alkaloid content having pharmacological activities. The species has been threatened due to deforestation and overexploitation therefore, a conservation strategy was needed. Asymbiotic seed germination has been used for conservation and commercial production of different orchids. The current study evaluated the effects of capsule maturity and media composition for asymbiotic seed germination and seedling development using capsules collected 118 and 148 days after pollination (DAP) of *D. crepidatum* flowers. The seed germination was highest for half strength Murashige and Skoog (MS) medium in 70 days of culture with 41±0.76% protocorm development for late capsule group (148 DAP capsules) and 36.33±0.96% for early capsule group (118 DAP capsules). Also, 148 DAP capsules showed early germination in terms of all three seed germination stages in comparison to 118 DAP capsules. The seedling development was studied across 12 different combinations of half strength MS media, where half strength MS medium supplemented with 2 ìgmL^{-1} 6-benzylaminopurine (BAP) and 1 ìgmL^{-1} 1 naphthaleneacetic acid (NAA) showed highest growth on measuring leaf number per shoot (3.25±0.47, $F_{11,36}$ = 7.075 with p < 0.05) over 100 days of sub-culture period. Thus, relatively more mature seeds collected from 148 DAP capsules were found to be more suitable for asymbiotic seed germination with the use of half strength MS medium. The culture method thus established may facilitate conservation and large-scale cultivation of this medicinal orchid.

Keywords: Capsule, Days after pollination (DAP), Murashige and Skoog Medium, Protocorm

Introduction

Dendrobium crepidatum Lindl. & Paxton, is an epiphytic or lithophytic orchid species, flourishes in southern China, northern Indochina (Laos, Myanmar, Thailand, Vietnam) and the eastern Himalaya (Assam, Arunachal Pradesh, Sikkim, Bangladesh, Nepal) regions (Chowdhery, 2001; Raskoti, 2009; Rokaya et al., 2013). It possesses characteristic green, terete and pendulous stem which bear lanceolate leaf and white pinkish flower. *D. crepidatum* is being used in traditional Chinese pharmacopoeias (Bao et al., 2001). The alkaloids reported from the plant possess anti-inflammatory properties (Hu et al., 2016) and promote nerve growth in cell lines (Li et al., 2013). The species however, is threatened due to deforestation and overexploitation in Nepal (Raskoti, 2009). To avoid extinction and tapping of the possible pharmaceutical resources, the *D. crepidatum* species needs a conservation strategy.

An *ex situ* conservation approach involving *in vitro* techniques like seed germination, mass propagation, cryopreservation etc. have been used for conservation of endangered orchids (Decruse et al., 2003; Mohanty et al., 2012a; Bhattacharya et al., 2017). Among these, seed germination on in vitro culture system could be investigated as a conservation strategy and tool for mass production of *D. crepidatum*. Seeds are much more desirable than other tissues for in vitro culture, as plant development by seed propagation promotes inherent genetic variation ensuring survival on habitat restoration in a variety of environmental conditions (McCargo, 1998). In nature, orchid species relies on symbiotic fungus for germination (Mitchell, 1989). However, almost all species can be grown asymbiotically in presence of suitable nutrient medium conditions (Manning & van Staden, 1987). An efficient propagation technique for orchids has been developed using orchid seeds asymbiotically

(Arditti & Ernst, 1993) which has been utilized for production of different commercial and endangered orchids (Kauth et al., 2006; Stewart & Kane; 2006, Sgarbi et al.; 2009, Mohanty et al., 2012b).

The asymbiotic seed germination methods developed for many different orchid species vary in their composition and are largely species-specific (Arditti & Ernst, 1993; Zeng et al., 2013). Success of asymbiotic germination of orchid seeds depends upon maturity of seed capsule, physical germination conditions and the growth media constituents (Arditti, 1967; Zeng et al., 2013). The effect of maturity of seed capsule has been measured on the basis of number of days after pollination (DAP) as an important criterion for successful in vitro asymbiotic seed germination in *Paphiopedilum* (Lee, 2007; Long et al., 2010). Exogenous auxins are considered non-essential for seed and seedling development of orchids (Tamanaha et al., 1979) while addition of 6-benzylaminopurine (BAP) improved seed germination and protocorm proliferation (David et al., 2010; Nongdam & Tikendra, 2014). Following study was performed to evaluate the effects of capsule maturity and culture media on seed germination of *Dendrobium crepidatum* and to assess in vitro growth of its seedlings across various media combinations.

Materials and Methods

Plant materials

Unripe green capsules of *Dendrobium crepidatum* were collected from the garden of Central Department of Botany, Tribhuvan University, Kirtipur, Nepal. Flowers of *D. crepidatum* were hand-pollinated in the last week of April, 2018. Capsules were collected from the pollinated flowers in two different batches: one at the end of August 2018 categorized as early capsule group (118 DAP capsules) and the other at the end of September 2018 categorized as late capsule group (148 DAP capsules).

Surface sterilization and seed inoculation

Collected capsules were washed for 30 min. under running tap water with the addition of 3 drops of Tween 20 solution. Surface sterilization was performed using 1% Sodium hypochlorite (NaOCl) solution for 7 min. inside a laminar flow cabinet, washed five times with sterile double distilled water and finally flame sterilized for few seconds after dipping in 70% ethanol for 5 minutes. The sterilized capsules were split open longitudinally with a sterile surgical blade and seeds were inoculated on MS (Murashige & Skoog, 1962) medium and MS medium supplemented with plant growth regulators (PGRs).

Culture medium and in vitro growth conditions

MS medium was prepared utilizing their component stock solutions that included macro and micronutrients, iron salts and vitamins, prepared in concentrated solutions and stored at 4 ºC before use. During preparation of MS medium, the stock solutions were added successively with thorough mixing in a conical flask for desired volume. Sucrose (3%) was added and a near medium volume was made with double distilled water then pH 5.8 was adjusted. MS medium was supplemented with PGRs, viz., 2,4 dichlorophenoxyacetic acid (2,4-D), 1 naphthaleneacetic acid (NAA) and 6 benzylaminopurine (BAP), individually and in combinations at different concentrations of 0.5 2.0 ìgmL^{-1}. Desired volume was then made by adding double distilled water and 0.8% agar (w/v) was added. Finally, the medium was autoclaved at 121ºC for 20 min. Cultures were maintained in a culture room at 25±2ºC under equivalent light-dark cycles provided by white fluorescent tubes.

In vitro growth parameters and data recording

Initially, seed germination based on protocorm development and seed germination stages was recorded for two capsule groups using MS medium of three strengths, viz., full, half and quarter, and MS medium supplemented with 2,4-D. A single capsule from each of the capsule groups was utilized for seed germination. Using a 0.5 cm diameter spatula, seeds were suspended in double distilled water in a sterilized test tube for an hour. The seeds were then separated from their suspension using Pasteur pipettes. The seed count was performed using a hand lens of 10X magnification in the Pasteur pipette. Finally in each MS medium, seeds were inoculated using Pasteur pipette while the double distilled water was removed as much as possible from the culture tube. Seed germination was examined by recording the protocorm development on different media compositions using the following formula:

$$\text{Protocorm development (\%)} = \frac{\text{Number of protocorms developed from inoculated seeds}}{\text{Total number of seeds inoculated}} \times 100$$

The process of seed germination was divided and recorded into three stages (seed swelling stage, green angular protocorm stage and leaf emergence stage), which were modifications of those given by Miyoshi & Mii (1995) for developmental stages of orchid embryos. These stages of seed germination were observed using a hand lens of 10X magnification and number of days for initiation of each stage was recorded across six replicates. Leaf emergence marked the initiation of seedling development.

Seedlings thus developed were transferred to the MS medium of the strength which was observed to best support the initial seed germination. Additionally, MS medium was further supplemented with NAA and BAP, individually and in combinations, and was used to establish seedling growth condition. Thus, a completely randomized experimental design with 12 different media as treatment factor over 4 replicates was performed. To assess the influence of various PGRs on in vitro seedling development, following growth parameters were measured: (1) Leaf number per shoot, (2) Leaf length, (3) Root number per shoot and (4) Root length.

Statistical analysis

In vitro seedling growth parameters viz., leaf number per shoot, leaf length, root number per shoot and root length were subjected to analysis of variance (ANOVA) with means compared by Duncan's multiple range tests (Duncan, 1955) using SPSS v25 program.

Results and Discussion

Selfing of four flowers of *Dendrobium crepidatum* resulted in 100% success of capsule formation indicating self-compatibility of the orchid species as suggested by Vasudevan & Van Staden (2010) in *D. nobile*. The major structural changes which occurred during in vitro culture of *D. crepidatum* seeds were studied with capsules collected in two groups. Initial seed swelling was observed due to absorption of water and nutrients from culture medium. The phenomenon was similar to that observed in all orchid seeds during in vitro germination prior to development of protocorm (Hossain et al., 2010). After 70 days of culture, protocorm development in initial seed germination medium was recorded. Comparatively higher percentage of protocorm development was observed

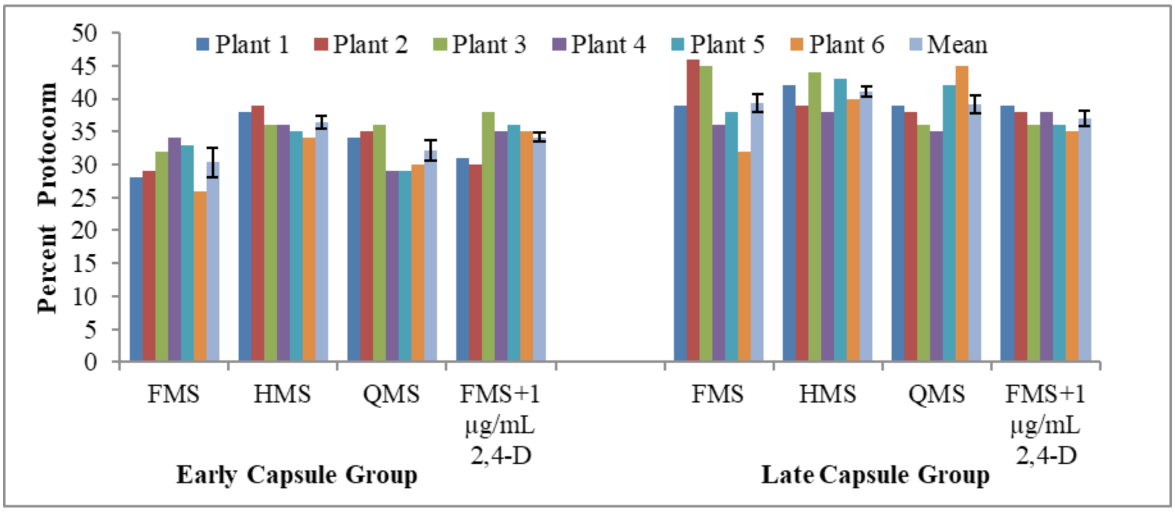

Figure 1: Effect of medium composition on protocorm development from seeds harvested from two different capsule groups of *Dendrobium crepidatum*.

in late capsule group than in early capsule group (Figure 1). This finding contradicts with the earlier observations for orchid seed germination as lower germination frequencies have been achieved by culturing mature seeds than immature seeds (Arditti et al., 1982; Rasmussen, 1995), since on maturity, integuments become impermeable to water (Kauth et al., 2008). However, to another view, sparse cuticular deposition in mature orchid seeds can make testa less hydrophobic resulting in greater germination of mature seeds (Hsu & Lee, 2012). Further, longer in vivo growth period after pollination may have resulted in greater histodifferentiation of developing embryo (Raghavan, 1997; Yeung, 2017) before in vitro culture making late capsule group more suitable for germination.

The half strength MS medium was found to be most suitable with 41±0.76% protocorm development for late capsule group and 36.33±0.96% for early capsule group. Henceforth, half strength MS medium was considered as an optimal medium for seedling development. The half strength MS medium has also been reported to show a significant seedling development in *Dendrobium transparens* (Sunitibala & Kishor, 2009) and in *D. chrysotoxum* (Kaur & Bhutani, 2011). Intervening callus formation was similar in all tested media causing decrease in protocorm development and differentiation. Cytokinin has been considered indispensable for orchid seed germination (Manning & van Staden, 1987), but for *D. crepidatum*, seeds were successfully germinated without supplementing any cytokinin which suggested presence of sufficient volume of endogenous cytokinin necessary for seed germination. Similar germination without PGR supplementation was reported in *Encyclia* aff. *Oncioides* (Znaniecka et al., 2005).

In this study, the effect of maturity of capsule on seed germination was by collecting the capsules at two different durations: early capsule group collected 118 days after pollination (DAP) and early capsule group collected 148 days after pollination. The capsules collected seeds of late capsule group showed early germination in terms of all three seed germination stages in comparison to those of early capsule group (Figure 2). The early seed germination from 148 DAP capsules may be due to appropriate age of the capsules for efficient protein mobilization during rehydration and embryonic unlignified testa allowing the permeability to nutrients (Long et al., 2010; Zeng et al., 2012). However, similar study of effect of maturity of capsule showed early seed germination from 90-120 DAP capsules in *Paphiopedilum godefroyae* (Lee, 2007) and 170-190 DAP capsules in *P. villosum var. densissimum* (Long et al., 2010). Hence, although capsule maturity affects seed germination, this effect seem to vary from orchid species to species as suggested by Deb & Pongener (2013). These germination responses, however, were slower with leaf emergence on 83.29±1.66 after inoculation for late capsule group and 89.87±1.92 days after inoculation for early capsule group, probably due to absence of PGRs in the medium as suggested by Roy et al. (2011) in *Vanda coerulea*, Decruse et al. (2013) in *Eulophia cullenii* and Nenekar et al. (2014) in *Eulophia nuda*.

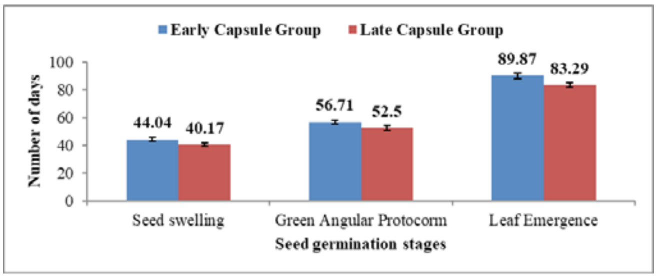

Figure 2: Changes recorded during seed germination from two different capsule groups of *Dendrobium crepidatum* over culture period.

Germinated seedlings attained maximum growth when sub-cultured on half strength MS medium supplemented with 2$\mu g.mL^{-1}$ BAP and 1$\mu g.mL^{-1}$ NAA (Table 1), the growth being measured on the basis of leaf number per shoot (3.25±0.47, $F_{11,36}$ = 7.075 with $p < 0.05$). This indicated a synergistic effect of auxin and cytokinin on leaf induction. Similar results of leaf development under the influence of cytokinin and auxin were observed in *Cattleya aurantiaca* (Melissa et al., 1994), *Vanda spathulata* (Decruse et al., 2003) and *Cymbidium aloifolium* (Deb & Pongener, 2011). The comparison of leaf numbers per shoot across all tested media using Duncan's multiple range test showed similar growths between half strength MS medium supplemented with 0.5µg.mL^{-1} NAA (2.5±0.28) or 1µg.mL^{-1} NAA (2.5±0.28) and half strength MS medium supplemented with 2µgm.L^{-1} BAP and 1µg.mL^{-1} NAA medium. All tested media supported seedling development during the study (Table 1) but the growth rate measured on the basis of leaf length, root number per shoot and root length did not validate homogeneity of variances of data.

Since symbiotic orchid seed germination revealed production of cytokinin by several mycorrhizal fungi (Crafts & Miller, 1974), the effects of addition of exogenous BAP on seed germination was studied. The seed germination rate decreased with the increasing concentration of BAP (Table 1) as there was high callus induction with increasing BAP concentration similar to the observations made by Roy et al. (2007), and by Nongdam & Tikendra (2014) in *Dendrobium chrysotoxum*. Among the tested media, the half strength MS medium supplemented with 2µg.mL^{-1} BAP and 1µg.mL^{-1} NAA showed maximum seedling growth measured as the leaf number per shoot.

Conclusion

In the current stuydy, asymbiotic seed germination and seedling growth of *Dendrobium crepidatum* was evaluated in terms of protocorm, leaf and root development. The seed germination competence was found to be influenced by simple nutritional requirements without using exogenous hormones. Earlier seed germination and greater protocorm development were recorded for relatively mature capsules collected 148 days after pollination. Further experiments are necessary to support the growth response of orchid seeds with the maturity of capsule. Use of half strength MS medium supplemented with 2µg.mL^{-1} BAP and 1µg.mL^{-1} NAA caused faster seedling development when measured on the basis of leaf number per shoot. The culture conditions thus established may facilitate conservation and large-scale cultivation of this medicinal orchid.

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Table 1: Effect of different PGRs on seedling development of *Dendrobium crepidatum* after 100 days of sub culture in half strength MS medium, where values represent mean±SE while '-' represent no response

NAA (µgmL^{-1})	BAP (µgmL^{-1})	Leaf number per shoot*	Leaf length in cm	Root number per shoot	Root length in cm
-	-	1.5±0.28^{abc}	0.89±0.18	-	-
0.5	-	2.5±0.28^{de}	1.27±0.09	0.25±0.25	0.162±0.16
1	-	2.5±0.28^{de}	1.46±0.08	1.00±0.41	0.727±0.24
1.5	-	1.75±0.25^{bcd}	1.15±0.02	1.00±0.41	0.627±0.22
2	-	2.00±0.00^{cd}	1.30±0.11	1.75±0.25	1.022±0.07
-	0.5	1.00±0.40^{ab}	0.577±0.20	1.25±0.25	0.952±0.15
-	1	1.25±0.25^{abc}	0.96±0.06	1.00±0	0.865±0.07
-	1.5	0.75±0.25^{a}	0.787±0.27	1.00±0.40	0.607±0.24
-	2	1.00±0.00^{ab}	0.535±0.04	-	-
1	1	2.00±0.40^{cd}	1.317±0.22	2.25±0.47	1.36±0.13
1	2	3.25±0.47^{e}	1.327±0.12	1.75±0.25	1.112±0.08
2	1	0.75±0.25^{a}	0.600±0.21	1.75±0.47	1.382±0.13

*The values followed by the same letter are not significantly different as determined by Duncan's Multiple Range Test ($p<0.05$)

References

Arditti, J. (1967). Factors affecting the germination of orchid seeds. *The Botanical Review*, *33*, 1–97.

Arditti, J., & Ernst, R. (1993). *Micropropagation of Orchids*. New York, USA: John Wiley & Sons Inc.

Arditti, J., Clements, M.A., Fast, G., Hadley, G., Nishimura, G., & Ernst, R. (1982). Orchid seed germination and seedling culture. A manual. *In*: J. Arditti (Eds.), *Orchid biology. Reviews and perspectives, II* (pp-243-370). Ithaca, New York, USA: Cornell University Press.

Bao, X.S., Shun, Q.S., & Chen, L.Z. (2001). *The Medicinal Plants of Dendrobium (Shi-Hu) in China, A Coloured Atlas*. Shanghai, China: Fudan University Press.

Bhattacharyya, P., Kumar, V., & van Staden, J. (2017). Assessment of genetic stability amongst micropropagated *Ansellia africana*, a vulnerable medicinal orchid species of Africa using SCoT markers. *South African Journal of Botany*, *108*, 294–302. doi: 10.1016/j.sajb.2016.11.007.

Chowdhery, H.J. (2001). Orchid diversity in North-East India. *Journal of the Orchid Society of India*, *15*, 1–17.

Crafts, C.B., & Miller, C.O. (1974). Detection and identification of cytokinins produced by mycorrhizal fungi. *Plant Physiology*, *54*, 586–588.

David, D., Gansau, J.A., & Abdullah, J.O. (2010). Effect of NAA and BAP on protocorm proliferation of Borneo Scented Orchid, *Vanda helvola*. *Asia-Pacific Journal of Molecular Biology and Biotechnology*, *18*, 221–224.

Deb, C.R., & Pongener, A. (2011). Asymbiotic seed germination and *in vitro* seedling development of *Cymbidium aloifolium* (L.) Sw.: A multipurpose orchid. *Journal of Plant Biochemistry and Biotechnology*, *20*(1), 90–95.

Deb, C.R., & Pongener, A. (2013). A study on the use of low cost substrata against agar for non-symbiotic seed culture of *Cymbidium iridioides* D. Don. *Australian Journal of Crop Science*, *7*, 642–649.

Decruse, S.W., Gangaprasad, A., Seeni, S., & Menon, V.S. (2003). A protocol for shoot multiplication from foliar meristem of *Vanda spathulata* (L.) Spreng. *Indian Journal of Experimental Biology*, *41*(8), 924–927.

Decruse, S.W., Reny, N., Shylajakumari, S., & Krishnan, P.N. (2013). *In vitro* propagation and field establishment of *Eulophia cullenii* (Wight) Bl., a critically endangered orchid of Western Ghats, India through culture of seeds and axenic seedling-derived rhizomes. *In Vitro Cellular & Developmental Biology - Plant*, *49*, 520–528.

Duncan, D.B. (1955). Multiple range and multiple 5Ø9Ü tests. *Biometrics: Journal of the Biometric Society*, *1*, 1–42.

Hossain, M.M., Sharma, M., Teixeira da Silva, J.A., & Pathak, P. (2010). Seed germination and tissue culture of *Cymbidium giganteum* Wall. ex Lindl. *Scientia Horticulturae*, *123*(4), 479–487.

Hsu, R.C.C., & Lee, Y.I. (2012). Seed development of *Cypripedium debile* Rchb. f. in relation to asymbiotic germination. *Horticultural Science*, *47*, 1495–1498.

Hu, Y., Zhang, C., Zhao, X., Wang, Y., Feng, D., Zhang, M., & Xie, H. (2016). (±)-Homocrepidine A, a pair of anti-inflammatory enantiomeric octahydroindolizine alkaloid dimers from *Dendrobium crepidatum*. *Journal of Natural Products*, *79*, 252–256.

Kaur, S., & Bhutani, K.K. (2011). *In vitro* propagation of *Dendrobium chrysotoxum* (Lindl.). *Floriculture and Ornamental Biotechnology*, *5*(1), 50-56.

Kauth, P.J, Vendrame, W.A., & Kane, M.E. (2006). *In vitro* seed culture and seedling development of *Calopogon tuberosus*. *Plant Cell, Tissue and Organ Culture*, *85*, 91.

Kauth, P., Dutra, D., & Johnson, T. (2008). Techniques and applications of *in vitro* orchid seed germination. *In*: J.A. Teixeira da Silva (Eds.), *Floriculture, Ornamental and Plant*

Biotechnology: Advances and Topical Issues (pp. 375–391). Iselworth, UK: Global Science Books.

Lee, Y.I. (2007). The asymbiotic seed germination of six *Paphiopedilum* species in relation to the time of seed collection and seed pretreatment. *International Society for Horticultural Science*, *755*, 381–386. doi: 10.17660/ActaHortic.2007.755.50.

Li, C.B., Wang, C., Fan, W.W., Dong, F.W., Xu, F.Q., Wan, Q.L., Luo, H.R., Liu, Y.Q., Hu, J.M., & Zhou, J. (2013). Chemical components of *Dendrobium crepidatum* and their neurite outgrowth enhancing activities. *Natural Products and Bioprospecting*, *3*, 70–73.

