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Sulekha Gotmare [α] & Esha Tambe [σ]

Abstract- The genus Cinnamomum consists of more than hundred species which occur in Asia and Australia. Cinnamon bark oil possesses a sweet and strong taste. It is employed in food and flavoring industry. Cinnamon has antimicrobial, antidiabetic, antiulcer and anti-inflammatory properties. In this study, cinnamon bark oil was extracted from whole cinnamon bark and powdered cinnamon by hydrodistillation. Whole cinnamon, powdered cinnamon, and market cinnamon oils, analyzed by using GCMS, showed variation in the chemical composition. Data from the current study was compared with cinnamon bark oil composition from five different countries, obtained from the literature survey.

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I. Introduction

Cinnamomum zeylanicum, belonging to Family Lauraceae, is considered as one of the oldest spices in the world (Siddig Ibrahim Abdelwahab et al., 2017). Cinnamon (*Cinnamomum verum, Cinnamomum zeylanicum* synonym) is a small evergreen tree, 10 – 15 meters (32.8 – 49.2 feet) tall and it is native Sri Lanka and South India (Vaibhavi Jakhetia et al., 2010). Cinnamomum has trinerved and fragrant leaves, fruits seated on a cupule and paniculate inflorescences and flowers with nine stamens (Syaliza Abdul Hammid et al., 2016). It is, commonly known as Dalchini in Hindi and Tvak in Sanskrit, cultivated on the Western Ghats and adjoining hills in April – July and October – December (Nanda Amalesh et al., 2015). *C. zeylanicum* is popularly known as cinnamon-of-India in Brazil, cinnamon-of-Ceylon, cinnamon-of-smell, true-cinnamon, cinnamon-of-China or just cinnamon (Felipe Queiroga Sarmento Guerra et al., 2012). Overall, approximately 250 species have been identified among the cinnamon genus, with trees scattered all over the world, mostly in Asia and some in South and Central America and Australia (Pasupuleti Visweswara Rao and Siew Hua Gan, 2014; Syaliza Abdul Hammid et al., 2016).

Cinnamon as a spice dates back in Chinese writings to 4000BC (K. V. Peter, 2001). The botanical name *Cinnamomum* is derived from Hebraic and Arabic term *amomum* which means fragrant spice plant (K. V. Peter, 2001). In ancient Eygpt, cinnamon was used medicinally, as a flavoring agent as well as embalming (K. V. Peter, 2001; Raaz K Maheshwari et al., 2013). It was so highly treasured that it was considered more precious than gold and around this time cinnamon received much attention in China (Raaz K Maheshwari et al., 2013). Due to its demand, cinnamon became one of the first commodities traded frequently between the Near East and Europe. The fact that Venetian merchants controlled the entire cinnamon trade in Europe fuelled other European explorers to travel in search of avenues of obtaining the precious spice. After Portuguese discovered Sri Lanka (Ceylon) at the end of the 15th century and took control over cinnamon trade, the Dutch removed them from power a century later, followed by the British in the year 1796. Since then the importance of cinnamon trade showed declination, as cinnamon began to be produced and cultivated in other areas (Raaz K Maheshwari et al., 2013).

The United States Department of Agriculture, Nature Resources Conservation Service provides the taxonomic description of cinnamon as: - *Kingdom: - Plantae – Plants, Subkingdom: - Tracheobionta – Vascular plant, Superdivision: - Spermatophyta – Seed plant, Division: - Magnoliophyta – Flowering plant, Class: - Magnoliopsida – Dicotyledons, Subclass: - Magnoliidae, Order: - Laurales, Family: - Lauraceae – Laurel family, Genus: - Cinnamomum Schaeff. (Cinnamon), Spices: - Cinnamomum verum J. Presl – cinnamon, synonym: - Cinnamomum zeylanicum.*

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Figure 1a

Figure 1b

Figure 1c

Figure 1a: Picture of cinnamon quills (Source: - Internet)

Figure 1b: Picture of cinnamon flowers (Source: - Meena Vangalapati et al., 2012)

Figure 1c: Picture of cinnamon leaves and seeds (Source: - Meena Vangalapati et al., 2012)