Long, B., Niemiera, A.X., Cheng, Z.Y., & Long, C.L. (2010). In vitro propagation of four threatened *Paphiopedilum* species (Orchidaceae). *Plant Cell Tissue and Organ Culture*, *101*, 151–162.

Manning, J.C., & van Staden, J. (1987). The development and mobilization of seed reserves in some African orchids. *Australian Journal of Botany*, *35*, 343-353.

McCargo, H. (1998). *Seed propagation of native wild flowers, marine organic farmers and gardenerassociation*. Retrieved from http://www.mofga.org/mofga/other/smofdec08.html.

Melissa, M., Sabapathi, D., & Smith, R.A. (1994). Influence of benzylaminipurine and 1-naphthaleacetic acid on multiplication and biomass production of *Cattleya aurantiaca* shoot explants. *Lindleyana*, *9*, 169–173.

Mitchell, R.B. (1989). Growing hardy orchids from seed at Kew. *The Plantsman*, *2*, 152-169.

Miyoshi, K., & Mii, M. (1995). Enhancement of seed germination and protocorm formation in *Calanthe discolor* (Orchidaceae) by NaOCl and polyphenol absorbent treatments. *Plant Tissue Culture Letters*, *12*(3), 267–272.

Mohanty, P., Das, M.C., Kumaria, S., & Tandon, P. (2012a). High-efficiency cryopreservation of the medicinal orchid *Dendrobium nobile* Lindl. *Plant Cell Tissue and Organ Culture*, *109*, 297–305. doi: 10.1007/s11240-011-0095-4.

Mohanty, P., Paul, S., Das, M.C., Kumaria, S., & Tandon, P. (2012b). A simple and efficient protocol for the mass propagation of *Cymbidium mastersii*: an ornamental orchid of Northeast India. *AoB Plants*, pls023. doi: 10.1093/aobpla/pls023.

Murashige, T., & Skoog, F. (1962). A revised medium for rapid growth and bioassays with tobacco tissue cuture. *Physiologia Plantarum*, *15*, 473-497. doi: 10.1111/j.1399-3054.1962.tb08052.x.

Nenekar, V., Shriram, V., Kumar, V., & Kishor, P.B.K. (2014). Asymbiotic in vitro seed germination and seed development of *Eulophia nuda* Lindl., An endangered medicinal orchid. *National Academy of Sciences, India Section B: Biological Sciences*, *84*(3), 837–846. Proceedings.

Nongdam, P., & Tikendra, L. (2014). Establishment of an efficient *in vitro* regeneration protocol for rapid and mass propagation of *Dendrobium chrysotoxum* Lindl. using seed culture. *The Scientific World Journal*, *2014*, 740150. doi: http://dx.doi.org/10.1155/2014/740150.

Raskoti, B.B. (2009). *The Orchids of Nepal*. Kathmandu , Nepal: Bhakta Bahadur Raskoti & Rita Ale.

Raghavan, V. (1997). *Molecular embryology of flowering plants*. Cambridge, UK: Cambridge University Press.

Rasmussen, H.N. (1995). *Terrestrial orchids from seed to mycotrophic plant*. New York, USA: Cambridge University Press.

Rokaya, M.B., Raskoti, B.B., Timsina, B., & Münzbergová, Z. (2013). An annotated checklist of the orchids of Nepal. *Nordic Journal of Botany*, *31*(5), 511-550.

Roy, J., Naha, S., Majumdar, M., & Banerjee, N. (2007). Direct and callus-mediated protocorm-like body induction from shoottips of *Dendrobium chrysotoxum* Lindl. (Orchidaceae). *Plant Cell, Tissue and Organ Culture*, *90*(1), 31–39.

Roy, A.R., Patel, R.S., Patel, V.V., Sajeev, S., & Deka, B.C. (2011). Asymbiotic seed germination, mass propagation and seedling development of *Vanda coerulea* Griff ex Lindl. (Blue Vanda): an in vitro protocol for an endangered orchid. *Scientia Horticulturae*, *128*(3), 325–331.

Sgarbi, E., Grimaudo, M., & Del Prete, C. (2009). In vitro asymbiotic germination and seedling development of *Limodorum abortivum* (Orchidaceae). *Plant Biosystems*, *143*, 114–119.

Stewart, S.L., & Kane, M.E. (2006). Asymbiotic seed germination and *in vitro* seedling development of *Habenaria macroceratitis* (Orchidaceae), a rare Florida terrestrial orchid. *Plant Cell, Tissue and Organ Culture*, *86*, 147.

Sunitibala, H., & Kishor, K. (2009). Micropropagation of *Dendrobium transparens* L. from axenic pseudobulb segments. *Indian Journal of Biotechnology*, *8*, 448-452.

Tamanaha, L.R., Shimizu, C.G., & Arditti, J. (1979). The effects of ethephon on *Cattleya aurantiaca* (Orchidaceae) seedlings. *Botanical Gazette*, *140*, 25–28.

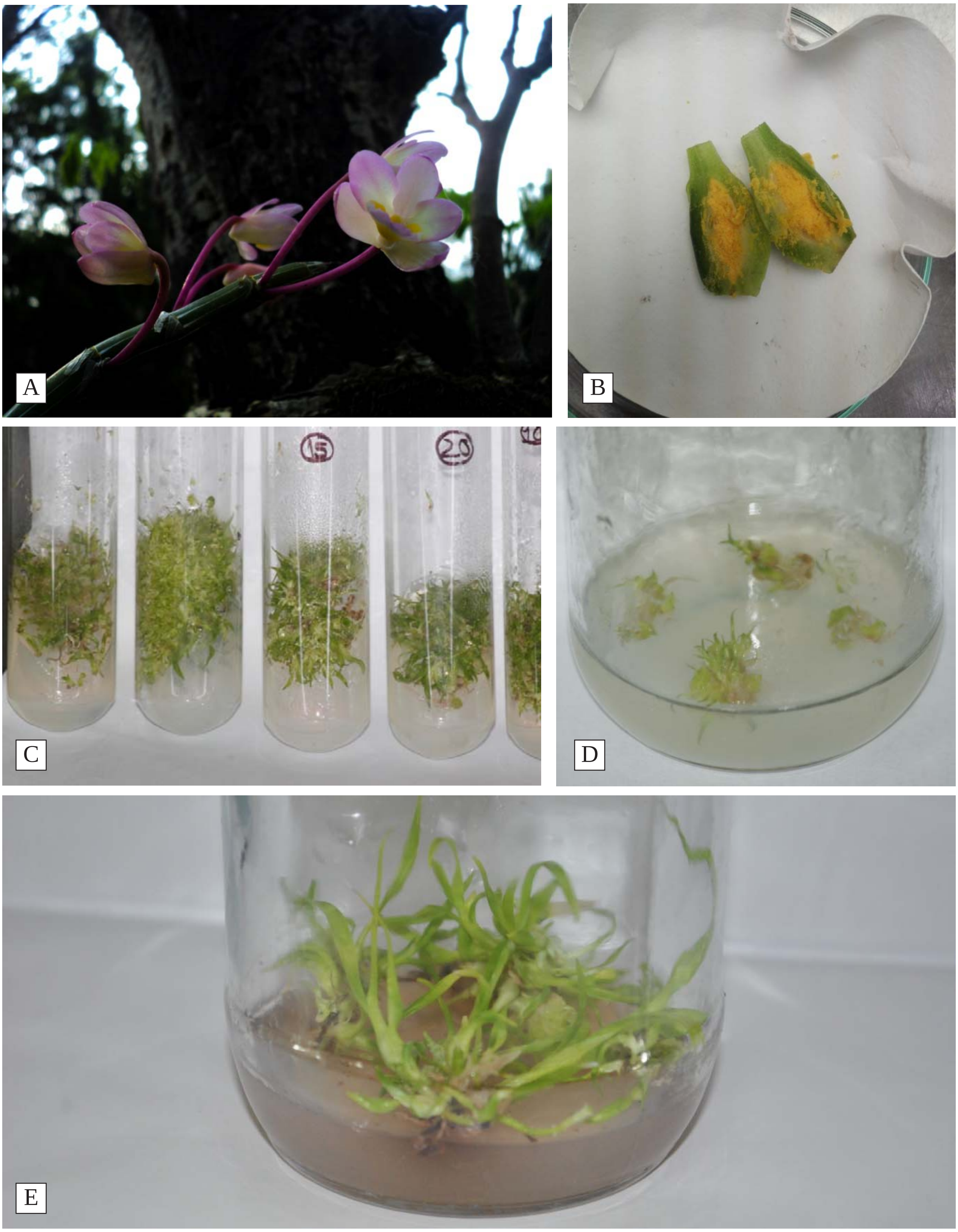
Vasudevan, R., & van Staden, J. (2010). Fruit harvesting time and corresponding morphological changes of seed integuments influence in vitro seed germination of *Dendrobium nobile* Lindl. *Plant Growth Regulation*, *60*, 237–246. doi: 10.1007/s10725-009-9437-1.

Yeung, E.C. (2017). A perspective on orchid seed and protocorm development. *Botanical studies*, *58*, 33. doi: 10.1186/s40529-017-0188-4.

Zeng, S., Wu, K., Teixeira da Silva, J.A., Zhang, J., Chen, Z., & Xia, N. (2012). Asymbiotic seed germination, seedling development and reintroduction of *Paphiopedilum wardii* Sumerh., an endangered terrestrial orchid. *Scientia Horticulturae*, *138*, 198–209. doi: 10.1016/j.scienta.2012.02.026.

Zeng, S.J., Zhang, Y., Teixeira da Silva, J.A., Wu, K.L., Zhang, J., & Duan, J. (2013). Seed biology and in vitro seed germination of *Cypripedium*. *Critical Reviews in Biotechnology*, *34*(4), 358-371.

Znaniecka, J., Krolicka, A., Sidwa-Gorycka, M., Rybcxynski, J.J., Szlachetko, D.L., & Lojkwoska, E. (2005). Asymbiotic germination, seedling development and plantlet propagation of *Encyclia* aff. *Oncidioides* – an endangered orchid. *Acta Societatis Botanicorum Poloniae*, *74*(3), 193-198.



Figures: In vitro culture of *Dendrobium crepidatum* A) Flowers of *Dendrobium crepidatum* Lindl. & Paxton, B) A longitudinally dissected capsule of *D. crepidatum*, C) Seedlings developed in half strength MS medium, D) Seedlings sub cultured in half strength MS medium supplemented with 1ìgmL^{-1} NAA, E) *D. crepidatum* plantlets developed in half strength MS medium supplemented with 2ìgmL^{-1} BAP and 1ìgmL^{-1} NAA.

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Phyto-cultural Heritage of Newah Community in Chapagaun, Nepal

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Abstract

The research dealt with the scientific documentation of plants used in the cultural activities of Newah community dwelling in Chapagaun of Nepal. The main purpose of this work was to enlist the plant species used during the rituals and festivals celebrated by this community, and also to assess the availability of such plants in the area. We employed open ended questionnaire to fill up the answers by interviewing well known priests, knowledgeable inhabitants and senior members of the community. We found that a total of 76 plants were most essential in their cultural practice. Among these, 46 plants belonging to 22 different families were used in the rituals from birth to death and 44 plants belonging to 23 different families were used in 14 different festivals celebrated throughout the year. Almost all the cultural activities basically utilized *Cynodon dactylon*, *Ficus religiosa*, *Jasminum multiflorum*, *Musa paradisiaca*, *Pisum sativum*, *Saccharum officinarum* and *Zingiber officinale* as the indispensable plant species. All the rituals and festivals involved a *puja* or worship set accompanied by a variety of seasonal flowers. Although many of the plant materials were not available in the wild, the Newah with their resilience kept their vibrant culture alive either by cultivating them in their home gardens or by purchasing them from outside. Only 21% of plant species essential for their culture were available in the natural habitat, 50% were cultivated in the home gardens or in the fields, whereas 29% were purchased in the packaged forms. Many plants of cultural value, once available in the wild have disappeared from their natural habitat. Hence, dissemination of knowledge about plants of cultural value among the youth would help conserve these plants for future generations.

Keywords: Biodiversity, Documentation, Festivals, Rituals,

Introduction

The Newah of Nepal are the people with a high degree of material culture and a distinctive social organization (Nepali, 2015). Chapagaun is one of the most prominent areas of Lalitpur district where Newah culture is still enthusiastically followed and preserved. Since Newah have their own perception of preserving and using biological diversity, they have a remarkable knowledge on regeneration and sustainable use of biological resources. Newah culture observes festivities throughout the year and there are rituals from the birth of a child to the death of a person. All these activities involve the extended family gatherings (Shrestha, 2013). Moreover, the rituals such as *Ihin* or getting a girl child worship *Aegle marmelos* or Bel fruit and *Janku* or worshipping the elderly people as gods are unique to Newah culture. Many plant species are considered indispensable for the specific deities during these rituals and festivals. Such beliefs invariably contribute to the conservation of particular plant species (Verschuuren, 2012), indicating the importance of phyto-cultural diversity of the community in the area.

Because of the mass migration of many other ethnicities into the Kathmandu valley, both Newah culture and the language are under threat today. Changing livelihood scenario due to modernization and less attractive economic potential of the traditional occupation as well as lack of transmission of the knowledge to the younger generations are the other threats the Newah community is facing today (Siwakoti & Joshi, 2020). Although Newah is a culturally rich ethnic group of Nepal, there is an inadequate literature about the cultural significance of plants used by the Newah community. It is universally accepted that the plants are better conserved when their apparent use and economical value are well known to the human society (Joshi & Joshi, 2005). Furthermore, the cultural beliefs and taboos help to preserve certain plants from exploitation and extinction (Kideghesho, 2009).

Hence, documentation of phyto-cultural heritage of the Newah community is essential for stopping the unprecedented loss of indigenous knowledge and also to conserve biological diversity. For the management of biological diversity, Strategic Plan for Biodiversity and Aichi Targets under Goal E and Target 18 (Convention on Biological Diversity [CBD], 2010) mentions that by 2020 the traditional knowledge, innovations and practices of indigenous and local communities relevant for the conservation and sustainable use of biodiversity and the customary use of biological resources are respected and fully integrated at all levels. Similarly, one of the cross-sectoral strategies for managing biodiversity according to Nepal National Biodiversity Action Plan 2014-2020 (Ministry of Forest and Soil Conservation [MoFSC], 2014) includes conservation and respect to traditional knowledge, innovations and practices of indigenous and local communities. The present research aims to enumerate the plants used by the Newah community of Chapagaun of Lalitpur, Nepal for their rituals and other religious activities as well as to assess the present status of such plants in the area.

Materials and Methods

Study area

Chapagaun lies in Godawari Municipality of Lalitpur district encompassing an area of approximately 7.27 km^2 (http://lalitpurmun.gov.np). It is located about 10 km south from Mangalbazar, the main city of Lalitpur district, at an altitude of approximately 1400 m (Shrestha, 2014). It lies between 27°36'00"N latitude and 85°18'00"E longitude (Figure 1). The total population of Chapagaun is 16081, out of which 6958 are Newah members (Central Bureau of Statistics [CBS], 2012).

Vegetation and flora

The sacred grove of Bajrabarahi temple is covered by approximately 0.15 km^2 of forest area where the most common trees are *Alnus nepalensis*, *Castanopsis indica*, *Choerospondias axillaris*, *Myrica esculenta*, *Prunus cerasoides*, and *Schima wallichi* (Shrestha, 2014). The forest is also habitat to many other plants of cultural and medicinal value. Rice, vegetables and mushroom are the most popular cash crops among the inhabitants.

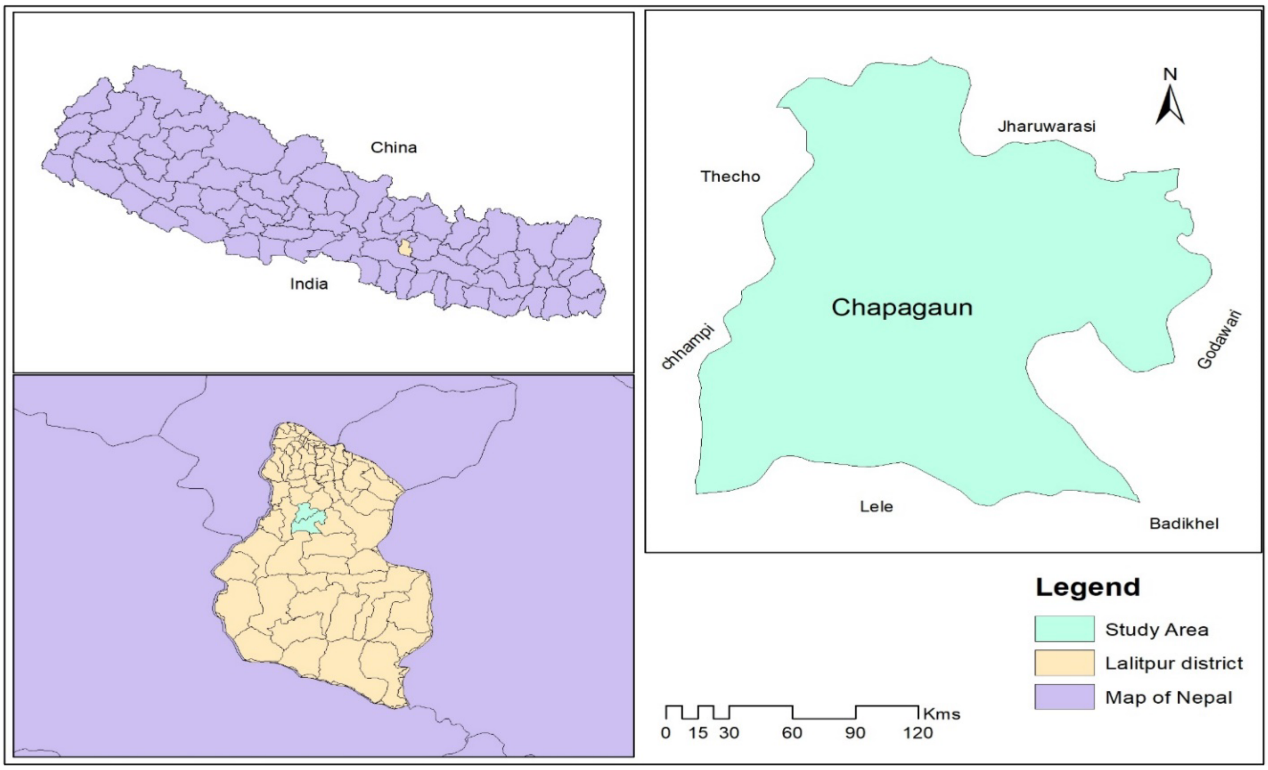

Figure 1: Site map of Chapagaun

Data collection

An open ended questionnaire was prepared for interviewing the key informants (Huntington, 2000). The informants comprised the priests locally known as *Baajyaa* and *Guvaju*, and also the knowledgeable community members as well as the elderly. The interviews were conducted during and after the rituals and festivals, and the questionnaire was filled up. Locally available plants of cultural value were collected, dried and mounted as herbarium specimens. Plants were identified first by their vernacular names, then by their common and scientfic names. The binomial nomenclature was based on the taxonomic keys, protologues and type specimens, acquired through the online databases such as http://www.biodiversitylibrary.org/ and http://plant.jstor.org/. The accepted scientific names were confirmed through online database http://www.plantlist.org/. The plant species were tabulated in an alphabetical order of the scientific names. The data were analyzed by using the charts.

Results and Discussion

The Newah community of Chapagaun have rich cultural heritage carried through generations. Traditional stewardship and management of plant resources in the forest area contribute to enhancing biodiversity. Although many plants of cultural value are not available in the wild, Newars have cultivated many such plants in their home gardens and in the fields. They purchase the unavialable plants from outside the city. People begin their day by showing deep spiritual respect to different dieties by offering a number of plant materials like *Curcuma longa* powder mixed with rice flour (Mhaasu sina), *Glycine max* (Mushya), *Gossypium arboreum* (Kapaya mah), *Hordeum vulgare* (Ta chho), *Oryza sativa* (Jaa ki), *Sesamum indicum* (Haamo) *and Valeriana jatamansi* (Naswankulcha) in the form of a '*puja*' or worship set. This *puja* set along with a variety of seasonal flowers is essential in all the rituals as well as the festivals.

The cultural activities of Newah of Chapagaun involve altogether 76 species of plants (Table 2). Out of these, 46 species belonging to 22 different families are used during eight rituals (Figure 2). A total of 44 plants belonging to 23 different families are utilized during different festivals (Figure 3). *Janku* or worshipping elderly person as god and *Mha puja* or worshipping one's own body is unique among Newah of the Kathmandu valley. *Cynodon dactylon*, *Ficus religiosa*, *Jasminum multiflorum*, *Musa paradisiaca*, *Pisum sativum*, *Saccharum officinarum* and *Zingiber officinale* have found place in almost all the rituals. Among 76 plant species, only 21% are found in the natural habitat, 50% are cultivated in their home gardens and in the fields, whereas 29% are purchased in the packaged form (Figure 4). Plants such as *Magnolia champaca*, *Thysanolaena latifolia* are found both in the wild and in the cultivated forms.

Table 1: Festivals and the day of celebration at Chapagaun, Lalitpur district

S. N.	Name of Festivals (Newah/Nepali)	Day of Celebration
1	*Mha puja*/Goverdhan Puja	Kartik shukla paru
2	*Kija Puja*/Bhai Tika	Kartik shukla pakshya dwitiya
3	*Sakhimila Punhi*	Kartik shukla punhi
4	*Yomari Punhi*	Marga shukla punhi
5	*Ghee-chaku sanhlu*/Maghe Shankranti	Magh shukla pakshya
6	*Sinkha Chahray*	Jestha krishna chaturdashi
7	*Gathamuga Cahray*/Gathemangal	Shrawan shukla chaturdashi
8	*Nag Panchami*	Shrawan shukla panchami
9	*Guh Punhi*/Janai Purnima	Shrawan shukla punhi
10	*Saparu*/Gai Jatra	Bhadra krishna paru
11	*Yenya Punhi*/Indra Jatra	Bhadra shukla punhi
12	*Mohani Nakha*/Dashain	Aswin shukla pratipada-Punhi
13	*Khicha Puja*/Kukur Tihar	Kartik krishna chaturdashi
14	*Laxmi Puja*	Kartik krishna aunsi

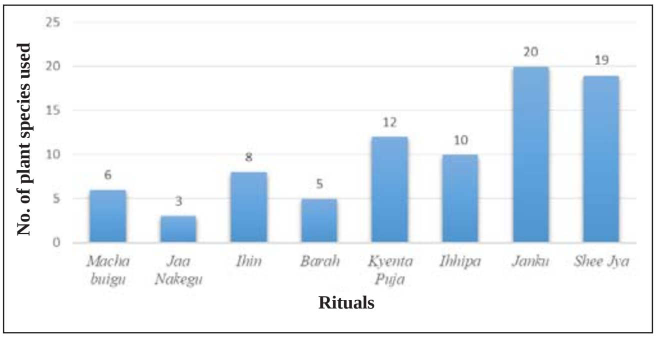

Figure 2: Number of plants used in the rituals.

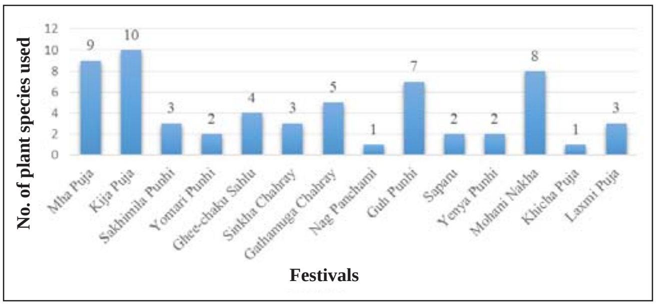

Figure 3: Number of plants used in the festivals.

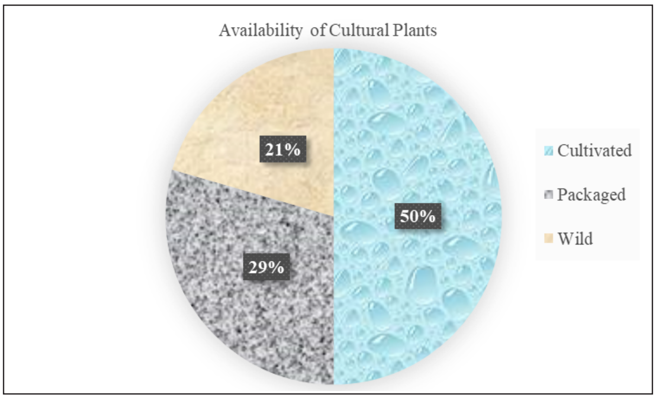

Figure 4: Availability of plant species in Chapagaun.

Chapagaun is named after the *Magnolia champaca* trees, commonly known as 'chaanp' (Shrestha, 2014), probably because these plants dominated the place in those days. Regrettably, only a few of them is found in the area at present. This should be a serious concern because of the infrequent chance of regeneration of these plants in the wild. Nevertheless,

Table 2: Plants used in different rituals and festivals at Chapagaun, Lalitpur district

S. N.	Name of Species	Family	Vernacular name	Common name	Uses		Availability
					Festivals	Rituals	
1	*Aegle marmelos* (L.) Correa	Rutaceae	*Bya*	Wood apple		*Ihin* (M_1)	P
2	*Allium sativum* L.	Amaryllidaceae	*Laba*	Garlic	*Ghee-chaku Sahlu*		C
3	*Amaranthus spinosus* L.	Amaranthaceae	*Bakan*	Slender amaranth	*Gathamuga Chahray*		W
4	*Anaphalis busua* (Buch.-Ham.) DC.	Compositae	*Bun swan*	Pearly everlasting	*Yenya Punhi*		W
5	*Areca catechu* L.	Arecaceae	*Gwey*	Betel nut		*Ihhipa* (M_3)	P
6	*Artemisia pallens* Wall. ex DC.	Compositae	*Dhawo swan*	Wormweed		*Ihhipa*	C
7	*Benincasa hispida* (Thunb.) Cogn.	Cucurbitaceae	*Bhuyu pasha*	Wax ground	*Mohani Nakha*		C
8	*Brassica juncea* (L.) Czern.	Brassicaceae	*Paka*	Indian Mustard		*Shee Jya* (D)	C
9	*Brassica rapa* L.	Brassicaceae	*Eeka*	Field Mustard		*Shee Jya*	C
10	*Buddleja asiatica* Lour.	Scrophulariaceae	*Sina swan*	Asian butterfly bush		*Shee Jya*	W
11	*Castanopsis indica* (Roxb. ex Lindl.) A. DC.	Fagaceae	*Syanguli*	Indian chest nut	*Mha Puja, Kija Puja*		W
12	*Chrysanthemum* sp.	Compositae	*Godawari swan*	Chrysanthamums	*Mha Puja, Kija Puja*		C
13	*Cicer arietinum* L.	Leguminosae	*Channa*	Chickpea	*Guh Punhi*	*Barah* (M_2)	P
14	*Citrus hystrix* DC.	Rutaceae	*Bhogatya*	Wild orange	*Mha Puja, Kija Puja*		C
15	*Citrus medica* L.	Rutaceae	*Tasi*	Citron	*Mha Puja, Kija Puja*		C
16	*Clitoria ternatea* L.	Leguminosae	*Aparajita swan*	Blue pea		*Janku* (W_2)	P
17	*Cocos nucifera* L.	Arecaceae	*Naikya*	Coconut	*Mohani Nakha*	*Jaa Nakegu* (RF), *Ihhipa*	P

S. N.	Name of Species	Family	Vernacular name	Common name	Uses		Availability
					Festivals	Rituals	
18	*Colocasia esculenta* (L.) Schott	Araceae	*Sakhi*	Cocoyam	*Sakhimila Punhi*		C
19	*Consolida ajacis* (L.) Schur.	Ranunculaceae	*Aji-fuli swan*	Toad flax		*Janku*	P
20	*Cynodon dactylon* (L.) Pers.	Poaceae	*Situ*	Bermuda grass	*Nag Panchami*	*Ihin, Kyenta Puja* (W_1), *Ihhipa, Janku, Shee Jya*	W
21	*Dendrocalamus hamiltonii* Nees & Arn. ex Munro	Poaceae	*Paa*	Tama bamboo		*Shee Jya*	W
22	*Desmostachya bipinnata* (L.) Stapf	Poaceae	*Kush*	Halfa grass		*Kyenta Puja, Janku, Shee Jya*	P
23	*Dioscorea alata* L.	Dioscoreaceae	*Gaa hee*	Nepal yam	*Ghee-chaku Sahlu*		C
24	*Drepanostachyum falcatum* (Nees) Keng f.	Poaceae	*Tee mah*	Himalayan bamboo		*Kyenta Puja*	W
25	*Eulaliopsis binata* (Retz.) C.E.Hubb.	Poaceae	*Sakhi khipa*	Sabai grass		*Kyenta Puja*	W
26	*Euphorbia pulcherrima* Willd. ex Klotzsch	Euphorbiaceae	*Lalpatya*	Poinsettia		*Ihhipa*	C
27	*Ficus benghalensis* L.	Moraceae	*Bar mah*	Banyan fig		*Ihin, Janku, Kyenta Puja, Shee Jya*	W
28	*Ficus racemosa* L.	Moraceae	*Dubosi*	Cluster Fig Tree		*Kyenta Puja, Janku, Shee Jya*	P
29	*Ficus religiosa* L.	Moraceae	*Wangala sima*	Sacred fig tree		*Macha Buigu* (BC), *Ihin, Kyenta Puja, Janku, Shee Jya*	W
30	*Gaultheria fragrantissima* Wall.	Ericaceae	*Dhashi kasi*	Indian winter green	*Gathamuga Chahray*		W
31	*Glycine max (*L.) Merr.	Leguminosae	*Mushya*	Soyabean	*Guh Punhi*	*Barah*	C
32	*Gomphrena globosa* L.	Amaranthaceae	*Gwega swan*	Globe amaranth	*Kija Puja*		C
33	*Hordeum vulgare* L.	Poaceae	*Jau mah*	Barley	*Mohani Nakha*		C
34	*Ipomoea batatas* (L.) Lam.	Convolvulaceae	*Hee chaku*	Sweet potato	*Sakhimila Punhi*		C
35	*Iris germanica* L.	Iridaceae	*Wochu swan*	Bearded iris		*Janku*	C
36	*Jasminum multiflorum* (Burm.f.) Andrews	Oleaceae	*Dapho swan*	Indian jasmine		*Macha Buigu, Ihin, Kyenta Puja, Ihhipa, Janku, Shee Jya*	C
37	*Jasminum sambac* (L.) Aiton	Oleaceae	*Chameli swan*	Jasmine		*Janku*	C
38	*Jasminum* sp.	Oleaceae	*Jee swan*			*Janku*	C
39	*Juglans regia* L.	Juglandaceae	*Khowshi*	Common Walnut	*Mha Puja, Kija Puja*		P
40	*Macrotyloma uniflorum (Lam.) Verdc.*	Leguminosae	*Kola*	Horse gram		*Janku*	P
41	*Magnolia champaca* (L.) Baill. ex Pierre	Magnoliaceae	*Cha swan*	Champak		*Ihin, Kyenta Puja, Janku, Shee Jya*	C, W

S. N.	Name of Species	Family	Vernacular name	Common name	Uses		Availability
					Festivals	Rituals	
42	*Malus domestica* Borkh.	Rosaceae	*Syaau*	Paradise Apple	*Mha Puja, Kija Puja*		P
43	*Mangifera indica* L.	Anacardiaceae	*Anh mah*	Mango		*Ihin, Kyenta Puja, Janku, Shee Jya*	P
44	*Mesua ferrea* L.	Calophyllaceae	*Rupkeshar*	Iron wood tree		*Janku*	P
45	*Musa paradisiaca* L.	Musaceae	*Kera mah*	Banana	*Mha Puja, Kija Puja, Guh punhi, Mohani Nakha*	*Macha Buigu,Jaa Nakegu, Ihin,Kyenta Puja, Ihhipa, Janku, Shee Jya*	P
46	*Narcissus tazetta* L.	Amaryllidaceae	*Gunakera swan*	Narcissus		*Ihhipa*	C
47	*Nelumbo nucifera* Gaertn.	Nelumbonaceae	*Pali swan*	Sacred lotus		*Janku*	C
48	*Nerium oleander* L.	Apocynaceae	*Kanhya swan*	Oleander		*Janku*	C
49	*Nyctanthes arbor-tristis* L.	Oleaceae	*Pallya swan*	Night flowering jasmine	*Yenya Punhi*		C
50	*Oryza sativa* L.	Poaceae	*Wah mah*	Rice	*Mohani Nakha, Yomari punhi*	*Shee Jya*	C
51	*Phaseolus lunatus* L.	Leguminosae	*Khocha simi*	Lima bean	*Laxmi Puja*		C
52	*Phaseolus vulgaris* L.	Leguminosae	*Simpu*	Common bean	*Guh Punhi*		P
53	*Phyllanthus emblica* L.	Phyllanthaceae	*Aambaa*	Indian gooseberry	*Mha Puja, Kija Puja*		W
54	*Pisum sativum* L.	Leguminosae	*Kasoo*	Field pea	*Guh Punhi, Saparu, Sinkha Chahray*	*Shee Jya*	C
55	*Prunus cerasoides* Buch.-Ham. ex D. Don	Rosaceae	*Foshi pasha*	Wild himalayan cherry	*Gathamuga Chahray*		W
56	*Psidium guajava* L.	Myrtaceae	*Aamashi*	Guava	*Kija Puja, Mha Puja*		C
57	*Pyrus pyrifolia* (Burm.f.) Nakai	Rosaceae	*Pashi*	Asian pear	*Saparu*		P
58	*Raphanus raphanistrum* subsp. *sativus* (L.) Domin	Brassicaceae	*Lain*	Radish	*Sakhimila Punhi*	*Jaa Nakegu, Ihhipa*	C
59	*Saccharum officinarum* L.	Poaceae	*Tu mah*	Sugarcane	*Mohani Nakha*	*Macha Buigu, Kyenta Puja, Ihhipa*	C
60	*Schleichera oleosa* (Lour.) Merr.	Sapindaceae	*Kusum swan*	Safflower		*Janku*	P
61	*Sesamum indicum* L.	Pedaliaceae	*Haamo*	Sesame	*Yomari Punhi*		P
62	*Sesbania grandiflora* (L.) Pers.	Leguminosae	*Agaste swan*	Vegetable hummingbird		*Janku*	P
63	*Sesbania sesban* (L.) Merr.	Leguminosae	*Laxmi swan*	River hemp	*Laxmi Puja*		P
64	*Sphagneticola calendulacea* (L.) Pruski	Compositae	*Vinllyah*	Creeping daisy		*Shee Jya*	C
65	*Spinacia oleracea* L.	Amaranthaceae	*Pala*	Spinach	*Ghee-chaku Sahlu*		C
66	*Tabernaemontana divaricata* (L.) R.Br. ex Roem. & Schult.	Apocynaceae	*Tanhllyah*	Pinwheel flower		*Shee Jya*	C
67	*Tagetes erecta* L.	Compositae	*Tapho swan*	Marigold	*Khicha Puja*		C

S. N.	Name of Species	Family	Vernacular name	Common name	Uses		Availability
					Festivals	Rituals	
68	*Thymus vulgaris* L.	Lamiaceae	*Emuu*	Thyme		*Macha Buigu*	P
69	*Thysanolaena latifolia* (Roxb. ex Hornem.) Honda	Poaceae	*Tuphi*	Bouquet grass	*Laxmi Puja*	*Shee Jya*	C, W
70	*Triticum aestivum* L.	Poaceae	*Chhawali*	Wheat	*Gathamuga Chahray*	*Barah*	C
71	*Urtica* sp.	Urticaceae	*Nhyakan*	Stinging neetle	*Gathamuga Chahray*		W
72	*Vicia faba* L.	Leguminosae	*Bakula*	Faba bean	*Guh Punhi*	*Barah*	C
73	*Vigna mungo* (L.) Hepper	Leguminosae	*Maay*	Black gram	*Guh Punhi, Sinkha Chahray*		P
74	*Vigna radiata* (L.) R.Wilczek	Leguminosae	*Muu*	Green gram	*Guh Punhi, Sinkha Chahray*		P
75	*Zea mays* L.	Poaceae	*Kani mah*	Corn	*Mohani Nakha*	*Barah*	C
76	*Zingiber officinale* Roscoe	Zingiberaceae	*Palu*	Ginger	*Ghee-chaku Sahlu, Mohani Nakha*	*Macha Buigu, Shee Jya*	C

Note: C= Cultivated, P= Packaged, W= Wild, BC= Birth of a Child, RF= Rice Feeding Ceremony, M_1= getting a girl child worship *Aegle marmelos* or Bel fruit, M_2= getting a girl worship Sun, W_1= Worshipping a Young Boy before Entering Adolescence M_3= Marriage Ceremony, W_2= Worshipping Elderly Person as God, D= Death ritual

the traditional practices help in conserving biological diversity at the species level. This notion aligns with the goal E of Convention on Biological Diversity (2010), which calls for the protection and promotion of the innovations and practices of indigenous and local communities embodying traditional lifestyles relevant for the conservation and sustainable use of biological diversity. Cultural practices amongst the communities are persistent in different parts of Nepal. The list of plants used in different rituals of Newah of other parts of Nepal (Rajbhandari, 2008) are mostly similar. However, the compulsory use of *Narcissus poeticus* is unique among the Newah of Chapagaun. Moreover, the use of *Ficus racemosa* during *Ihin* is not in practice in the area. Extensive use of rice kernels and grains is the common cultural practice of Newah community of Chapagaun. *Yomari punhi* is one such festival, which celebrates the harvest of rice by making delicious preparations from the flour of rice grains. The grains of rice and pounded rice are used in other communities of Nepal as well, such as during the marriage ceremony of Magar community of Junigaun, Palpa (Ahearn, 2011). Leaves and flowers of *Buddleja asiatica* are offered during the worship of each deity in Chapagaun. This plant is valued by other communities such as Thangmi of Dolakha and Tamang of Kathmandu valley. The white flowers of this plant are made into necklaces and worn by the female relatives of the groom during Thangmi wedding rituals (Turin, 2003). The leaves of this plant are used for the worship by the Tamang (Shrestha, 1988). Newah of Chapagaun and Lepcha of Ilam (Manandhar, 2002) use *Zingiber officinale* in most of their cultural activities. India shares many phyto-cultural similarities with the Nepalese indigenous communities. *Mangifera indica* or mango leaves are considered indispensible for the wedding ceremony and Ganesh Chaturthi worship in India (Subrahmanian et al., 1997). Newah of Chapagaun also utilize the leaves of mango in a number of rituals such as *Ihin*, *Kyenta puja*, *Janku* and *Shee jya*. The community of Apatani, Arunachal Pradesh, India uses the leaves of *Castanopsis indica* for the preparation of *Agyang* (altar) during the rituals of *Tamu agyang* and *Kharung agyang* (Bamin & Gajurel, 2015). Newah of Chapagaun consider the fruits of this plant essential during the rituals of *Mha puja* and *Kija puja* (Rajbhandari, 2007). Indigenous communities of other parts of the world also show common cultural practice regarding the use of plants. In Oman, flowers of *Jasminum sambac* are sprinkled on the child's head by other children on their first birthdays (Walsh, 2004). The Newah of Chapagaun offer the flowers of this plant during *Janku* or the worship of the senior member of the family as a god. Across North America, *Euphorbia pulcherrima* flowers are used for Christmas decorations (Bussell, 2009), amusingly these flowers are considered indispensible in the *Ihhipa* or the wedding ceremony of Newah of Chapagaun. Plants such as *Cynodon dactylon*, *Juglans regia*, *Urtica dioca* are frequently used by Newah of Chapagaun in their cultural practices whereas the aboriginal people of Sikles, Kaski district of Nepal use them for curing a variety of diseases (Gurung et al., 2008). Similarly, plants such as *Gaultheria fragrantissima* and *Urtica dioca* are used by the Newah of Pharping village of Kathmandu valley for treating a range of diseases (Balami, 2004), while the Newah of Chapagaun utilize them for the cultural activities.

Conclusion

The cultural knowledge of Newah community has passed on from generations through language, practical teachings and a few printed materials. Nevertheless, it has shaped their ways of life, sense of conservation and love towards nature. Many plants used in the past for the cultural activities have disappeared from their natural habitat. Therefore, the locals who directly take part in the cultural activities grow the plants themselves. The dissemination of knowledge about plants of cultural value among the youth would help conserve such plants in a larger scale.

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References

Ahearn, L.M. (2011). *Living language: An introduction to linguistic Anthropology*. West Sussex, United Kingdom: Wiley-Blackwell.

Balami, N. (2004). Ethnomedicinal uses of plants among the Newar community of Pharping village of Kathmandu District, Nepal. *Tribhuvan University Journal, 24*(1), 13-19.

Bamin, Y., & Gajurel, P.R. (2015). Traditional use and conservation of some selected plants used in festivals and rituals in Apatani plateau of Arunachal Pradesh, India. *International Journal of Conservation Science, 6*(2), 189-200.

Biodiversity Heritage Library. Retrieved from https://www.biodiversitylibrary.org/

Bussell, G. (2009). Get ready for holiday flowers. *Southern Living. 44* (12), 88.

Central Bureau of Statistics. (2012). *National Population and Housing Census 2011*. Kathmandu, Nepal: Government of Nepal.

Convention on Biological Diversity. (2010). *Strategic Plan for Biodiversity 2011-2020 and the Aichi Targets*. Montreal: Author.

Ministry of Forest and Soil Conservation. (2014). Nepal National Biodiversity Strategy and Action Plan 2014-2020. Kathmandu, Nepal: Government of Nepal.

Gurung, L.J., Rajbhandary, S., & Ranjitkar, S. (2008). Medicinal Plants in Nepal: An Anthology of Contemporary Research, Jha, P.K., Karmacharya, S.B., Chettri, M.K., Thapa, C.B. & Shrestha, B.B. (Eds.), *Indigenous Knowledge of Medicinal Plants in Mid Hill of Nepal: A Case Study of Sikles Area of Kaski District* (pp. 152-163). Kathmandu, Nepal: Ecological Society.

Huntington, H.P. (2000). Using traditional ecological knowledge in science: Methods and applications. *Ecological Applications, 10*, 1270-1274.

Joshi, A.R., & Joshi, K. (2005). *Ethnobotany and Conservation of Plant Diversity in Nepal, Status, Bibliography and Agenda for Sustainable*

Management. Kathmandu, Nepal: Repro Vision Press Pvt. Ltd.

JSTOR Global Plants: Retrieved from https://plants.jstor.org/

Kidegheso, J.R. (2009). The potentials of traditional African cultural practices in mitigating overexploitation of wildlife species and habitat loss: experience of Tanzania. *International Journal of Biodiversity Science & Management*, *5*(2), 83-94.

Lalitpur Metropolitan City, Government of Nepal. Retrieved from http://lalitpurmun.gov.np/en/geographical-resource-map

Manandhar, N.P. (2002). *Plants and people of Nepal*. Portland, Oregon, USA: Timber Press.

Nepali, G.S. (2015). *The Newars*. Kathmandu, Nepal: Mandala Book Point.

Rajbhandari, B.M. (2007). *Salana Manaudai Aairaheko Chadparva*. Kathmandu: Mass Printing Press.

Rajbhandari, B.M. (2008). *Garvadekhi Maronaparanta Samamako Sanskar*. Kathmandu: Mass Printing Press.

Shrestha, P.P. (2014). Chapagaun Lai Najikbata Niyalda. *In*: M.P. Chapagain, B. Karmacharya, M. Maharjan, R. Acharya, & B.T. Shrestha (Eds.), *Chapagaun ko Chinari* (pp. 65–69). Chapagaun, Lalitpur: Chapagaun V.D.C. Office, Government of Nepal.

Shrestha, P. (1988). Contribution to the Ethnobotany of the Tamangs of Kathmandu valley. *Contributions to Nepalese Studies, 15*(2), 247-266.

Shrestha, S.K. (2013). *Audience and cosmos: A Performative Reading of Indra Jatra.* (M.Phil. Thesis). Central Department of English. Tribhuvan University, Kathmandu, Nepal.

Siwakoti, M. & Joshi, N. (2020). Plants and Culture of Newar Community. *In*: M. Siwakoti, M., P.K. Jha, S. Rajbhandary and S.K. Rai (Eds.), *Plant Diversity in Nepal*, (pp. 125-141). Kathmandu, Nepal: Botanical Society of Nepal.

Subrahmanian, N., Hikosaka, S., & Samuel, G.J. (1997). *Tamil social history.* Madras, India: Institute of Asian Studies.

The Plant List (2013). Version 1.1. Published on the Internet; http://www.theplantlist.org/ (accessed 13th April, 2017).

Turin, M. (2003). Ethnobotanical notes on Thangmi plant names and their medicinal and ritual uses. *Contribution to Nepalese Studies*, *30*(1), 19-52.

Verschuuren, B. (2012). Integrating biocultural values in nature conservation: perceptions of culturally significant sites and species in adaptive management. *In*: G. Pungetti, G. Oviedo & D. Hooke (Eds.), *Sacred Species and Sites: Advances in Biocultural Conservation* (pp. 231-246). United Kingdom: Cambridge University Press.

Walsh, T. (2004). *Jasmine Scents of Arabia*. Riyadh: Saudi Research & Publishing Company.

Ethnobotanical Knowledge of the Tharu Community Living in Tulsipur Sub-metropolitan City, Dang, Nepal

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Abstract

An ethnobotanical work was conducted in Tulsipur sub metropolitan city of Dang District, Rapti, Nepal. The main objective was to document the indigenous knowledge of the Tharu community in utilization of plant for various purposes. Ethnobotanical survey was made by direct field visit, herbarium collection and identification was done with the help of national and international literatures. The Tharus of Tulsipur Municipality were found to have diversified knowledge in utilization of plant resources in 8 different forms. From the study area, 114 species belonging 96 genera of 44 families that were utilized for various purposes were documented. Among the taxa, Poaceae and Leguminosae have more species (12 species each). Among the reported plant species, 55 species were food plant, 33 medicinal plant, 14 fodder plant, 11 used for cultural purposes, 11 for making utensil, 7 were wild vegetable plant, 7 were firewood plant and 7 were timber plant. From the research 18 plants were found to have multiple uses. The Tharus of the study area were found to have good knowledge on food plants followed by medicinal plants.

Keywords: Ethnobotany, Fodder plants, Food plants, Medicinal plants, Wild vegetable

Introduction

Nepal has a rich and varied flora due to its diversified topography and variable climatic conditions. The physical setup of the country comprises altitudinal gradient ranges from tropical region to alpine region. About 44.7% of the country's area is covered by forest (Government of Nepal [GoN], 2017). From the ancient period man lives closely associated with nature and are depended on it for their survival. Many living groups of people, having diversified ethnic history of rituals and performance, who are more of less isolated form modem world and are closely associated with their ambient vegetation is the emporia of ethno botanical research (Pal & Jain, 1998). The surrounding environment directly and indirectly influences the human life and culture.

The interaction between plant and people is as long as human being existence in this planet (Shah et al., 2015). Plants are the part of our environment. People uses plants around them for many proposes like; food, shelter, dyes, cosmetics, clothing, medicine etc. from their surrounding vegetation. They gathered the knowledge from the environment, use them and pass them through generation to generation with or without written documents. But many have disappeared due to several reasons. Without proper documentation, these resourceful of information or knowledge may be disappeared for ever. So a recent branch of botany, ethnobotany arise which provide the proper documentation and preservation system of traditional plant use information, accumulated in a community through generation by generation in relation with their culture. Ethnobotany is a term used to encompass studies to describe local people's interaction with the natural surroundings (Eldeen et al., 2016). Ethnobotany is the scientific study of the relationships that exist between peoples and plants.

Mother tongue statistics of Nepalese people represent 100 different ethnic groups and more than 100 languages are spoken in Nepal, among which Tharu is one (Central Bureau of Statistics[CBS], 2011). Traditionally, ethnic groups are known to use large number of wild plants for various purposes like medicine, food, fodder, fuel, culture, etc (Mishra & Mishra, 2014).

Many people have done ethnobotanical works in remote areas or in VDCs (Acharya & Acharya, 2009; Aryal, 2009; Acharya, 2012). Some work only deals with the medicinal plants only or wild plants only (Chapagain, (2004), Bhatt, (2012), Bajpai et al., (2016), etc.). So this work was carried out in order to know whether the Tharu people living in the developed places i.e. municipalities either have lost or retained their knowledge of utilizing both wild and cultivated plant for various purposes.

Materials and Methods

Study area

Tulsipur sub-metropolitan city was chosen as the study area. The rich floral diversity, the vast forest and large ethic population provide the ideal condition for ethnobotanical study in this area. The study was conducted in ward number 6, 9,12,13,15 and 20 of Tulsipur municipality. It is situated between 28°08' N latitude and 82°00' E longitude with the total area of 384.63 sq. km. Its altitude ranges from 529 m asl to 2050 m asl (Figure 1). It is a cosmopolitan and culturally diverse place. This place has majority of the Tharus ethnicity, Brahmins and Chhetris came from the Southern mountains where Nepali is the dominant language. Tulsipur contains a total population 141528 (CBS, 2011), where the Tharus people constitute more than 20% of the total population.

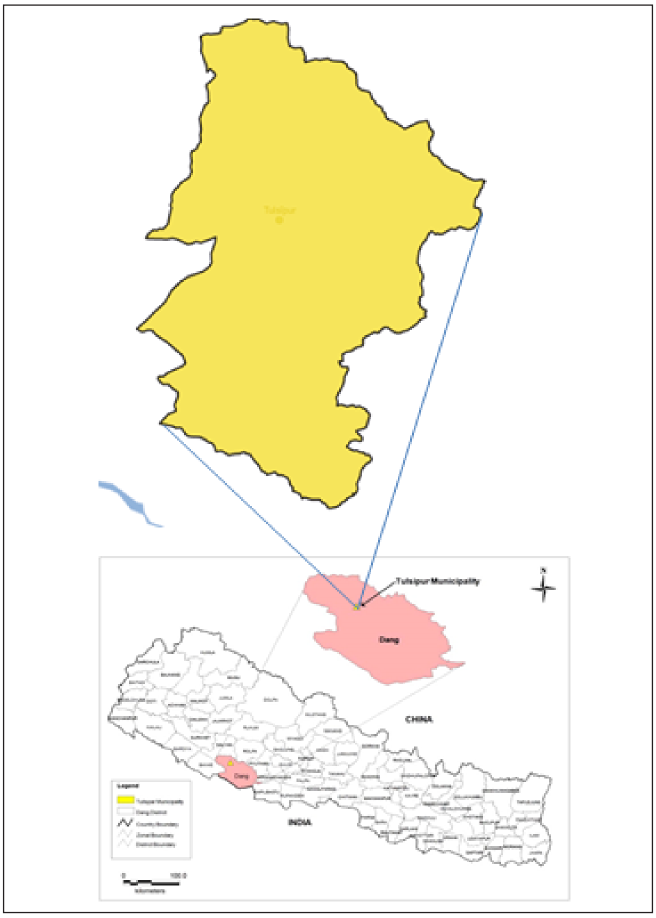

Figure 1: Map of the Study area

Data collection

A total of 12 weeks was spent for the field work in the study area. The area was visited frequently during the study period (2016-2018). 5 teachers, 3 herbalists, 25 farmers, 10 students, 12 social leaders, 7 traditional healers, 11 housewives were interviewed mainly concerning their knowledge on food, fodder, medicine from the plants and their parts, local names, with miscellaneous uses etc. Household survey was conducted on a random basis to obtain information on people's perception on conservation, use of plants, availability and method of use of plant parts, preference of plants for certain use etc. Structured and unstructured interview methods were used for the study. It was basically focused on the history, culture, tradition, socio-economic and present condition of the study. The secondary data was collected from different reports of ethnobotanical studies from central library of Tribhuvan University (TU), Kathmandu University and Department of Plant Resources, Government of Nepal. Similarly various books, journals published by native and foreign institutions, newspapers and documents from internet were consulted for relevant information.

The plant specimens those were collected and preserved were identified with the help of the references materials of National Herbarium (KATH), Tribhuvan University Central Herbarium (TUCH), Central Department of Botany, experts, taxonomists and other standard literatures like Hara et al. 1982, Press et al. 2000.

Results and Discussion

The Tharus of Tulsipur sub-metropolitan city are found to possess a very rich ethnobotanical knowledge. They have been making use of a large number of plants species for various purposes such as medicine, fodder, firewood, timber, ceremonies, etc. A total 114 plant species representing 44 families with 96 genera have been reported to use among the Tharus of the study area. Both cultivated and non-cultivated plant species have been documented in the present study.

The Tharu community was found to use more herbs (39%) species followed by trees (35%), shrubs (17%) and vines (9%).This proportion was similar to other studies (Shrestha & Dhillion, 2003; Uprety et al., 2010; Singh et al., 2012).The higher uses for herbs may be due to easy availability and its multipurpose uses like food, fodder, medicine, wild vegetables, etc. (Figure 2)

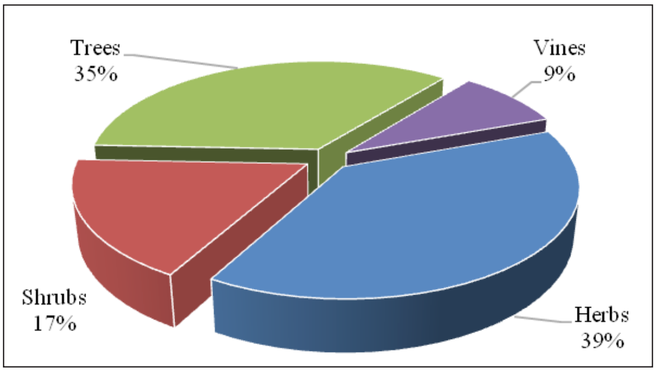

Figure 2: Used plants habit

In the study area, maximum number (55 species) of food plants was found to be used followed by medicinal plants (33 species) and fodder plants (14 species).The lower number of medicinal plants may be due to lack of faith on healers and wider use of modern medicine and inadequacy of plant availability (Mandar & Chaudhary, 1992). Less number of firewood plants and timber plants used may be due to restriction for firewood and timber collection in the forest. People also use gas and dung cake for cooking (Figure 3).

Fodder plants like *Bambusa* sp., *Leucaena leucocephala*, *Oryza sativa*, *Zea mays*, firewood plants like *Lantana camara*, *Dalbergia sissoo* and food crops like *Raphanus raphanistrum* subsp. *sativus*, *Allium cepa*, *Brassica rapa* etc. are also used by the Tharus of Kapilvastu district, Nepal (Aryal, 2009).

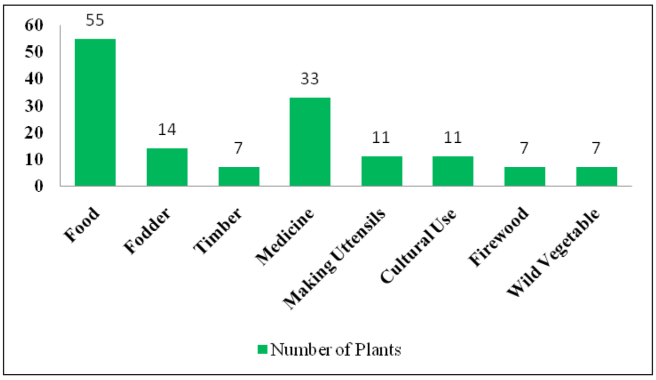

Figure 3: Application of plant and their number

Medicinal plants *Acorus calamus*, *Ageratum conyzoides*, *Artemisia vulgaris* and *Calotropis gigantea* are the common medicinal plants used by the Tharu people of Chitwan, Kapilvastu and Dang District (Dangol & Gurung, 1991; Aryal, 2009). *Achyranthes aspera*, *Musa paradisiaca* and *Plumbago zeylanica* are the common medicinal plants used by the Tharu people of India and Dang (Kumar & Bharati, 2014).

Maximum numbers of leaves were found to be used followed by fruit, stem, seed and roots. This is due to multiple uses of leaves (Figure 4). The different parts of plants such as leaves, flower, fruit, bark, latex, branch twig, rhizome, etc. were used as medicine and other purposes. The study showed that the people use different parts of the same plants for different purposes (food, fodder, firewood, timber, etc.) and for different aliments (Rokaya et al., 2012).

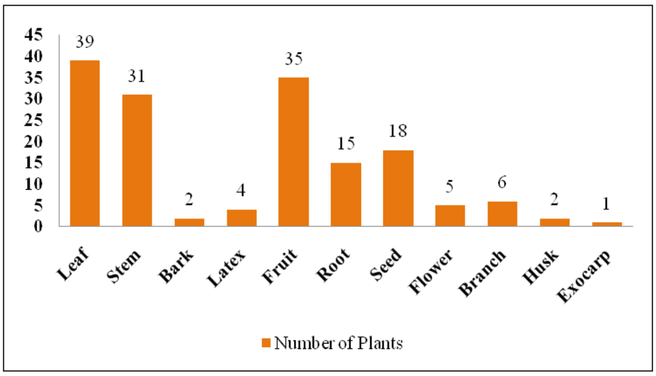

Figure 4: Plants parts used

Maximum numbers of food plants used were cultivated, while minimum numbers of plants used as medicine, making utensils, timber, firewood, fodder were wild (Figure 5). This situation of using large number of uncultivated plants could in long term, lead to the depletion of resources or even extinction of the plant species if they are harvested in large amount.

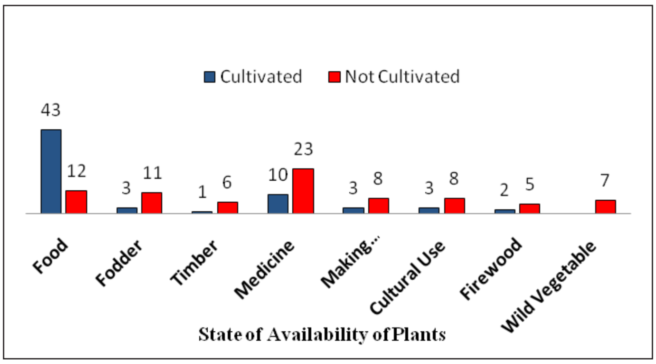

Figure 5: State of availability of used plants

Conclusion

The Tharus Community of Tulsipur sub metropolitan city was found to have very diversified knowledge in the utilization of the available plant resources. They were found to use 114 plant species belonging to 96 genera and 44 families. Various parts of the plants like leaves, root, branch, bark etc. were found to be used for various different purposes like food, fodder, timber, medicine, etc. Eighteen plants were also found to have multiple uses. We also believe that there may be other species of plants used for other purposes by the Tharu community.

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References

Acharya, R. & Acharya, K.P. (2009). Ethnobotanical study of medicinal plants used by Tharu community of Parroha VDC, Rupandehi District, Nepal. *Scientific World*., *7*(7), 80-84.

Acharya, R. (2012). Ethnobotanical study of Medicinal plants of Resunga hill used by the Magar community of Balagaun VDC, Gulmi District, Nepal. *Scientific World*., *10*(20), 54-65.

Aryal, S.P. (2009). *Ethnobotany of Tharu: Incorporation of culture and biodiversity conservation (A case study of Jaganagar VDC of Kapilvastu District*. Nepal: Tribhuvan University.

Bajpai, O., Pandey J. & Chaudhary, L.B. (2016). Ethno medicinal uses of Tree species by Tharu tribes in the Himalayan Terai Region of India. *Research Journal of Medicinal Plants. 10*, 19-41.

Bhatt, P. (2012). *Ethnobotanical study of medicinal plants used by Tharus of Kanchanpur District, Far-Western Nepal*. Nepal: Tribhuvan University.

Central Bureau of Statistics. (2011). *Population Census of Nepal* 2011. Thapathali, Kathmandu, Nepal: Author.

Chapagain, D.J. (2004). *Medicinal plants used by the Tharu community in the buffer zone of Royal Bardiya N.P., Nepal*. Nepal: Tribhuvan University.

Dangal, D.R. & Gurung, S.B. (2008).Ethnobotany of the Tharu tribe of Chitwan District, Nepal. *Pharmaceutical Biology*, *29*(3), 103-109.

Eldeen, I.M.S., Effendy, M.A.W. & Muhammad, T.S.T. (2016). Ethnobotany: Challenges and future perspectives. *Research Journal of Medicinal Plants*., *10*, 382-387.

Government of Nepal. (2017). *Four investment program: Invent plan for Nepal inventory in fourts for property at a time of transformation*. Kathmandu: Author.

Kumar, R. & Bharati, K. (2014). Ethnomedicines of Tharu Tribes of Dudhwa National Park, India. *Ethnobotany Research & Applications*, *12*, 1-14.

Hara, H., Charter, A.O. & Williams, L.H.J. (1982). *An Enumeration of the Flowering Plants of Nepal*., London: British Museum of Natural History

Mandar, L.N. & Chaudhary, R.P. (1992). Medicinal plants and their traditional use by tribal people of Saptari district, Nepal. *First Nepal Botanical Conference*. Aug. 11-12, Kathmandu, Nepal. Proceedings.

Mishra, S. & Mishra, M.K. (2014). Ethnobotanical study of plants with edible underground parts of south Odisha, India. *International Journal of Research in Agriculture and food science.*, *4*(2), 51-8.

Pal, D.C. & Jain,S.K. (1998). *Tribal Medicine. Naya Prakash*. Calcutta. 317.

Press, J.R., Shrestha, K.K. and Sutton, D.A. 2000. *Annonated Checklist of the Flowering Plants of Nepal.*, London: The Natural History Museum.

Rokaya, M.B., Munzbergovaa, Z., Timsina, B. & Bhattarai, K.R. (2012). Rheum austral D. Don: A review of its botany, ethnobotany, phytochemistry and pharmacology. *Journal of Ethnopharmacology*, *141*, 761-774.

Shah, A.H., Khan, S.M., Shah, A.H., Mehmood, A., Rahmen, I. & Ahmad.(2015). Cultural uses of plants among basikhel tribe of district for ghar Khyber Pakhtunkhwa, Pakisthan. *Pak.J. Bot*., *47*, 17-35.

Shrestha, P.M. & Dhillion, S.S. (2003). Medicinal plants diversity and use in highlands of Dolakha district, Nepal. *Journal of Ethnopharmacology.*, *86*, 81-96.

Singh, A.G., Kumar, A. & Tiwari, D.D. (2012). An ethnobotanical survey of medicinal plants used in terai forest of western Nepal. *Journal of Ethnobotany and Ethnomedicine.*, *8*(19).

Uprety, Y., Asselin, H., Boon, E.K., Yadav, S. & Shrestha, K.K. (2010). Indigenous use and bioefficacy of medicinal plants in the Rasuwa district, Central Nepal. *Journal of Ethnobiology and* Ethnomedicine, *6*(3).

Table 1: Plants with their parts used and purpose of use

S.N.	Botanical Name	Vernacular Name	Family	Nature	Part Used	Used For
1	*Abelmoschus esculentus* (L.) Moench.	Ramtoria	Malvaceae	C	Fruit	Fo.
2	*Acacia catechu* (L.f.) Willd.	Khayar	Fabaceae	NC	Stem, Bark	Ut., Me.
3	*Achyranthes aspera* L.	Datiwon	Amaranthaceae	NC	Root	Me.
4	*Achyranthes bidentata* Blume	Apamagra	Amaranthaceae	NC	Twig of branch, Root	Me
5	*Acorus calamus* L.	Bojho	Araceae	C	Rhizome	Me.
6	*Adenium obesum* (Forssc) Roem. & Schult.	Malati	Apocynaceae	NC	Flower	Cu.
7	*Aegle marmelos* (L.) Correa	Bel	Rutaceae	NC	Leaf, Fruit, Branch	Me.,Cu.
8	*Ageratum conyzoides* L.	Raunne	Asteraceae	NC	Leaf, Shoot	Me., Fd.
9	*Allium cepa* L.	Pyaj	Alliaceae	C	Leaf, Tuber	Fo.
10	*Allium sativum* L.	Lasun	Alliaceae	C	Leaf,Tuber	Fo.
11	*Aloe vera* (L.) Burm.f.	Gheu kumara	Asphodelaceae	C	Latex	Me.
12	*Arisaema tortuosum* (Wall.) Scnott	Banko	Araceae	NC	Shoot	Wv.
13	*Artemisia vulgaris* Mattf.	Paati	Compositae	NC	Leaf	Me.
14	*Artocarpus heterophyllus* Lam.	Katahar	Moraceae	C	Fruit, Seed	Fo.
15	*Azadirachta indica* A. Juss.	Neem	Meliaceae	NC	Leaf	Me.
16	*Bahunia malabarica* Roxb.	Malu	Fabaceae	NC	Leaf, Bark	Ut.
17	*Bambusa malingensis* McClure	Bans	Poaceae	C	Young shoot,leaf, Matured shoot	Fo., Fd., Ut., Tm., Fw.
18	*Bauhinia tomentosa* L.	Emli	Fabaceae	NC	Fruit	Fo.
19	*Bauhinia veriegata* (L.) Benth.	Koiralo	Fabaceae	NC	Flower	Wv.
20	*Bombax ceiba* L.	Simal	Malvaceae	NC	Stem	Tm.
22	*Brassica juncea* (L.) Vassilii	RayoSaag	Brassicaceae	C	Leaf	Fo.
21	*Brassica rapa* L.	Tori	Brassicaceae	C	Leaf twig, Seed	Fo.
23	*Cajanus cajan* (L.) Millsp.	Rahar	Leguminosae	C	Grain	Fo.
24	*Calotropis gigantea*(L.) Dryand.	Aakh	Apocynaceae	NC	Latex	Me.
25	*Capsicum annuum*L.	Khursani	Solanaceae	C	Fruit	Fo.
26	*Carica papaya* L.	Mewa	Caricaceae	C	Fruit	Fo.
27	*Carissa macrocarpa* (Eckl.) A.DC.	Karauti	Apocynaceae	NC	Fruit	Fo.
28	*Centella asiatica* (L.) Urb.	Ghortapre	Apiaceae	NC	Leaf	Me.
29	*Cheilanthes micropteris* Sw.	Kuthrukay	Pteridaceae	NC	Shoot	Wv.
98	*Choerospondias axillaris* (Roxb.) B.L.Burtt & A.W.Hill	Amaro	Anacardiaceae	NC	Fruit	Fo.
30	*Cicer arietinum* L.	Chana	Leguminosae	C	Seed	Fo.
31	*Citrus aurantifolia* (Christ.) Swingle	Kagati	Rutaceae	C	Fruit	Fo.
32	*Citrus junos* Siebold ex Tanaka	Jyamir	Rutaceae	C	Fruit	Fo.
33	*Citrus limon* (L.) Osbeck	Nibuva	Rutaceae	C	Fruit	Fo.
34	*Clerodendrum infortunatum* L.	Tite	Lamiaceae	NC	Leaf	Fd.
35	*Colocasia esculentas* (L.) Schott	Pidalu	Araceae	C	Leaf, Tuber	Fo.
37	*Cucumis sativus* L.	Kakro	Cucurbitaceae	C	Fruit	Fo.
38	*Cucurbita pepo L.*	Pharsi	Cucurbitaceae	C	Leaf twig, Fruit	Fo.
39	*Cynodon dactylon* (L.) Pers.	Dubo	Poaceae	NC	Shoot	Fd.
40	*Dalbergia sissoo* DC.	Sisoo/ Sisau	Leguminosae	NC	Stem, Leaf, Branch	Tm., Me., Fw

S.N.	Botanical Name	Vernacular Name	Family	Nature	Part Used	Used For
42	*Datura metel* L.	Dhaturo	Solanaceae	NC	Seed	Me.
41	*Desmodium oojeinense* (Roxb.) H. Ohashi	Sadan	Leguminosae	NC	Stem	Ut.
43	*Dioscorea alata* L.	Tarul	Dioscoreaceae	NC	Tuber	Fo.
44	*Diploknema butyracea* (Roxb.) H.J.Lam	Cheuri	Sapotaceae	NC	Fruit, Seed	Fo.
45	*Dolichos aciphyllus* R.Wilczek	Hiundesimi	Leguminosae	C	Fruit, Leaf	Fo., Me.
46	*Dryopteris cochleata* (D.Don.) C.Chr.	Neuro	Dryopteridaceae	NC	Shoot	Wv.
36	*Eichhornia crassipes* (Mart.) Solms	Jaluko	Pontederiaceae	NC	Leaf	Wv.
47	*Eucalyptus oblique* L'Her.	Masala	Myrtaceae	NC	Stem	Tm.
48	*Ficus benghalensis* L.	Bar	Moraceae	NC	Leaves, Latex	Cu., Me.
49	*Ficus lacor* Buch. (Ham.)	Kavro	Moraceae	NC	Flower	Fo.
50	*Ficus religiosa* L.	Pipal	Moraceae	NC	Leaf	Cu.
51	*Garuga pinnata* Roxb	Jingat	Burseraceae	NC	Leaf, Branch	Fd., Fw.
52	*Glycine max* (L.) Merr.	Bhatmas	Leguminosae	C	Grain	Fo.
53	*Hedychium densiflorum* Wall.	Besar	Zingiberaceae	C	Tuber	Fo., Me.
54	*Hordeum vulgare* L.	Jahu	Poaceae	C	Grain	Fo.
55	*Imperata cylindrical* (L.) Raeusch.	Siru	Poaceae	NC	Shoot	Ut.
56	*Ipomoea batatas* (L.) Lam.	Sakharkhand	Convolvulaceae	C	Root	Fo.
57	*Justicia adhatoda* L.	Asuro	Acanthaceae	NC	Leaves	Me.
58	*Lagenaria siceraria* (Molina.) Standl.	Lauka	Cucurbitaceae	C	Fruit	Fo., Ut.
59	*Lantana camara* L.	Banmara	Verbenaceae	NC	Shoot	Fw.
60	*Lawsonia intermis* L.	Mehendi	Lythraceae	NC	Leaves	Me.
61	*Lens culinaris* Medik.	Masuro	Leguminosae	C	Grain	Fo.
62	*Leucaena leucocephala* (Lam.) de Wit.	Ipil/ Epilepil	Leguminosae	C	Leaf,Branches,Stem	Fd., Fw., Tm
63	*Linum usitatissimum* L.	Arsi	Linaceae	C	Seed, Husk (Testa)	Fo., Me.
64	*Litsea monopetala* (Roxb.) Pers.	Kutmiro	Lauraceae	NC	leaf	Fd.
65	*Luffa cylindrical* (L.) M.Roem.	Ghiraula	Cucurbitaceae	C	Fruit	Fo.
66	*Mangifera indica* L.	Aap	Anacardiaceae	C	Fruit	Fo.
67	*Mentha spicata* L.	Pudina	Lamiaceae	C	Leaf	Me.
68	*Morus australis*Poir.	Kimbu	Moraceae	C	Fruit	Fo.
69	*Murraya koenigii* (L.) Spreng.	Ban Bakaino	Rutaceae	NC	Leaves	Fd.
70	*Musa paradisiaca* L.	Kera	Musaceae	C	Fruit, Root	Fo., Me.
71	*Nasturtium officinale* R.Br.	Simsag	Brassicaceae	NC	Shoot	Wv.
72	*Ocimum basilicum* L.	Babari	Lamiaceae	NC	Flowering twig	Cu.
73	*Ocimum tenuiflorum* L	Tulsi	Lamiaceae	C	Leaf twig	Me.
74	*Opuntia monacantha* (Willd.) Haw.	Seuri	Cactaceae	NC	Latex	Me.
75	*Oryza sativa* L.	Dhan	Poaceae	C	Grain, Shoot, Husk	Fo., Fd., Fw., Ut., Cu.
76	*Pennisetum alopecuroides* (L.) Spreng.	Napear	Poaceae	NC	Shoot	Fd.
77	*Phoenix dactylifera* L.	Khajuri	Arecaceae	NC	Fruit	Fo.
78	*Phyllanthus emblica* L.	Amala	Phyllanthaceae	NC	Fruit	Fo.

S.N.	Botanical Name	Vernacular Name	Family	Nature	Part Used	Used For
79	*Pisum sativum* L.	Kerau	Leguminosae	C	Grain, Leaf twig	Fo.
80	*Plumbago zeylanica* L.	Cheet/ Chito	Plumbaginaceae	NC	Root	Me.
81	*Pogostemon cablin* (Blanco) Benth.	Rudelo	Lamiaceae	NC	Leaf twig	Me.
82	*Premna serratifolia* L.	Agnimantha	Lamiaceae	NC	Stem	Cu.
83	*Prunus persica* (L.) Batsch	Aaru	Rosaceae	C	Fruit	Fo.
84	*Psidium guajava* L.	Belauti/ Amba	Myrtaceae	C	Fruit	Fo.
85	*Punica granatum* L.	Anar	Lythraceae	C	Fruit	Fo.
86	*Pyrus bourgaeana* Decene.	Naspati/ Naaspaati	Rosaceae	C	Fruit	Fo.
87	*Raphanus raphanistrum* subsp. *sativus* (L.) Domin	Mula	Brassicaceae	C	Leaves, Root	Fo.
89	*Saccharum bengalense* Retz.	Bankash	Poaceae	NC	Shoot	Ut.
90	*Saccharum officinarum* L.	Ukhu	Poaceae	C	Shoot	Fo.
91	*Saccharum spontaneum* L.	Kans	Poaceae	NC	Shoot	Ut.
88	*Semecarpus anacardium* L.f.	Bhalayo	Anacardiaceae	NC	Fruit, Leaf	Fo., Cu.
92	*Shorea robusta* Gaertn.	Saal	Dipterocarpaceae	NC	Leaf, Stem	Ut., Cu., Tm.
93	*Sida rhombifolia* L.	Bishkhapro	Malvaceae	NC	Leaves	Me.
94	*Solanum lycopersicum* L.	Tamatar	Solanaceae	C	Fruit	Fo.
95	*Solanum melongena* L.	Bhyanta	Solanaceae	C	Fruit	Fo.
96	*Solanum subinerme* Jacq	B	Solanaceae	NC	Seed, Leaf	Me.
97	*Solanum tuberosum* L.	Aalu	Solanaceae	C	Tuber	Fo.
99	*Stephania hernandifolia* (Willd.) Walp.	Ghoryaful	Menispermaceae	NC	Leaf twig	Wv.
100	*Stylosanthes guianensis* (Aubl.)Sw.	Stailo	Leguminosae	C	Shoot	Fd.
101	*Syzygium cumini* (L.) Skeels	Jamun	Myrtaceae	NC	Fruit	Fo.
102	*Tectona grandis*L.f.	Treak	Lamiaceae	C	Stem	Tm.
103	*Terminalia alata* Heyne ex Roth	Saj	Anacardiaceae	NC	Branch	Fw.
104	*Terminalia bellirica* (Gaertn.) Roxb.	Barrho	Combretaceae	NC	Fruit	Me.
105	*Terminalia chebula* Retz.	Harro	Combretaceae	NC	Fruit	Me.
106	*Themeda triandra* Forssk.	Khar	Poaceae	NC	Shoot	Fd.
107	*Tinospora sinensis* (Lour.) Merr.	Gurjo	Menispermaceae	NC	Tuber	Me.
108	*Triticum aestivum* L.	Gahu	Poaceae	C	Grain	Fo.
109	*Vicia faba* L.	Bakula Simi	Leguminosae	C	Fruit	Fo.
110	*Vigna mungo* (L.) Hepper.	Mas	Leguminosae	C	Grain	Fo.
111	*Woodfordia fruticosa* (L.) Kurtz	Dhaira	Lythraceae	NC	Leaf, Flower	Fd., Me.
112	*Zea mays* L.	Makai	Poaceae	C	Grain, Shoot, Leaf, Exocarp,Bracts	Fo., Fd., Ut., Me.,Cu.
113	*Zingiber officinale* Roscoe	Aduwa	Zingiberaceae	C	Rhizome	Me.
114	*Ziziphus jujuba* Mill.	Bayar	Rhamnaceae	NC	Fruit	Fo.

Note: Here, C= Cultivated, NC= Not cultivated, Fo.= Food, Fd.= Fodder, Me.= Medicinal Use, Fw.= Firewood, Tm.= Timber, Wv.= Wild vegetable, Ut.= Utensils making, Cu.= Cultural uses

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Documentation of Indigenous Knowledge on Plants used by Tamang Community of Nuwakot District, Central Nepal

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Abstract

The present work documents 45 plant species belonging to 36 genera of 24 families used by the Tamang community of Kispang Rural Municipality of Nuwakot district. The Tamang people were found dependent on the plant resources to fulfill their basic requirements. Primary data collection methods during field visit included semi-structured interview with local knowledgeble people of the community. The information collected included local name of plants, uses, form of use, and parts used. The highest numbers of plant (32 species) were used as edible and medicine followed by others. The most commonly used plant part was leaves (26 species). This study revealed that the Tamang community of study site has good indigenous knowledge of using plants for various purposes.

Keywords: Ethnobotany, Plant resources use, Tamang people

Introduction

Nepal being a multiethnic and multilingual country consists of 125 caste/ethnic groups. The population of Tamang is 1,539,830 which cover 5.8 percent of total population of Nepal (National Population and Housing Census [NPHC], 2011). They are one of the major ethnic groups of Nepal. The documentation of indigenous knowledge on the utilization of local plant resources by different ethnic groups or communities is one of the main objectives of ethnobotanical research (Malla & Chhetri, 2009). Plant resources can be used for various purposes such as food, fodder, fiber, firewood, timber, making tools, making household appliances, medicines, aroma, ornament, cultures, festivals etc. (Kunwar & Bussmann, 2008; Bhattarai & Acharya, 2015; Shah et al., 2017; Shah et al., 2018). The practice of using plant resources vary according to tradition, climatic conditions and vegetation type of the place.

Several studies have been conducted on medicinal plants and their traditional use in different parts of Nepal. Studies regarding the use of plants by Tamang community have also been conducted in the past (Shrestha, 1988; Tamang, 2003; Malla & Chhetri, 2009; Luitel et al., 2014). Most of the studies are done on traditional medicinal practices. Plants are used for many purposes other than medicinal (Bhattarai, 2009). Ethnobotanical study of Tamang community in Kispang Rural Municipality of Nuwakot district has remained unexplored. Kispang Rural Municipality, Manegaun being one of the oldest place where Tamang people live. Documentation of traditional knowledge is necessary before the knowledgeable generation gets completely lost (Shah et al., 2017). Ethnobotanical studies help for conservation of cultural tradition, sustainable use of plants as well as for socio-economic growth of ethnic communities (Malla & Chhetri, 2009; Mesfin et al., 2013).

This study helps to documentation of traditional knowledge and indigenous practices to use the plants in Tamang community and conserve the used parts in ethnobotanical museum & ethnobotanical garden of National Botanical Garden (NBG), Godawari.

Materials and Methods

Study Area

Nuwakot district lies between 27°54' to 27°91 N and 85°14' to 85°24' E. Its total area is about 1,121 square kilometers. The height ranges from 300 m to 5,000 m from the sea level. This study was carried out on Tamang community of Nuwakot district in Kispang VDC, Manegaun in May 2019.

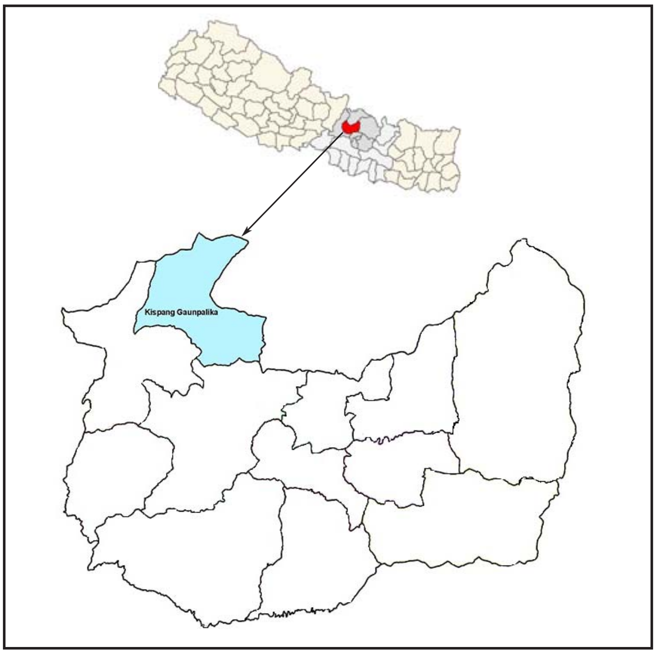

Figure 1: Map of study area (Kispang, Nuwakot)

Plant species were collected and necessary information were noted down in the field. To obtain detail information, the plant specimens collected from the field were exhibited during semi-structured interviews done with 43 respondents mostly including traditional healers and knowledgeable persons both male and female. The collected information included local name of plants, uses, form of use and parts used. The graphs were prepared by using MS-Excel.

Voucher specimens collected during field visit were preserved as herbarium in National Botanical Garden. They were identified using standard literatures (Hara et al.,1978, 1982; Hara & Williams,1979; Press et al., 2000) and comparing with specimens preserved at National Herbarium and Plant Laboratories (KATH), Godawari.

Results and Discussion

Altogether 45 plant species belonging to 24 families and 36 genera were collected and their local name, uses, used parts and utilization patterns were noted down. Most of the plants (32 species) were used for edible and medicinal followed by miscellaneous uses, fodder, firewood, religious and other purposes as shown in Figure 2. Some of the common medicinal uses were in fever, toothache, pressure, sugar, gastrointestinal, cut and wounds, eye problem, etc. (Yadav & Rajbhandary, 2016). Miscellaneous uses include making toothpaste, soap, shampoo, toothbrush of stem, tapari etc. Several species were found to be used for more than one purpose. Uses of plants along with its local names, utilization pattern and used parts are listed in Table 1.

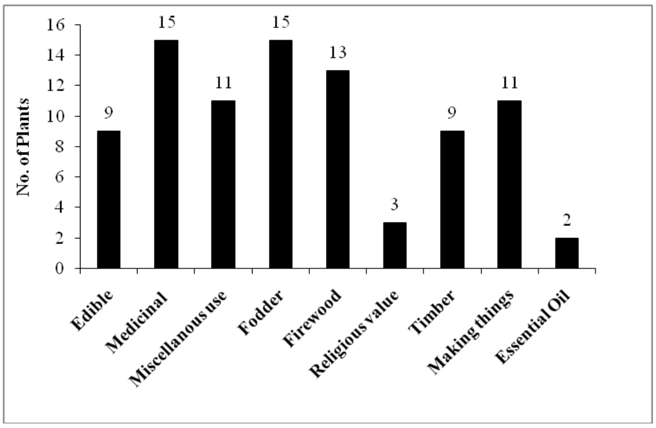

Figure 2: Number of plants used by Tamang people for various purposes

Among different plant parts, leaves of most of the plants (26 species) were used by Tamang people for various purposes followed by fruit (17 species), wood (9 species), etc. Stem, flower, whole plant, young shoot and root were also used in some cases (Figure 3).

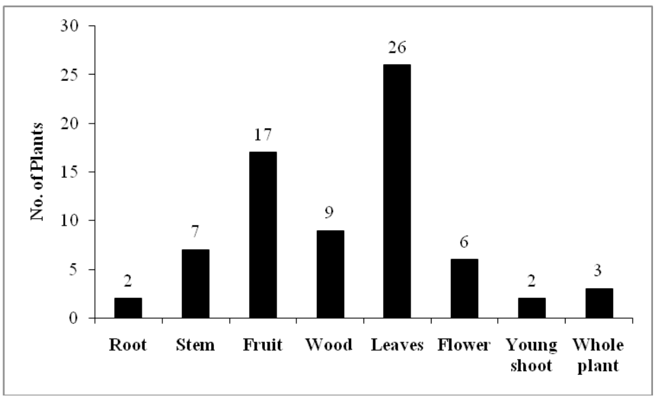

Figure 3: Number of plant parts used by Tamang people

Malla & Chhetri, (2009) and Shah & Lamichhane, (2017) also found that tribal people of Kavrepalanchowk including Tamang used plants and plant parts for various purposes in their daily life. The study by Luitel et al. (2014) found that, leaves and fruits were frequently used parts by people because they are easily available and contain high concentration of bioactive compounds. As seen from this study leaves and fruits were also used in most of the plants for edible as well as medicinal purpose. Similarly, the work conducted by Mesfin et al. (2013) in Northern Ethiopia, Yadav & Rajbhandary, (2016) in Nuwakot district and Shah et al. (2018) in Dhading district also found that leaves of plants were mostly harvested for medicinal purpose which do not affect their population and is better for sustainable utilization of plant.

Conclusion

The study showed that people of Tamang community have good indigenous knowledge of using wild plants for various purposes, most importantly as wild edible fruits and medicinal value. This study showed that such knowledge seems to be decreasing in younger generation because of global modernization. Hence, it is necessary to preserve and properly document them to keep a record of the diversified utilization of various plants for future.

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References

Bhattarai, K.R. & Acharya, S.K. (2015). Documentation of ethnobotanical knowledge of Tharupeople on the utilization of the plant resources in Gadariya and Phulwari VDCs of Kailali district, west Nepal. *Bull. Department of Plant Resources,3*, 741-50.

Bhattarai, S. (2009). *Ethnobotany and Antibacterial activities of selected medicinal plants of Nepal Himalaya*. (PhD Thesis), Central Department of Botany, Tribhuvan University, Kathmandu, Nepal.

Hara, H., Stearn, W.T. & Williams, L.H.J. (1978). *An Enumeration of the Flowering plants of Nepal*. Vol.I. London, UK: British Museum (Natural History).

Hara, H. & Williams, L.H.J. (1979). *An Enumeration of the Flowering plants of Nepal*. Vol II. London, UK: British Museum (Natural History).

Hara, H., Chater, A.O. & Williams, L.H.J. (1982). *An Enumeration of the Flowering plants of Nepal*. Vol III. London, UK: British Museum (Natural History).

Kunwar, R.M. & Bussmann, R.W. (2008). Ethnobotany in the Nepal Himalayas. *Journal of ethnobiology and ethnomedicine*, *4*, 24.

Luitel, D.R., Rokaya, M.B., Timsina, B. & Munzbergova, Z.(2014). Medicinal plants used by Tamang community in the Makwanpur district of Central Nepal. *Journal of Ethnobiology and Ethnomedicine*, *10*, 1-11.

Malla, B & Chhetri, R.B. (2009). Indigenous knowledge on ethnobotanical plants of Kavrepalanchowk district. *Kathmandu University Journal of Science, Engineering and Technology*, *5*(II), 96-109.

Mesfin, K., Tekle, G. & Tesfay, T. (2013). Ethnobotanical Study of Traditional Medicinal Plants Used by Indigenous People of Gemad District, Northern Ethiopia. *Journal of Medicinal Plants Studies*, *1*(4), 32-37.

National Population and Housing Census. (2011). National Planning Commission Secretariat, Central Bureau of Statistics: Author.

Press, J.R., Shrestha, K.K. & Sutton, D.A. (2000). Annotated Checklist of the Flowering plants of Nepal.London:The Natural History Museum.

Shah, S., & Lamichhane, D. (2017). Documentation of Indigenous Knowledge on Plants Used by

Tamang Community of Kavrepalanchok district, central Nepal. *J. Pl. Res*, *15*(1), 45-51.

Shah, S., Lamichhane, D., Panthi, Ram. & Nepali, K.B. (2018). Traditional Knowledge to Tamang Community on Utilization of Plant Resources in Dhading District, Central Nepal. *J. Pl. Res*, *16*(1), 134-140.

Shrestha, P. (1988). Contribution to the Ethnobotany of the Tamangs of Kathmandu Valley. *CNAS Journal*, *15*(2), 247-266.

Tamang, G.(2003). An Ethnobiological Study of the Tamang People. *Our Nature*, *1*, 37-41.

Yadav, S & Rajbhandary, S. (2016). Medicinal Plants Used Against Gastrointestinal Disorder by the Tamang People in Rasuwa District, Central Nepal. *Journal of Plant Science*, *10*, 19-23.

Table 1: List of plants used by Tamang people of Kispang VDC Manegaun for various purposes

S.N.	Scientific Name	Nepali Name	Tamang Name	Family	Parts Used	Form of use	Uses	Life form
1	*Achyranthes bidentata* Blume	Datiwan	Phrekphrek	Amaranthaceae	Whole plant	Twigs	Stem used as toothbrush at the time of toothache and religious value	H
2	*Ageratina adenophora* (Spreng.) R.M. King & H.Rob.	Banmara	Risaiba	Compositae	Leaves	Leaf paste	In cuts, green manure	H
3	*Albizia julibrissin* Durazz.	Siris	Tyewa	Leguminosae	Stem, Leaves		Firewood, Bananaripening	T
4	*Alnus nepalensis* D. Don	Uttis	Bomsing	Betulaceae	Wood, leaves		Fodder, timber, firewood	T
5	*Artemisia indica* Willd.	Titepati	Chyenchin	Compositae	Whole plant	Leaf juice	As incense, in cough, cuts, religious value	H
6	*Bauhinia purpurea* L.	Tanki	Konar	Leguminosae	Leaves, flower		Fodder	T
7	*Bauhinia vahlii* Wight & Arn.	Bhorla	Ghumlapte	Leguminosae	Leaves		Fodder, to make Segu	C
8	*Bauhinia variegata* L.	Koiralo	Ampu	Leguminosae	Leaves, Flower		Fodder, flowers for making pickle	T
9	*Choerospondias axillaris* (Roxb.) B.L.Burtt & A.W.Hill	Lapsi	Kalang	Anacardiaceae	Fruits and seed	Dry seed	Fruits for making pickle, seed pest for wound	T
10	*Cinnamomum camphora* (L.) J. Presl	Kapur	Sarchengen	Lauraceae	Leaves & fruits		Fodder, firewood, insecticide	T
11	*Dalbergia sissoo* DC.	Sisau	Sisoo	Leguminosae	Wood		Timber	T
12	*Diploknema butyracea* (Roxb.) H.J.Lam	Chiuri	Singmar	Sapotaceae	Fruits & seed	Dry seed	Fruits are edible, seed used to prepare banaspati ghee	T
13	*Ficus hispida* L.f.	Khasreto	Khosere	Moraceae	Leaves, stem		Fodder, Firewood	T
14	*Ficus lacor* Buch.-Ham.	Kavro	Dawah	Moraceae	Leaves, buds	Dry buds	Fodder, buds for vegetable & pickle	T
15	*Ficus racemosa* L.	Timilo	Mako	Moraceae	Leaves, fruit, buds		Fodder, fruit edible	T
16	*Ficus semicordata* Buch.-Ham. ex Sm.	Khaniyo	Kyosing	Moraceae	Leaves, fruit		Fodder, fruit edible	T
17	*Gaultheria fragrantissima* Wall.	Dhasingre	Chyanchabal	Ericaceae	Fruit, leaves	Leaf paste	Fruit edible, as medicine in scabies, extraction of oil, making ointment, toothpaste	S
18	*Jatropa curcas* L.	Sajiwan	Usure	Euphorbiaceae	Stem & Latex	Fresh latex	Toothache, wound heal	S
19	*Lagerstroemia parviflora* Roxb.	Bot Dhayero	Botadhairo	Lythraceae	Leaves, stem		Fodder, Firewood	T
20	*Leucaena leucocephala* (Lam.) de Wit	Ipil Lipil	Dyahung	Leguminosae	Leaves		Firewood	T
21	*Litchi chinensis* Sonn.	Litchi	Litchi	Sapindaceae	Fruits		Fruits edible	T
22	*Litsea monopetala* (Roxb.) Pers.	Kutmero	Chachache	Lauraceae	Leaves		Fodder	T
23	*Lyonia ovalifolia* (Wall.) Drude	Angeri	Domsin	Ericaceae	Leaves,wood	Leaf juice	In skin diseases, scabies, timber, firewood	T
24	*Magnolia champaca* (L.) Baill. ex Pierre	Champ	Chyambe	Magnoliaceae	Leaves, flower		Fodder, Religious value	T

S.N.	Scientific Name	Nepali Name	Tamang Name	Family	Parts Used	Form of use	Uses	Life form
25	*Mangifera indica* L.	Aanp	Aampa	Anacardiaceae	Fruits, Flowers	Dry fruits and leaves	Fruits edible, dry flower and fruits for pickle, religious	T
26	*Melia azedarach* L.	Bakaino	Gorkha	Meliaceae	Leaves, stem		Fodder, Firewood	T
27	*Morus alba* L.	Kimbu	Botyero	Moraceae	Leaves, fruits		Fodder, Fruits are edible,	T
28	*Oroxylum indicum* (L.)Kurz	Tatelo	Pate	Bignoniaceae	Fruit, leaves, flower	Leaf paste	In wounds, religious purpose	T
29	*Pinus roxburghii* Sarg.	Khotesalla	Thangsing	Pinaceae	Wood, stem		Timber, firewood	T
30	*Potentilla fulgens* Diels	Bajradanti	Kripangdu	Rosaceae	Root	Dry root	Fever and headache	H
31	*Prunus cerasoides* Buch.-Ham. ex D.Don	Painyu	Pyursing	Rosaceae	Fruit	Fruit bark cooked pest	Fruit edible, in sprain, as anthelminthic	T
32	*Prunus persica* (L.) Batsch	Aaru	Khale	Rosaceae	Fruit		Fruit edible	T
33	*Psidium guajava* L.	Amba	Ambaru	Myrtaceae	Leaves & Fruits		Fruits are edible, leaf extract for cough & diarrhea.	T
34	*Quercus semecarpifolia* Sm.	Khasru	Yemen	Fagaceae	Wood, Leaves		Timber, for carpentry, firewood, fodder	T
35	*Rhododendron arboreum* Sm.	Laligurans	Paramendo	Ericaceae	Stem, flower	Flower juice	Firewood, as medicine in neck pain, flowers are pickled	T
36	*Rubus ellipticus* Sm.	Ainselu	Polang	Rosaceae	Fruit, root		Fruit edible, making toothpaste, headache	S
37	*Sapindus mukorossi* Gaertn.	Rttha	Lyumdang	Sapindaceae	Fruits	Dry Fruits	As soap, insecticides	T
38	*Schima wallichii* (DC.)Korth.	Chilaune	Kyasing	Theaceae	Wood, Leaves		Firewood, Timber, manure	T
39	*Shorea robusta* Gaertn.	Saal	Jesing	Dipterocarpaceae	Leaves and wood		Timber, fodder, leaves for making plate	T
40	*Smilax aspera* L.	Kukurdino	Nagikre	Liliaceae	Stem, Leaves		Making dhyangro, fodder	C
41	*Spondias pinnata* (L.f.) Kurz	Amaro	Chyapang	Anacardiaceae	Fruits and Stem		Firewood and pickle	T
42	*Terminalia alata* B. Heyne ex Roth	Saaj	Dharsing	Combretaceae	Wood		Timber	T
43	*Terminalia bellirica* (Gaertn.) Roxb.	Barro	Barba	Combretaceae	Fruits	Dry fruit	Throat pain	T
44	*Terminalia chebula* Retz.	Harro	Arba	Combretaceae	Fruits, Leaves, Wood	Dry fruit	Timber, Fodder, fruits for throat pain	T
45	*Toona ciliata* M.Roem.	Tooni	Kyabai	Meliaceae	Wood		In carving, firewood	T

Note: H= Herb, S= Shrub, T=Tree, C= Climber

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Folklore Medicinal Plants Used Against Typhoid and Fever in Lwangghalel, Kaski District, Central Nepal

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Abstract

Typhoid is an infectious contagious disease of concern throughout the developing nations around the world.Different types of plant species are used traditionally against typhoid and fever by ethnic peoples of Lwangghalel, Kaski district, central Nepal. This study aims to document traditional medicinal plants used traditionally by local indigenous people and traditional healers to treat typhoid and fever. Group discussions, forest walk and in situ individual interviews were part of the methodology using open-ended semi-structured questionnaires. Thirty plants were cited by elder people and traditional healers to treat typhoid and fever. Most of them were used in the form of decoction taken orally. The most frequently utilized medicinal plant parts were root (33.33%) and bark (16.67%) followed by whole plant (13.33%), shoot (13.33%), rhizome (10%), fruit (10%) and tuber (3.33%). Herbs (73%) were the primary source of medicine, followed by shrubs (10%) and trees (17%). Knowledge about medicinal plants and its practices existed only among elder people and traditional healers. Further detail documentation with involvement of local stakeholder is important, so that it can be accessible to a large number of populace.

Keywords: Antipyretic, Ethnobotany, Herbal healers, Medicine,*Salmonella enterica,*Traditional knowledge

Introduction

Humans have been using plant resources and its products to fulfil multifarious daily requirements since time immemorial. Plant and plant products enhanced human culture earlier than the domestication of agriculture and fire (Khadka, 2019). Typhoid is an infectious contagious disease of concern throughout the developing world. One of the major causes of typhoid is *Salmonella enterica* serotype typhi which continues to be major public health problem in much of the developing world, especially in Asia including Nepal (Parry et al., 2002; Lewis et al., 2005). Fever represents a complex adaptive response of the host to various immune challenges whether infectious or non-infectious (Ogoina, 2011). Fever is a regulated rise in body temperature above normal daily fluctuations occurring in conjunction with an elevated thermoregulatory set point (Mackowiak, 2000; Ogoina, 2011; Mabey & Whitty, 2012). Outbreaks of drug-resistant typhoid fever and other fever have been recorded in different Asian countries (Mermin et al., 1999; Lewis et al., 2005), so traditional medicinal plants can be used as potential alternative to find new drugs against typhoid and fever.It is a type of enteric fever, known as Myadhe jaro in Nepali. The term “ethnobotany” was first given by John Harshberger in 1896 though the history of the field began long before that. Ethnobotany is the study of relationship between aboriginal society and their plant environment (Schults, 1962). It reflects the relation between plant and people. Ethnobotany is a part of traditional knowledge which is passed from generation to generation orally.

Plant and people interaction among indigenous inhabitants around the world has lured the attention of natural resources researchers to add a noble compound for the collective benefit of modern humans (Khadka, 2019). Ethnobotany uses both the anthropological and botanical approaches to understand the hidden knowledge system between plants and indigenous communities (Ford, 1978; Davis, 1995). Ethnomedicinal studies are significant value to discover contemporary drugs from indigenous medicinal plant resources (Umair et al., 2017). Traditional medicinal plants provide healthcare for up to 80% of the world’s population (Saslis-Lagoudakis et al., 2014). This study aims to document traditional medicinal plants used traditionally by local indigenous people and traditional healers to treat typhoid and fever.

In Nepal, the concept of ethnomedicine has been developed since the late 19th century (1885-1901 A.D). Ethnobotanical study started in 1955 by Banerji with the publication of paper on medicinal and food plants of eastern Nepal (Manandhar, 2002). The first book "Chandra-Nighantu" regarding medicinal plants was published by the Royal Nepal Academy in 1969 (2025 B.S.). Later, a number of ethnobotanical studies on different ethnic groups of Nepal have been carried out by different botanist and other researchers. Some major studies worked out were by Pandey (1964), Adhikari & Shakya (1977), Ghimire et al., (2000), Lama et al., (2001), Rajbhandari (2001), Manandhar (2002), Shrestha & Dhillion (2003), Mahato & Chaudhary (2005), Malla & Chhetri (2009), Singhet al., (2012), Panthi & Singh (2013) and Kunwar et al., (2013; 2014).

Materials and Methods

Study area

Present research work was carried out in Lwangghalel, Machhapuchhre Rural Municipality-8 and 9, Kaski (Figure 1) to assess the knowledge on traditional medicinal plants used for typhoid and fever. Lwangghalel lies between 28°13' N to 28°47' N latitudes and 83°84' E to 83°94' E longitudes with an elevation range of 1100-5554 m asl upto the tip of Mardi Himal.

According to Central Bureau of Statistics [CBS] (2017), the total area of Lwangghalel is 151.37 square kilometre (Sq km) with 78.84 sq km of forest area which is 52.08 % of total area. Climate is warm-temperate at lower elevation and cool-temperate to alpine at higher elevation.The mean annual precipitation recorded in the nearest Lumle station for the last 30 years was 459.48 mm (range: 462.86 mm in 1988 to 393.51 mm in 2017) (Department of Hydrology and Meterology [DHM], 2019).

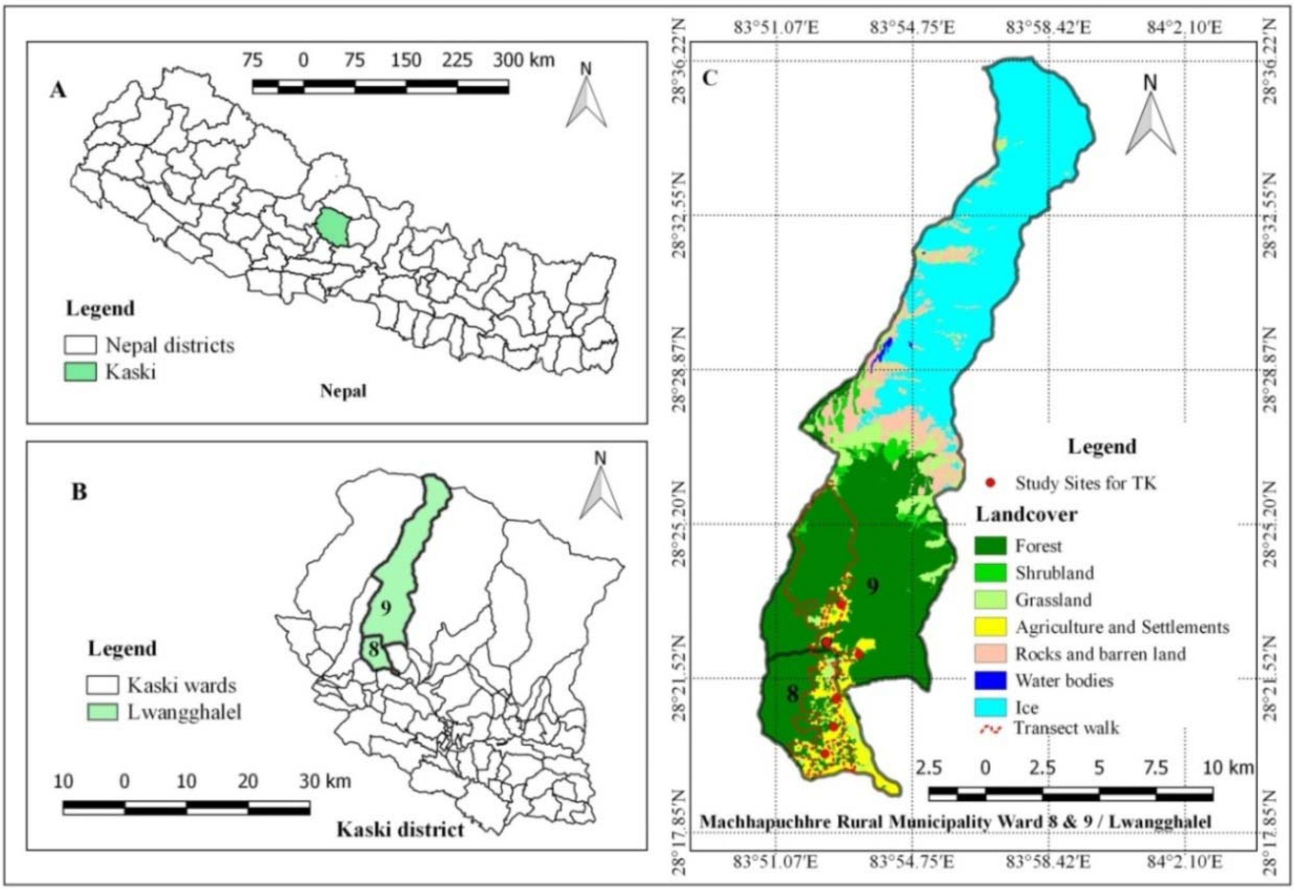

Figure 1: Map of Study Area (A) Nepal, (B) Kaski district and (C) Machhapuchhre Rural Municipality(MRM) ward 8 and 9 or Lwangghalel (Source for landcover map: ICIMOD 2010).

Sampling

The study was done in Machhapuchhre Rural Municipality (MRM), Kaski district covering rural and remote ten villages (Sidhing, Ghalel, Kalimati, Lwang, Koleli, Takru, Kuibang, Saitighatta, Lumre and Idhikhola). Since the area lies within Annapurna Conservation Area Project (ACAP), collection of rare and medicinal plants is restricted. The total population of study area was 4,211 dominated by Brahmin and Chhetri (38.26%), followed by Gurung (16.33%), Kami (15.48%), Tamang (11.10%), Damai (6.58%), others (6.36%) and Magar (5.89%) (CBS, 2011). Indigenous local people have limited access to modern health serviceshaving only one primary health center and seven health posts(CBS, 2017). So, local people rely on traditional system of medicine for their basic health care needs.

Data collection and analysis

The study area was visited from October 2018 to July 2019. A total of five field visits were made in each study site.For ethnobotanical study, both anthropological approach and ethnobotanical approaches (Ford, 1978; Davis, 1995) were followed. Group discussions, focus group discussions, transect walk and individual interviews were the part of the methodology. Open-ended semi-structured questionnaire were used for the interviews (Martin, 1995; Cunningham, 2001). Nepali and local languages were used for interviews as far as possible. Before taking interviews permission was taken either from the head of the ethnic communities or local level stakeholders. Their rights on their knowledge and natural resources were not violated. As an anthropological approach individual interviews, three focus group discussions and two group discussions were done. Questionnaires were explained in Nepali and local dialects. During the survey, a total of 65 individual including Lamas and Jhakri (9), Vaidhya (4), Pujari (8), community leader (6) and a local herbalist (13) were reached out for study and only 59 allowed us to interview further. Similarly, as an ethnobotanical approach different plant specimens cited by local indigenous peoples were collected. Ethnobotanical data along with its botanical name, voucher number, family, habitat, local name, parts used and modes of administration were entered in MS Excel sheet. The collected plant specimens were identified with the help of relevant literature (Stainton, 1987 and 1988; Press et al.,2000) and deposited at Tribhuvan University Central Herbarium (TUCH), Central Department of Botany, Tribhuvan University, Kirtipur, Kathmandu.But the rare and restricted plants were only photographed and identified during transect walk.

Results and Discussion

Diversity and uses of medicinal plants

In this study, total 30 plants belonging to 27 families and 29 genera were found to have medicinal use for typhoid and fever. Out of 27 families, Gentianaceae, Roasaceae and Rubiaceae each has 2 species and other families like Amaranthaceae, Asparagaceae, Asteraceae, Berberidaceae, Fagaceae, Malvaceae, Solanaceae and Zingiberaceae each has 1 species. Most frequently cited medicinal plants to treat typhoid and fever by local indigenous people of Lwangghalel were *Aconitum gammiei*, *Neopicrorhiza scrophulariiflora*, *Swertia chirayita*, *Mussaenda frondosa*, *Rubus ellipticus* and *Iris domestica* as reflected in Figure 2. Among 30 reported medicinal plants 8 were used against typhoid, 10 against fever and 12 against both typhoid and fever. *Abelmoschus manihot*, *Achyranthes bidentata*, *Berberis aristata*, *Eurya acuminata*, *Imperata cylindrical*, *Oxalis corniculata and Rubus ellipticus* were preferred in typhoid known as *kupath* among local indigenous communities of Lwangghalel. The scientific name, voucher number, family name, local name, parts used, habitat and preparation of the medicinal plants are summarized in Table 1.

Plant parts used and their growth forms

The most common plant parts used was root (10 species), followed by bark (5 species), shoot and whole plants each (4 species), rhizome and fruits each (3 species) and tuber (1 species) as reflected in Figure 3. Among different plant parts used, roots were preferred as it generally contains a greater amount of active compounds in comparison with

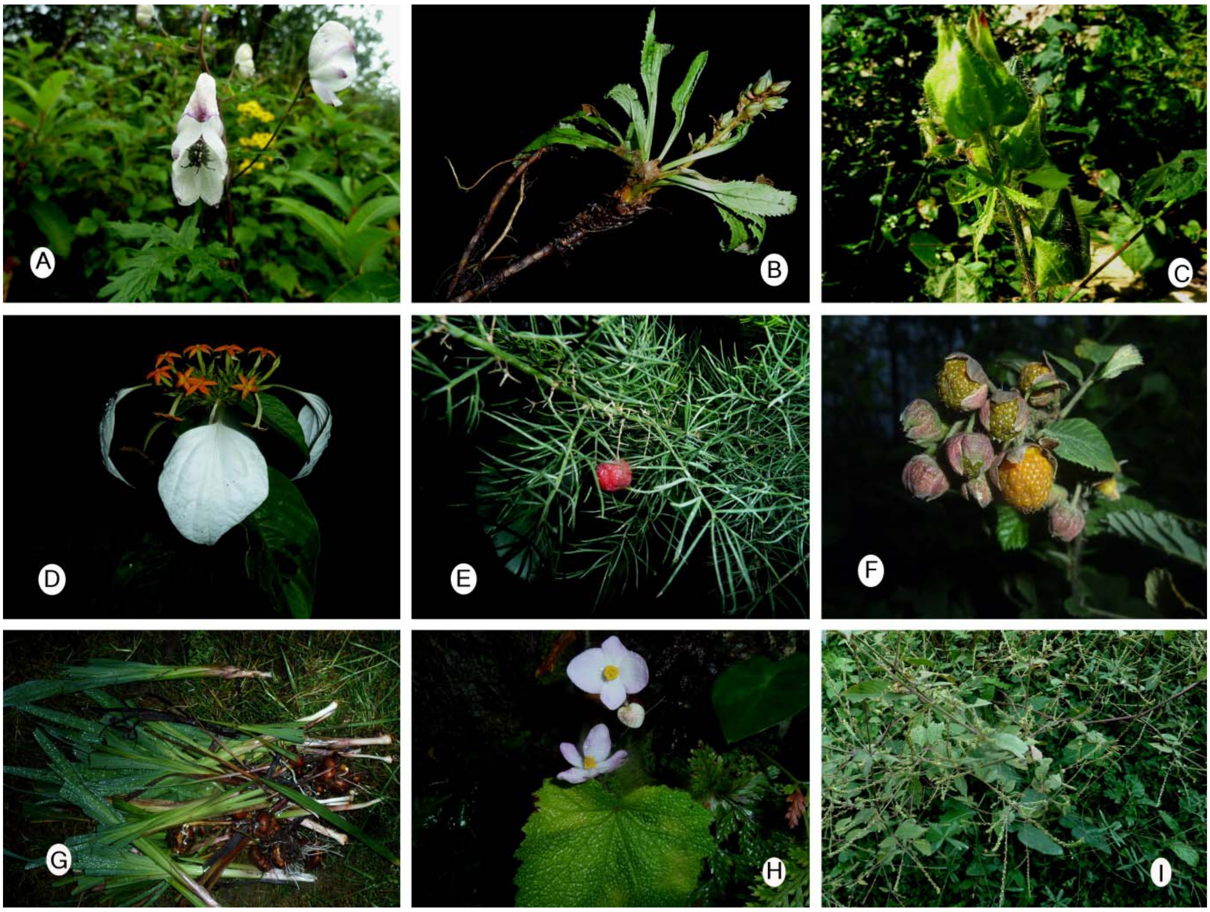

Figure 2: More frequently used medicinal plants to treat typhoid and fever A) *Aconitum gammiei* Stapf , B) *Neopicrorhiza scrophulariiflora* (Pennell) D.Y.Hong, C) *Abelmoschus manihot* (L.) Medik, D) *Mussaenda frondosa* L., E) *Asparagus racemosus* Willd., F) *Rubus ellipticus* Sm., G) *Iris domestica* (L.) Goldblatt & Mabb., H) *Begonia picta* Sm. and I) *Achyranthes bidentata* Blume

other parts (Bhattarai et al., 2006). Information on plant parts used might be beneficial for choosing these parts for further research on bioprospecting and phytochemicals determination. But the unsustainable and massive collections of roots, rhizomes and other important plant parts could lead to complete destruction and decline of the plant from its natural habitats (Ghimire et al., 2008).

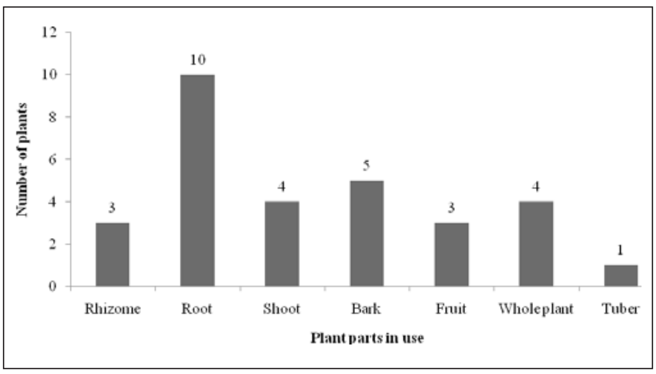

Figure 3: Medicinal plant parts preferred by local people

The total plants cited by local indigenous people were grouped into herb, shrub and tree based on their habit. Herb species were found dominant with 22 (73 %) species followed by tree 5 (17%) and shrub 10 (3%) species as demonstrated in Figure 4.

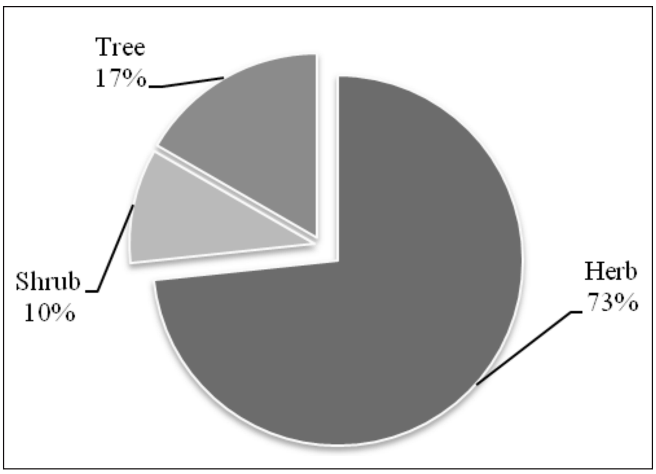

Figure 4: Growth form of medicinal plants used by local people

Herbs like *Aconitum gammiei*, *Berberis aristata*, *Nardostachys jatamansi* and *Neopicrorhiza scrophulariiflora* have habitat far from the human settlements above 2000 m elevation from sea level were collected in harvesting season. Herb diversity is dominant and increases with elevation in Lwangghalel (Khadka, 2019), easy to harvest and use so herbs growth forms were more preferred than others.

Dosage forms and routes of administration

Medicinal plants were used in the decoction, infusion, juice and powder form. Few plants were taken in chewable forms. The most frequently used dosages form was found to be decoction (50%), followed by infusion and juice each (20%), chewable (6.67%) and powder (3.33%). Route of administration for all the dosage forms was oral route. Most of the medication was prepared freshly when needed either from fresh or dried plant parts. The diagrammatic representation of the dosage form is displayed in Figure 5.

Most commonly preferred dosage forms were decoction, infusion and juice. As the routes of administration was oral, dosage forms decoction and infusion with boiling process to extract phytochemicals ensures the absence microbial infection. Similarly, the use of juice is easy for oral administration. The details about the amounts and time period of medication was not disclosed by the traditional healers as it was their main occupation and were not intended to share. Also the use of wild animals body parts along with some secret ingredients were not shared due to their traditional beliefs that the medication becomes ineffective after sharing to unknown persons.

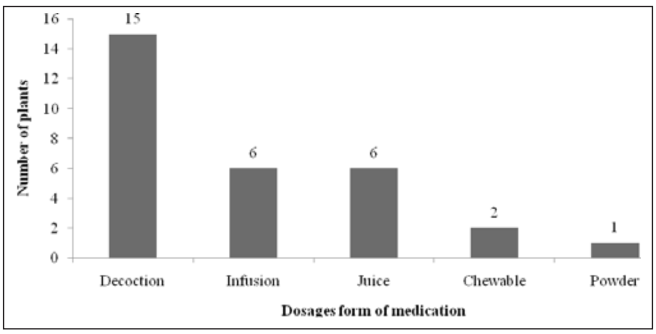

Figure 5: Dosages form of medication in the treatment of typhoid fever by local people

The 30 medicinal plants recorded from this study were found to be popular among the traditional herbalism as plants having antipyretic properties. Voucher specimens of *Nardostachys jatamansi* and *Neopicrorhiza scrophulariiflora* were not collected because they were either rare or restricted for collection or restricted to particular habitat. The indigenous local people of Lwangghalel are rich in ethnomedicinal knowledge and rely on plant based remedies for common health problems including fever, intestinal disorders, cattle diseases and other common health related issues. Typhoid fever was known as Kharo Jaro, Kam Jaro or Kupath among indigenous local peoples. The symptoms of typhoid fever were less appetite, increase in body temperature and fever, headache, abdominal pains and weakness. Typhoid fever and other fever were common during summer and rainy season due to lack of health education, poor hygiene, contaminated food and water. The combination of more than two medicinal plants listed in Table 1 had more effect than single plant. *Begonia picta* and *Oxalis corniculata* were mostly preferred as mixture with other ingredients as these plants improves the bitter taste of medicines. Most of these medicinal plants have multifarious uses to treat other diseases as *Azadirachta indica* was used as an ingredient to prepare medicine for uric acid (Khadka, 2019), whereas *Azadirachta indica* leaf was used in the treatment of typhoid fever in Nigeria (Igberaese and Ogbole, 2018). *Achyranthes bidentata* used to treat typhoid fever contains phytosterols have antibacterial activity (Devi et al., 2007). Root decoction of *Berberis aristata* used for typhoid is supported by its antibacterial activity against *Salmonella typhi* (Sohni et al., 1995, Singh et al., 2008). The reported use of *Centella asiatica*to treat typhoid and fever is justified by the antibacterial activities of its phytochemicals (Roy et al., 2013). According to Khadka (2019) *Neopicrorhiza scrophulariiflora* was used in typhoid, fever, diarrhoea, dysentery and to remove worms from digestive tracts, *Begonia picta* and *Elaeagnus parvifolia* against fever, typhoid and gastrointestinal problems, roots and rhizome of *Aconitum gammiei*, *Iris domestica*, *Mussaenda frondosa* and *Rubus ellipticus* to treat typhoid and fever in Lwangghalel, Kaski district. Among 45 medicinal plants reported by Bhattarai et al., (2013) from Panchase region, *Achyranthes aspera* and *Rubus ellipticus* were used in typhoid and *Swertia chirayita* in fever, which were similar to present findings.

Herbal healers, elderly people, traditional practitioners were rich in traditional knowledge about plant resources and its use. They still believe and are depended in herbalism. Mostly the forest dwellers, herders, lamas, jhakris and aged community leaders were knowledgeable about such resources. They were also interested to document such type of their ancestral knowledge but they want their rights and some returns too without misusing such knowledge.

Conclusion

Ethno-medicinal study focusing on typhoid fever has not been carried out in this area. Traditional medicinal plants could be alternative drug source as drug-resistance typhoid fever is common in developing world. The information and findings presented here are primarily based on interviews, group discussions, transect walk and field observation with indigenous local traditional healers, elderly peoples, lamas and social workers. Although being rich in medicinal plants only traditional practitioners and elder people had knowledge about multifarious uses of those resources. The reason behind this is that there were no health services in rural areas as nowadays, so they were totally dependent on traditional treatment for various diseases.

This study provides authentic data related to the ethnomedicinal uses of local flora to treat typhoid and fever and will lure the attention of pharmacologist, phytochemists, researchers working on antipyretic and antimicrobial drugs and traditional healers to find out the potential natural products in those plants. Further detail documentation with involvement of local stakeholder is important as some traditional medicine practitioners kept secrecy about medicinal uses of plants, so that it can be accessible to a large number of populace.

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References

Adhikari, P.M., & T.P. Shakya. (1977). Pharmacological Screening of Some Medicinal Plants of Nepal. *Journal of Nepal Pharmaceutical Association*, *5* (1), 41-50.

Banerji, M. L. (1955). Some edible and medicinal plants from East Nepal. *J. Bombay Natl. Hist. Soc.*, *53*, 153-155.

Bhattarai, K., Måren, I., & Chaudhary, R. (2013). Medicinal plant knowledge of the Panchase region in the Middle Hills of the Nepalese Himalayas. *Banko Janakari*, *21*(2), 31-39.

Bhattarai, S., Chaudhary, R.P., & Taylor, R.S. (2006). Ethnomedicinal plants used by the people of Manang district, central Nepal. *Journal of Ethnobiology and Ethnomedicine*, *2*, 41.

Central Bureau of Statistics. (2011). *Population Census of Nepal* 2011. Thapathali, Kathmandu, Nepal: Author

Central Bureau of Statistics.(2017).*Rural Municipality/Municipality's Profile of Kaski*;: Pokhara, Kaski: Author

Cunningham, A.B. (2001). *Applied Ethnobotany: People, Wild Plant Use and Conservation*. London and Sterling, VA, USA: Earthscan Publication Ltd.

Davis, E.W. (1995). Ethnobotany: an old practice, a new discipline. In: *Ethnobotany: Evolution of a Discipline* (R.E. Schultts and S.V. Reis, eds.), pp. 40-51. Oregon: Dioscoriodes Press.

Devi, P.U., Murugan, S., Suja, S., Selvi, S., Chinnaswamy, P. & Vijayanand, E. (2007). Antibacterial in vitro lipid peroxidation and phytochemical observation on *Achyranthes bidentata* Blume. *Pak. J. Nutr.*, *6*, 447-451.

Department of Hydrology and Meterology. (2019). Meterology Data Management Section, Kathmandu, Nepal: Author

Ford, R.L. (1978). *The nature and status of ethnobotany*. In Anthropological Papers (R.L. Ford, eds.), Museum of Anthropology, University of Michigan, USA.

Ghimire, S.K., Gimenez, O., Pradel, R., McKey, D., & Aumeeruddy-Thomas, Y. (2008). Demographic variation and population viability in a threatened Himalayan medicinal and aromatic herb Nardostachys grandiflora: Matrix modelling of harvesting effects in two contrasting habitats. *Journal of Applied Ecology, 45,* 41–51.

Ghimire, S.K., Shrestha, A.K., Shrestha, K.K., & Jha, P.K. (2000). Plant resources use and human impact around Royal Bardiya National Park, Nepal. *Journal of Natural History Museum,19*, 3-26.

Harshberger, J. W. (1896). The purposes of ethno-botany. *Botanical gazette*, *21*(3), 146-154.

Igberaese, P.O., & Ogbole, O.O. (2018). Ethnobotanical survey of plants used in the treatment of typhoid and its complication(s) in Esan North East local government area, Uromi, Edo state. *Nig. J. Pharm. Res*, *14*(2), 175-188.

International Centre for Integrated Mountain Development. (2010). Land cover of Nepal 2010. Kathmandu, Nepal: Author. http://rds.icimod.org/Home/DataDetail?metadataId=9224

Khadka, B. (2019). *Ethnobotany, diversity and distribution pattern of vascular plants in Lwangghalel, Kaski district, central Nepal*. (Master's Thesis), Central Department of Botany, Tribhuvan University, Kirtipur, Kathmandu, Nepal.

Kunwar, R.M., Mahat, L. Acharya, R.P.,& Bussmann, R.W. (2013). Medicinal plants, traditional medicine, markets and management in far-west Nepal. *Journal of Ethnobiology and Ethnomedicine*, *9*(1), 24-34.

Kunwar, R.M., Pandey, M.L., Kunwar, L.M.,& Bhandari, A. (2014). *Medicinal Plants and Ethnomedicine in Peril: A Case Study from Nepal Himalaya, Evidence-Based Complementary and Alternative Medicine*, http://dx.doi.org/10.1155/2014/792789.

Lama, Y.C., Ghimire, S.K.,& Aumeeruddy-Thomas, Y.(2001). *Medicinal plants of Dolpo: Amchis' knowledge and conservation*. Kathmandu, Nepal: WWF Nepal Program.

Lewis, M.D., Serichantalergs, O., Pitarangsi, C., Chuanak, N., Mason, C.J., Regmi, L.R., Pandey, P., Laskar, R., Shrestha, C.D., & Malla, S. (2005). Typhoid fever: A massive, single-point source, multidrug-resistant outbreak in Nepal. *Clinical Infectious Diseases*, *40*(4), 554-561.

Mabey, D., & Whitty, C.J.M. (2012). Approach to the febrile patient. E. Parry, R. Godfrey, D. Mabey, G. Gillm (eds.), *Principles of medicine in Africa*, 3rd ed., (pp. 191-197) UK: Cambridge University Press.

Mackowiak, P. (2000). *Temperature Regulation and the pathogenesis of fever in Principles and Practice of infectious diseases*, 5th (pp. 604-619). Philadelphia: Churchill Livingstone.

Mahato, R.B., & Chaudhary, R.P. (2005). Ethnomedicinal plants of Palpa district, Nepal. *Ethnobotany*, *17*, 152-163.

Malla, B., & Chhetri, R.B. (2009). Indigenous knowledge on ethnobotanical plants of Kavrepalanchowk district, Kathamandu University. *Journal of Science, Engineering and Technology*, *2*(5), 96-109.

Manandhar, N.P. (2002). *Plants and people of Nepal*. Portland, Oregon, USA: Timber Press

Martin, G.J. (1995). *Ethnobotany: A Methods manual*. London : Chapman and Hall.

Mermin, J.H., Villar, R., Carpenter, J., Roberts, L., Samaridden, A., Gasanova, L., Lomakina, S., Bopp, C., Hutwagner, L., Mead, P., Ross, B., & Mintz, E.D. (1999). A massive epidemic of multidrug-resistant typhoid fever in Tajikistan associated with consumption of municipal water. *J Infect Dis*, *179*, 1416-22.

Ogoina, D. (2011). Fever, fever patterns and diseases called 'fever'–a review. *Journal of infection and public health*, *4*(3), 108-124.

Pandey, P.R. (1964). Distribution of Medicinal Plants in Nepal. *Symposium on Medicinal Plants (Ceylon)*. 15-18 December.

Panthi, M.P., & Singh, A.G. (2013). Ethnobotany of Arghakanchi district, Nepal: Plants used in dermatological and cosmetic disorders. *International Journal of Applied Sciences and Biotechnolgy, 1*(2), 27-32

Parry, C.M., Hien, T.T., Dougan, G., White, N.J., & Farrar, J.J. (2002). Typhoid fever.*N Engl J Med*, *347*, 1770-82.

Press, J.R., Shrestha, K.K., & Sutton, D.A. (2000). *Annotated Checklist of the Flowering Plants of Nepal*. London, UK: The Natural History Museum.

Rajbhandari, K.R. (2001). *Ethnobotany of Nepal*.Ethnobotanical Society of Nepal (ESON), Kathmandu: Kishor Offset Press (P.) Ltd.

Roy, D.C., Barman, S.K., & Shaik, M.M. (2013). Current updates on *Centella asiatica*: phytochemistry, pharmacologyand traditional uses. *Med. Plant Res., 3,* 20-26.

Saslis-Laguodakis, C.H., Hawkins, J.A., Greenhill, S.J., Pendry, C.A., watson, M.F., Tuladhar-Douglas W., Baral, S.R., & Savolainen, V. (2014). The evolution of traditional knowledge: environment shapes medicinal plant use in Nepal. *Proceeding of the Royal Society of London B: Biological*, *281*(1780), 20132768.

Schults, R.E. (1962). The role of ethnobotanist in the search of new medicinal plants. *Lloydia*, *25*(4), 45-64.

Shrestha, P.M., & Dhillion, S.S. (2003). Medicinal plant diversity and use in the highlands of Dolakha district, Nepal. *Journal of Ethnopharmacology*, *86*(1), 81-96.

Singh, A.G., Kumar, A., & Tewari, D.D. (2012). An ethnobotanical survey of medicinal plants used in Terai forest of western Nepal. *Journal of Ethnobiology and Ethnomedicine*, *8*(1), 19.

Singh, M., Srivastava, S., & Rawat, A.K. (2008). Antimicrobial activities of Indian *Berberis* species. *Fitoterapia, 78*, 574–576.

Sohni, Y.R., Kaimal, P., & Bhatt, R.M. (1995). Prophylactic therapy of *Salmonella typhi* septicemia in mice with a traditionally prescribed crude drug formulation. *J. Ethnopharmacol., 45*, 141-147.

Stainton, J.D.A. (1987). *Concise Flowers of Himalaya*. New Delhi, India: Oxford University Press.

Stainton, J.D.A. (1988). *Flowers of the Himalaya: A supplement*. New Delhi, India : Oxford University Press,

Umair, M., Altaf, M. & Abbasi, A.M. (2017). Anethnobotanical survey of indigenous medicinal plants in Hafizabad district, Punjab-Pakistan. *Plos one*, *12*(6), e0177912.

Table 1: Enumeration of medicinal plant species of oral dosage used to treat typhoid and fever

S. N.	Botanical name	Voucher number	Family	Local name (Nepali)	Habitat	Uses	Parts used	Preparation
1	*Abelmoschus manihot* (L.) Medik.	L11	Malvaceae	Kapase	H	T	Root	Decoction
2	*Achyranthes bidentata* Blume	KL429	Amaranthaceae	Datiwan	H	T	Root	Juice
3	*Aconitum gammiei* Stapf	KL321	Ranunculaceae	Nirmasi	H	F & T	Rhizome	Decoction
4	*Asparagus racemosus* Willd.	L49	Asparagaceae	Kurilo	H	F & T	Root	Decoction
5	*Azadirachta indica* A.Juss.	KL444	Meliaceae	Nim	T	F & T	Bark	Decoction
6	*Begonia picta* Sm.	KL141	Begoniaceae	Magar kachi	H	F & T	Whole plant	Chewable
7	*Berberis aristata* DC.	KL349	Berberidaceae	Chutro	S	T	Bark	Infusion
8	*Castanopsis tribuloides* (Sm.) A.DC.	KL219	Fagaceae	Katus	T	F	Bark	Decoction
9	*Centella asiatica* (L.) Urb.	B131	Apiaceae	Ghodtapre	H	F & T	Shoot	Juice
10	*Cirsium verutum* (D. Don) Spreng.	KL449	Compositae	Thakailo	H	F	Root and shoot	Juice
11	*Curcuma angustifolia* Roxb.	KL452	Zingiberaceae	Besar/Haledo	H	F	Rhizome	Powder
12	*Elaeagnus parvifolia* Wall. ex Royle	KL401	Elaeagnaceae	Guyeli	S	F	Fruit	Chewable
13	*Erythrina stricta* Roxb.	KL466	Leguminosae	Fadelo	T	T	Bark	Decoction
14	*Eurya acuminata* DC.	L16	Pentaphyllaceae	Jhyanu	T	T	Young shoot	Juice
15	*Galium aparine* L.	KL411	Rubiaceae	Tite jhar	H	F & T	Shoot	Infusion
16	*Imperata cylindrica* (L.) Raeusch.	KL459	Poaceae	Siru ghans	H	T	Root	Infusion
17	*Iris domestica* (L.) Goldblatt & Mabb.	KL423	Iridaceae	Khadgaa dhari	H	F & T	Rhizome	Decoction
18	*Mussaenda frondosa* L.	KL247	Rubiaceae	Dhobino	H	F & T	Root	Decoction
19	*Nardostachys jatamansi* (D.Don) DC.		Caprifoliaceae	Jatamasi	H	F & T	Root	Decoction
20	*Neopicrorhiza scrophulariiflora* (Penn ell) D.Y.Hong		Plantaginaceae	Kutki	H	F & T	Root	Decoction
21	*Nephrolepis cordifolia* (L.) C. Presl	KL460	Nephrolepidaceae	Paniamala	H	F	Tuber	Juice
22	*Ocimum tenuiflorum*L.	B163	Lamiaceae	Tulasi	H	F	Shoot	Infusion
23	*Oxalis corniculata* L.	KL462	Oxalidaceae	Chariamilo	H	T	Whole plant	Decoction
24	*Physalis peruviana* L.	B164	Solanaceae	Isamgol	H	F	Fruit	Decoction
25	*Prunus persica* (L.) Batsch	KL467	Rosaceae	Aaru	T	T	Bark	Decoction
26	*Rubus ellipticus* Sm.	S133	Rosaceae	Ainselu	S	F & T	Root	Decoction
27	*Solena amplexicaulis* (Lam.) Gandhi	KL402	Cucurbitaceae	Gol kakri	H	F	Fruit	Juice
28	*Swertia chirayita* H. Karst.	B167	Gentianaceae	Chiraito	H	F & T	Whole plant	Infusion
29	*Swertia paniculata*Wall.	B118	Gentianaceae	Chiraito	H	F	Whole plant	Infusion
30	*Thysanolaena latifolia* (Roxb. ex Hornem.) Honda	KL465	Poaceae	Amriso	H	F	Root	Decoction

Note: T-Typhoid, F-Fever, H-Herb, S-Shrub, T-Tree

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Eleusine coracana (L) Gaertn (Finger millet): A Crop With Medicinal Value

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Abstract

Finger millet is one of the food crops with rich nutritional composition. It has been cultivated from 85 m to 3130 m altitudes of Nepal. A total of 121 household surveys were conducted in 11 villages of two ecological zones (Tarai /Siwalik and Mid-hills) of Central Nepal from September 2017 to March 2018 using questionnaire to document the ethno-medicinal value of finger millet. Locally finger millet is recognized as medicinal value besides food. More than 80% of local people were aware about its medicinal values and 83% of local people used it as medicine to cure the diarrhoea of cattle and 81% of people indicated it is very good for bone. The medicinal values of finger millet mentioned by the informants were compared with the published scientific facts. The comparison revealed that 15 different types of human ailments can be cured by using finger millet and its products. The human ailment categories are mostly related with lifestyle related diseases like diabetes, blood pressure, cardiovascular diseases and so on.

Keywords: Food, Human ailments, Lifestyle related diseases, Medicinal value

Introduction

Six species of millet are cultivated found in Nepal. Finger millet [*Eleusine coracana* (L.) Gaertn.], foxtail millet [*Setaria italica* (L.) P.Beauv.]. proso millet [*Panicum miliaceum* L.], barnyard millet [*Echinochloa frumentacea* Link], sorghum [*Sorghum bicolor* (L.) Moench] and pearl millet [*Pennisetum typhoides* (Burm.f.) Stapf & C.E.Hubb.Hubb. (Ghimire et al., 2017). Two wild varieties of millets *Eleusine indica* (L.) Gaertn. and *Eleusine africana* Kenn.-O'Byrne have also been reported from Nepal. The commonly called Kodo (Finger millet) is cultivated throughout the country. It is a fourth cereal crop of Nepal and second important crop of mid-hill and Himalaya. Mid-hills and Himalayas cover about 10% of total cultivated land of country and nearly 75% of finger millet cultivation prevails in mid-hill and mountain (Upreti, 2002; Ministry of Agriculture, Land Management and cooperatives [MoAC], 2017).

Finger millet has been cultivated from low land Terai region in Kachorwa village (85 m asl) of Bara district (Amgain et al., 2004) to high hill area in Borounse village (3130 m asl) of Humla district (Baniya et al., 1992). Finger millet is called "kuanna" or an unholy cereal not worth for worshipping. It as a neglected or orphan crop but provide staple food to millions of marginal community in the world and also to the population using on subsistence farming system in Nepal. It is also referred as "crop for poor" or a "famine food" (Vietnameyer et al., 1996). Finger millet grain has low infestation of pest in storage and can be stored for long periods due to its small grain size.

Finger millet is also used, for making Jadh (alcoholic liquor made from finger millet without distillation), Raksi and Tumba (alcohol made from finger millet) and animal feed. But in many parts of the country including Tarai and Siwalik regions, it is used in snacks eg. haluwa (sweet dish) and roti (bread). It is generally considered to be very nutritious rich food for pregnant women and also used for treatment of animal diarrhea (Rana et al., 2000) and for diabetic patients (Sri-Lankan Ministry of Agriculture [LMA], 2004). Finger millet has very high nutritional value particularly iron, calcium, and manganese (International Crops Research Institute for the Semi-Arid Tropics [ICRISAT], 2004). Being high nutritional value with lots of medicinal properties, there are enough opportunities of income generation through in agro-based industry (Sthapit et al., 1993).

Till date finger millet only occupies the traditional consumption pattern in Nepal. Systematic documentation of health benefit of finger millet and its products has not been done in Nepal. Beside nutrition, its plant parts have been used as folk medicine.

This study was focused on documenting opinions of small holder farmers on medicinal value of finger millet in different ecological regions in central Nepal. The finger millet is one of the main crops in mountains regions especially in remote areas. Besides, it has been used as folk medicine to treat different diseases directly or indirectly. These practices are based on knowledge transfer from generation to generation. However, finger millet grains are mostly used in preparation of traditional products like dhedo, roti, haluwa, alcoholic beverages like rakshi, chhyang, tumba, jandh which have religious and cultural importance in different ethnic communities of Nepal. Do the local people use finger millet as folk medicine? is the main research question of this study and an attempt has been made to compare the perception and experiences of local people on health benefit of finger millet with the published secondary facts. There were no previous studies on medicinal values of finger millet from Nepal. However, there are several literatures quoted many health benefits of finger millet globally.

Materials and methods

Study area

This study was conducted in eleven villages of four Palikas (municipalities) in three different districts of central Nepal. These Palikas were selected based on accessibility, cultivation of finger millet and population composition with diversity of communities. These Palikas of CHAL are important finger millet producers. These municipalities were located at different altitudes to represent different ecological regions. Two rural municipalities (Hupsekot and Bungdi Kaali) lie at less 500 m in Nawalparasi and two municipalities (Waling in Syangja and Besishahar in Lamjung) at 1000 - 2000 m. Majority households of the selected villages in Nawalparasi, Syangja and Lamjung districts cultivate finger millet for their food requirements. Farming system was traditional type integrated with animal husbandry.

Questionnaire preparation and household survey

The semi-structured questionnaire was prepared to gather the medicinal value of finger millet and survey

Table 1: Details of respondent in two ecological regions of Central Nepal, (Figures in parentheses indicates number of respondents above 70 years)

S.N.	Regions	Village Name	Districts	Total House holds	Total no. of elder (70 yrs) people in village		
					Total	Male	Female
1	Siwalik	Sankhadev	Hupesekot RM-6, Nawalparasi	151	25(6)	11(2)	14(4)
2		Machedi	Bungdikali RM-1, Nawalparasi	80	26(16)	10(5)	16(11)
3		Dhobaji	Hupesekot RM-1, Nawalparasi	42	13(13)	6(6)	7(7)
4	Mid-hills	Tamadi	Waling-14,Syangja	42	14(11)	5(4)	9(7)
5		Dhikidanda	Waling-12,Syangja	15	6(5)	3(3)	3(2)
6		Kapase	Waling-14,Syangja	55	21(16)	9(7)	12(9)
7		Gaire- Kattike	Waling-12,Syangja	18	6(6)	3(3)	3(3)
8		Dobate	Waling-12,Syangja	17	8(8)	5(5)	3(3)
9		Sirsekot	Waling-12,Syangja	37	15(12)	8(8)	7(4)
10		Kokhe	Waling-12,Syangja	45	20(12)	11(8)	9(4)
11		Chandidanda-baseni	Beshishahar-8,Lamjung	51	19(16)	10(10)	9(6)
	Total				173(121)	81(61)	92(60)

was conducted from September 2017 to March 2018. A total of 121 individuals (61 male and 60 female) were selected for face-to-face interviews. The respondents were mostly aged people (>70 years), permanent resident and small holder farmers following traditional system of agriculture. Respondents were Brahmin (23), Magar (35), Gurung (39) and Dalit (5) from Mid-hill (Syangja and Lamjung districts) and 13 Brahmin and 5 Magar from Nawalparasi. The questionnaire had two major sections: (i) detailed information of the informant including socio-economic and agricultural practices and (ii) crop calendar of finger millet and its uses including medicinal value, local landraces, type of diseases, mode of uses, duration of uses etc.

Secondary literature

We compiled the medicinal values of finger millet from international and national journals, books, proceedings and internet base sources. The medicinal values were compiled in to 15 different categories based on human ailments.

Results and Discussion

Perceptions on medicinal values of finger millet

Eighty percent local people knew the medicinal value of finger millet from Nawalparasi (Siwalik region) and more than 81% people have mentioned medicinal value of this crop in mid hills of CHAL. However, 100% respondents were aware about medicinal value of finger millet in Sakhadev, Tamadi and Dhikidanda villages. Only fifty percent respondent from Dobate and Gaire Kattike of Syangja mentioned medicinal values (Table 2).

The people of Nepal have rich ethno-botanical knowledge on plant resources. Wild medicinal herbs have been used in traditional medicinal practice; however, several crops species including finger millet have been used to treat human and animal ailments.

In an average more than 80% people in CHAL knew about the medicinal values of finger millet (Table 2). People realize that finger millet as food is much nutritious and good for hard working people. Though several respondent mentioned medicinal values of finger millet. However, few statements are mentioned here to quote the value of this crop as medicine and other in the villages. Kaumaya Gurung (87 F) from Kokhe village of Syangja feels appetite just after having rice meal but she do not want any more food throughout the day after having dhido (a product of finger millet). She could not swell the rice well but can easily eat dhido and roti of finger millet. She told that food habitat of her family has been changed from finger millet and maize to rice culture and also mentioned that her health and

Table 2: Perception of respondents on medicinal values of finger millet (figures in parentheses is percentage)

S.N.	Regions	Village Name	Total no. of respondents	Medicinal value of finger millet		
			Total	Yes	No	Don't know
1	Siwalik	Sankhadev	6	6(100)	0	0
2		Machedi	16	10(62.5)	2(12.5)	4(25)
3		Dhobaji	13	12(92.3)	0	1(7.7)
	Total in Siwalik		35	28(80)	2(5.7)	5(14.28)
4	Mid-hills	Tamadi	11	11(100)	0	0
5		Dhikidanda	5	5(100)	0	0
6		Kapase	16	13(81.25)	1(6.2)	2(12.5)
7		Gaire- Kattike	6	3(50)	0	3(50)
8		Dobate	8	4(50)	2(25)	2(25)
9		Sirsekot	12	10(83.34)	0	2(16.67)
10		Kokhe	12	9(75)	1(8.3)	2(16.67)
11		Chandidanda-baseni	16	15(93.75)	0	1(6.25)
Total in Mid-hills			86	70(81.4)	4(4.6)	12(13.95)
Grand Total			121	98(81)	6(4.95)	17(14.04)

physical is still strong only due to finger millet food during her life. Similarly, Dorna Kandel (78 M) from Machedi village of Nawalparasi would be happier when main food in his kitchen is dhido or roti of finger millet. He added the product of finger millet is real food for us who has been still active physically. He also mentioned that he has not taken a single tablet of medicine for headache throughout his life which may be due to benefit of finger millet diet. Chandri Thapa (98 F) from Kapase village of Syangja mentioned that finger millet is a real food for villagers who has been working daily in the farm. She added that it is not only food but also life supporting medicine. She remembered that they spend about ½ tons of finger millet to produce liquor (kodo ko raksi) yearly which is a strong medicine for people and has to take about 60 ml daily at the time of sleep in night. Therefore, the views and experience of local people from different villages of central Nepal reveals the medicinal value of finger millet.

Ailments wise perceptions

A total of seven different types of ailments have been used by local people to cure by finger millet and its production in their villages. More than 83% respondent mentioned that this crop is very good to control diarrhea of castles and 81% mentioned good food for bone of human beings. Respondents could not identify the scientific detail of diseases and its therapeutic value and mechanism. They commonly mentioned that finger millet is good for health and makes the man strong without diseases. The common perception regarding the therapeutic value includes, it makes bone strong, increases stamina of people, good for health, help to heal wound, joins the fractured bones, control diarrhea in human as well as in cattle, good for common cold and cough, etc. Overall perception regarding the health benefit of finger millet with percent is presented in Table 3.

The local perceptions on medicinal value (categorical) of finger millet (Table-3) can be summarized with possible logic in the following ways.

1. Consumption of finger millet regularly, protect from malnutrition, degenerative diseases and premature aging because of the nutraceutical importance of finger millet as it contains high calcium (0.38%), protein (6%–13%), dietary fiber (18%), carbohydrates (65%–75%), minerals (2.5%–3.5%), phytates (0.48%), tannins (0.61%), phenolic compounds (0.3–3%) and trypsin inhibitory factors (Chandra et al., 2016).
2. Strengthen the bones of human due to good source of natural calcium (0.38%) (Chandra et al., 2016) and reduces the risk of fracture and prevent from osteoporosis. Join the fracture of bones due to high natural calcium.
3. Due to antioxidant property of finger millet, good for late aging and metabolic diseases.
4. Finger millet diet has lower glycemic responses (lower ability to increase blood sugar level) which lower digestibility and absorption of starch as a result person having finger millet diet can avoid appetite.
5. Nature of phytochemicals of finger millet controls the blood sugar level which is beneficial for diabetic patients.
6. Due to high natural iron and calcium content in finger millet, it is good food for pregnant and delivered women as well as protect from anemia.

Table 3: Total number of perceptions of respondent on medicinal values (categorical) of finger millet out of 121 in CHAL

S.N.	Health benefit of finger millet (Ailments)	Total no of respondent
1	Makes bone strong	98 (81%)
2	Finger millet consumption people are resistant to common diseases	67(55.3%)
3	Flour controls the diarrhea in goat, lamb, sheep, calf of caw, buffaloes	101(83.4%
4	Wound healing property	81(66.7%)
5	Flour soup is good for children during cough and cold	34(28%)
6	Prevent premature aging	29(24%)
7	Good for pregnant and delivered women	73(60.3%)

Table 4: Medicinal value of finger millet quoted in different published literatures

S. N.	Uses	Reference
1	Antioxidant (Due to high total phenolic and flavonoids, level of enzymatic and non enzymatic antioxidant)	Sripriya, et al., 1996; Hedge, et al., 2005a, Viswanath, et al., 2009; Chandrasekara & Shahidi, 2010; Veenashri & Muralikrishna, 2011; Devi et al., 2011; Amadou et al., 2011; Mohamed et al., 2011
2	Antimicrobial activities (Polyphenol extract from finger millet flour active against *Bacillus cereus*, *Aspergillus niger* and Fermented finger millet extract-suppress growth of *Salmonella* sp., *Escherichia coli* Inhibition of *Salmonella typhimurium* and *Escherichia coli* by fermented flour of finger millet Protocatechuic, caffeic, gallic, parahydroxy benzoic acid, polyphenols, and quercitin from finger millet inhibited the growth of several pathogenic bacteria	Antony, et al., 1998; Viswanath, et al., 2009; Usha et al., 1998; Chethan and Malleshi, 2007
3	Anti-ulcerative property (Diet with finger millet prevent mucosal ulceration	Tovey, et al., 1975; Chandra et al., 2016
4	Wound healing property	Rajesekaran et al., 2004; Hedge et al., 2005b
5	Diarrhea control (fermented drink by lactic acid bacteria used as therapeutic agent against diarrhea)	Manzoni et al., 1999; Venkateswaran & Vijayalakshi, 2010
6	Improve haemoglobin (rich in natural source of iron- improve haemoglobin in blood)	Tatala et al., 2007
7	Protection from diseases Cancer, Diabetes, heart disease, hypertension, metabolic syndrome, and Parkinson's disease (due to phyto-chemical properties of finger millet) Prevent cardiovascular diseases by reducing plasma tri-glycerides, Reducing tumor incidence, lowering blood pressure, risk of heart disease	Srivastava & Sharma, 2012; Lee et al., 2010; Chandrasekara & Shahidi, 2011; Taylor & Emmambux, 2008; Taylor, et al., 2006; Gull et al., 2015; Manach et al., 2005; Scalbert et al.,2005; Saleh et al., 2013; Gupta et al. 2012
8	Good for abdominal patients (millet do not contain gluten therefore good for abdomen)	Chandrasekar & Shahidi, 2010
9	Delay ageing (By reducing glycosylation of body proteins)	Doraiswamy et al., 1969.
10	Checks constipation, high blood cholesterol formation and intestinal cancer due to high fiber content in grain,	Usha, 2004; Enas et al., 2003
11	Folk medicine for leprosy, liver diseases	Watt & Breyer-Brandwijk, 1962
12	Folk medicine for Measles, Pleurisy, Pneumonia, Small pox	Duke & Wain, 1981
13	Anti-inflammatory, antiviral	Chethan & Malleshi, 2007
14	Prevention and management of diabetes	American Diabetes Association 2005; Shobana et al., 2009; Kim et al., 2011
15	Miscellaneous uses from Ayurvedic perspective (Promotes weight loss, healthy choice for vegans, mental relaxation, lowers triglycerides, improve lactation, child growth and weaning, promote hair growth, women friendly, geriatric tonic, gluten free food, lower the risk of gall stones and fights diseases.	Chandra et al., 2016

Medicinal value of finger millet in literatures

Based upon ethnobotanical practices, finger millet was effective against 15 types of human ailments, viz. antioxidant; antimicrobial activities; anti-ulcerative ; wound healing property; diarrhea control; improve haemoglobin; Inhibition of pathogenic bacterial strains; protection from non communicable diseases; delay ageing; checks constipation; leprosy, pneumonia, measles; anti-inflammatory and antiviral; prevent cardio-vascular diseases; prevent diabetics; miscellaneous (Table 4**)**.

Health benefit of finger millet such as prevention of diabetes (Kim et al., 2011), cardiovascular diseases (Saleh et al., 2013), lowering blood pressure (Gupta et al. 2012), antioxidant, anti-ulcerative, anti-microbial, anti- inflammatory, anti-viral, anti- cancer has also been documented as therapeutic property of finger millet. Its medicinal properties are also valued in Ayurvedic medicine though it is known as Kuanna.

Finger millet is still second main food stuff in large parts of mid-hills and mountains region, but consumption rate is decreasing due to changing food habit of people. Health benefit of finger millet should be communicated to people in urban area, so that finger millet would become popular. Finger millet can serve as an ideal crop in term of nutrition and medicinal benefits to people of Nepal.

Conclusion

Finger millet could be an idol crop of Nepal due to rich in nutrition as well as medicinal value for several lifestyle related diseases like diabetes, cardio vascular disease. Locally finger millet is recognized as medicinal value besides food. More than 80% of local people were aware about its medicinal values and more than 83% of local people used it as medicine to cure the diarrhoea of cattle and 81% of people indicated it is very good for bone.

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References

Amadou, I., Amza, T., Shi, Y.H.& Le, G.W.(2011). Chemical analysis and antioxidant properties of foxtail millet bran extracts. *Songklanakarin J Sci Technol*, *33*(5), 509–15.

American Diabetes Association. (2005). Diagnosis and classification of diabetes mellitus. D*iabet Care*, *28*, 37–42.

Amgai R.B., Joshi, B.K., Shrestha, P. Chaudahry, B. Adhikari, N.P & Baniya, B.K. (2004). Intra- and interpopulation variation in finger millet (*Eleusine coracana* (L.) Gaertn) landraces grown in Kachorwa, Bara, Nepal. In: *On-farm Conservation of Agricultural Biodiversity in Nepal* (BR Sthapit, MP Upadhyay, PK Shrestha, DI Jarvis, eds). Volume 1. 2nd National Workshop on CUAPGR, 2017 223 Amount and Distributionof Genetic Diversity On-farm. Nagarkot, Nepal, pp.84-95. Proceedings.

Antony, U., Moses, L.G. & Chandra, T. S. (1998). Inhibition of *Salmonella typhimurium* and *Escherichia coli* by fermented flour of finger millet (*Eleusine coracana*), *World J. Biotechnol*, *14*(6), 883–886.

Baniya, B.K., Riley, K.W., Dongol, D.M.S. & Sherchand, K.K .(1992). *Characterization of Nepalese hill crops landraces (Barley, Buckwheat, Finger millet, Grain Amaranth, Foxtail, Proso and Barnyard millets)* (pp. 162). Kabre, Dolakha, Nepal: NARC-IBPGR,

Chadra, D., Chandr, S. & Sharma Pallavi, A.K. (2016). Review of finger millet (*Eleusine coracana* L. Gaertn), a power house of health benefiting nutrients. *Food Science and Human Wellness 5, 149–155.*

Chandrasekara, A. & Shahidi, F. (2011). Determination of antioxidant activity in free and hydrolyzed fractions of millet grains and characterization of their phenolic profiles by HPLC-DAD-ESI-MS, *J. Funct. Foods, 3*(3), 144–158.

Chandrasekara, A. & Shahidi, F. (2010). Content of insoluble bound phenolics in millets and their contribution to antioxidant capacity, *J. Agric. Food Chem.*, *58* (11), 6706–671.

Chethan, S. & Malleshi, N.G.(2007). Finger millet polyphenols: characterization and their nutra - ceutical potential, *Am. J. Food Technol*., *2* (7), 582–592.

Devi, P.B., Vijayabharathi, R., Sathyabama, S., Malleshi, N.G. & Priyadarisini, V.B. (2011). Health benefits of finger millet (*Eleusine coracana* L.) polyphenols and dietary fiber: a review. *J Food Sci Technol*, DOI: 10.1007/s13197-011-0584-9.

Doraiswamy T.R., Singh, N. & Daniel, V.A. (1969). Effects of supplementing ragi (*Eleusine coracana*) diets with lysine or leaf protein on the growth and nitrogen metabolism of children. *British Journal of Nutrition*, *23*, 737-743.

Duke, J.A. & Wain, K.K. (1981) *Medicinal Plants of the World*, Computer index with more than 85,000 entries (Vol. III), London, UK: Longman Group Ltd.

Enas, A., Kumar, S., Chennikkara, H. & Bjurlin, M.A**.** (2003) Prudent diet and preventive nutrition from pediatrics to geriatrics: Current knowledge and practical recommendations: *Indian Heart* Journal, *55*, 310-318.

Ghimire, K.H., Bhandari, B., Gurung, S.K., Dhani, N. B. & Baniya, B.K. (2017). Diversity and utilization status of millets genetic resources in Nepal. Conservation and Utilization of Agricultural Plant Genetic Resources in Nepal (BK Joshi, HB KC and AK Acharya, eds). 2nd National Workshop, 22-23 May 2017 Dhulikhel; NAGRC, FDD, DoA and MoAD; Kathmandu, Nepal. Proceedings.

Gull, A., Prasad, K., & Kumar, P. (2015). Effect of millet flours and carrot pomace on cooking qualities, color and texture of developed pasta. *LWT Food Science and Technology*, *63*, 470–474. http://dx.doi.org/10.1016/j.lwt.2015.03.008

Gupta, N., Srivastava, A.K. & Pandey ,V.N. (2012). Biodiversity and nutraceutical quality of some indian millets. *Proceedings of the National Academy of Sciences*, India Section B: *Biological Science*. DOI: 10.1007/s40011-012-0035-z.

Hegde, P.S., Rajasekaran, N.S. & Chandra, T.S.(2005a). Effects of the antioxidant properties of millet species on oxidative tress and glycemic status in alloxan-induced rats, *Nutr.Res*., *25*(12), 1109–1120.

Hegde, P.S., Anitha, B. & Chandra, T.S. (2005b). In vivo effect of whole grain flour of finger millet (*Eleusine coracana*) and kodo millet(*Paspalums crobic- ulatum*) on rat dermal wound healing, *Indian J.Exp.Biol*., *43*(3), 254–258.

International Crops Research Institute for the Semi-Arid Tropics. (2004). Finger millet. http://www.icrisat.org/web/ASP/mainsection.asp/ (assessed: 15 July,2004).

Jennings, H.M., Merrell, J., Thompson, J.L. & Heinrich, M. (2014). Food or medicine? The food-medicine interface in households in Sylhet. *J Ethnopharmacol*., *167*, 97–104.

Kim, J.S., Hyun, T.K. and Kim. M.J. (2011). The inhibitory effects of ethanol extracts from sorghum, foxtail millet and proso millet on *á*-glucosidase and a-amylase activities. *Food Chem*., *124*, 1647–51.

Lee, S. H., Chung, I. M., Cha, Y. S., & Park, Y. (2010). Millet consumption decreased serum concentration of triglyceride and C-reactive protein but not oxidative status in hyperlipidemic rats. *Nutrition Research*, *30*, 290–296.

Sri- Lankan Ministry of Agriculture. (2004). Finger millet: *Eleucine coracana* : Department of Agriculture. http://www.gov.lk/Agriculture/Agridept/Techinformations/Cgrains/Fmillet.htm.

Manach, C., Mazur, A. & Scalbert A. (2005). Polyphenols and prevention of cardiovascular diseases. *Current Opin Lipidol*, *16*, 77–84.

Manzoni, M. Bergomi, S. Rollini,M. & Cavazzoni, V. (1999). Production of stains by filamentous fungi, *Bio technol. Lett*., *21*(3), 253–257.

Ministry of Agriculture, Land Management and cooperatives. (2017). *Statistical Information on Nepalese Agriculture (2073/74*). Kathmandu, Nepal: Author.

Mohamed, E.A., Ahmed, I.A.M. & Babiker, E.E. (2011). Preservation of millet flour by refrigeration: changes in total protein and amino acids composition during storage. *World Acad Sci Eng Technol*, *52*, 11-15.

Rajasekaran, N.S., Nithya, M. Rose, C. & Chandra, T.S.(2004). The effect of finger millet feeding on the early responses during the process of wound healing in diabetic rats, *BBA-Mol. Basis Dis., 1689* (3) , 190-201.

Rana, R.B., P. Chaudhary, D. Gauchan, S.P. Khatiwada, B.R. Sthapit, A. Subedi, M.P. Upadhyay & D. I. Jarvis. (2000) . *In situ* crop conservation: findings of agro-ecological, crop diversity and socio-economic baseline survey of Kachorwa eco-site, Bara, Nepal. NP Working Paper No. 1/2000. NARC/LIBIRD/IPGRI, Kathmandu, Nepal/Pokhara, Nepal/Rome, Italy.

Saleh, A.S.M., Zhang, Q., Chen, J. & Shen, Q. (2013). Millet grains: nutritional quality, processing and potential health benefit. *Comprehensive Reviews in Food Science and Food Safety*, *12*, 281-295. doi: 10.1111/1541-4337.12012

Scalbert, A., Manach, C., Morand, C., Remesy, C. & Jimenez, L.(2005). Dietary polyphenols and the prevention of diseases. *Crit Rev Food Sci Nutr*., *45*, 287-306.

Shobana, S., Sreerama, Y.N., & Malleshi, N.G. (2009). Composition and enzyme inhibitory properties of finger millet (*Eleusine coracana* L.) seed coat phenolics: mode of inhibition of glucosidase and amylase, *Food Chem*., *115* (4) , 1268–1273.

Sripriya, G., Chandrasekharan, K., Murty, V.S. & Chandra , T.S. (1996). ESRspectro- scopic studies o n free radical quenching action of finger millet (*Eleusine coracana*), *FoodChem*., *57*(4), 537–540.

Srivastava, K. & Sharma, A. K. (2012) . Nutraceutical importance of finger millet (Eleusine coracana) for improved human health.*The European Journal of Plant Science and* Biotechnology, *6*(2), *91-95.*

Sthapit, B.R., P.M. Pradhanang, R.J. Khadka & Subedi, K.D. (1993). Finger millet relay intercropping system in the hills of Nepal: its constraints, research findings and prospects. *In* Advances in small millets (K.W. Riley, S.C. Gupta, A. Seetharam and J.N. Mushonga, eds.), New Delhi, India: Oxford and IBH Publishing Company.

Tatala, S., Ndossi, G., Ash, D. & Mamiro, P. (2007). Effect of germination of finger millet on nutritional value of foods and effect of food supplement on nutrition and anemia status in Tanzania children, Tanzan. *J. Health Res*., *9* (2), 77-86.

Taylor, J. R. N., & Emmambux, M. N. (2008). Gluten free foods and beverages from millets. In E. *Gluten free cereal products and beverages* (pp. 1-27). Gallagher (Eds.), Burlington, USA: Elsevier.

Taylor, J. R. N., Schober, T. J., & Bean, S. R. (2006). Novel food and non-food uses for sorghum and millets. *Journal of Cereal Science*, *44*, 252-271. (http://dx.doi.org/10.1016/j.jcs.2006.06.009)

Tovey, F. I. Jayaraj, A.P. & Clark, C.G. (1975). The possibility of dietary protective factors in duodenal ulcer, *Postgrad. Med. J*., *51* (596), 366-372.

Upreti, R.P. (2002). Production system with millets as component. *In*. Enhancing the Contribution of Nutritious but Neglected Crops to Food Security and to Incomes of Rural poor's: Nepal Component-Nutritious Millets. Inception Workshop. 2-3 May 2007, NARC Kathmandu, Nepal. Proceedings.

Usha, A . (2004). Nutrition in HIV/AIDS. *iJM Diet and Nutrition.*, *7*(2), 12-18.

Usha, A, George, M.L. & Chandra, T.S.(1998). Inhibition of *Salmonella typhi- murium* and *Escherichia coli* by fermented flour of finger millet (*Eleusine coracana*). *World Journal of Microbiology and Biotechnology.*, *14*, 883-88.

Veenashri, B.R. & Muralikrishna, G . (2011). Invitro anti-oxidant activity of xylo- oligosaccharides derived from cereal and millet brans –a comparative study, *Food Chem.*, *126*(3), 1475-1481.

Venkateswaran, V. & Vijayalakshmi, G. (2010). Finger millet (*Eleusine coracana*)— an economically viable source for anti-hypercholesterolemic metabolites production by *Monascus purpureus*, *J.Food Sci.Techno*l., *47*(4), 426–431.

Vietnameyer, N.D., Borlaugh, N.E., Axtell, J., Burton, G.W., Harlan, J.R., & Rachie. K.O. (1996). Fonio. In: *Lost Crops of Africa* (pp. 39-58) (Vol 1), National Academy Press, New York: Grains BOSTID Publications.

Viswanath, V., Urooj, A., & Malleshi, N.G. (2009). Evaluation of antioxidant and antimicrobial properties of finger millet polyphenols (*Eleusine coracana*), *Food Chem*., *114*(1), 40–346.

Watt, J.M. & Breyer-Brandwijk, M.G. (1962). *The Medicinal and Poisonous Plants of Southern and Eastern Africa*, 2nd Ed (pp. 1457), Edinburgh: E. and S. Livingstone.

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