In-vitro and *in-vivo* studies in animals and human beings from different parts of the world have demonstrated numerous health benefits of cinnamon such as anti-inflammatory, anti-microbial activity, reducing cardiovascular diseases, boosting cognitive function and reducing the risk of colon cancer (Priyanga Ranasinghe et al., 2013). Cinnamon is one such dietary component that has been shown to contain biologically active substances which help in the regulation of blood glucose level by insulin–mimetic properties (Priyanga Ranasinghe et al., 2017). It enhances glucose uptake by activating insulin receptor kinase activity, auto-phosphorylation of the insulin receptor and glycogen synthase activity (Priyanga Ranasinghe et al., 2017). The volatile oils obtained from the different parts cinnamon plant such as bark, leaf, and root barks vary significantly in chemical composition, which suggests that they might differ in their pharmacological effects as well (Priyanga Ranasinghe et al., 2013). Das Manosi et al. mentioned in their review article that the aqueous extract of *C. zeylanicum* Blume inhibited the replication of the influenza virus. Alcoholic extracts of cinnamon were found most effective in reducing the growth of *Helicopter pylori*. He also mentioned nematicidal activity of cinnamon against male, female and juveniles of pinewood nematods *Bursaphelenchus xylophilus* (Das Manosi et al., 2013). In this study whole cinnamon oil, powdered cinnamon oil and market cinnamon oil were analyzed by GCMS. This data was compared with other research papers.

II. Materials and Methods

Materials: Innamon bark (in the form of quills) and cinnamon powder packets were purchased from D Mart of Thane city, Maharashtra, India. Market cinnamon oil was purchased online from the manufacturer. The leaflet of the market cinnamon oil states that the following information: -

Botanical name: Cinnamomum zeylanicum

Parts of plant: Bark

Extraction method for cinnamon oil: Initially 125g of cinnamon was weighed to extract the oil. The percentage yield obtained from 125g was very less. Hence extraction was carried out using 300g of whole dried cinnamon barks and 215g of powdered cinnamon. The cinnamon dried bark and powdered cinnamon were transferred in a round bottom flask which was connected to the condenser. To this, 700 ml distilled water was added and the assembly was placed in a heating mantle. Initially, the contents were heated to 60°C and then increased gradually to 100°C. The extraction process was carried out for 3 hours (till no more oil drops were coming out of the condenser). The oil extracted from whole dried cinnamon barks is called as whole cinnamon oil and the oil extracted from powdered cinnamon is called as powdered cinnamon oil. Both the extracted was collected in glass bottles and stored in the refrigerator.

Source: - Genuine picture captured at Research lab of S.H.P.T College of Science, SNDT Women's University. PC: - Esha Tambe

Figure 2: Extraction of cinnamon oil from whole dried cinnamon barks using distillation flask.

GCMS method of analysis: The components of cinnamon oils were separated and identified on Perkin Elmer Clarus 600 C mass spectrometer. Separation of a mixture of compounds present in the cinnamon oils was achieved on GsBP-5ms (30m x 0.25mm ID x 0.25μm film thickness) capillary column. This column has the composition of 5% diphenyl and 95% dimethyl poly siloxane (non- polar) with the temperature range of -60°C to 350°C. For analysis using mass spectrometer as a detector, Helium (He) gas was used as carrier gas. The flow rate of carrier gas was set to 1ml/min. The injector was maintained at 220°C whereas MS source and Inlet line temperature was set at 280°C. All the three cinnamon oils were injected as neat samples. 0.2μl of cinnamon oil samples were injected with a split ratio of 50:1 to prevent the capillary column from overloading. The column oven was temperature programmed by initially keeping the temperature at 80°C for 2 mins then increasing the temperature by 5°C/min till 150°C and finally achieving 250°C by the rise of 5°C/min. The total run time was 41 mins. The mass range was set from 15 to 350 amu. Fragmentation was achieved by electron ionization (70eV). The identification of all the three cinnamon oils components was accomplished by comparing the mass spectra with those available in Wiley (Flavours and Fragrances of Natural and Synthetic Compounds 3) and NIST Library.

III. Results and Discussions

Extraction Yield: The percentage yield for whole cinnamon oil and powdered cinnamon oil was calculated by using the following equation (Shimaa A. Moawad et al., 2015): -

$$\text{Volatile oil (\%)} = \frac{\text{Weight of the volatile oil recovered in g}}{\text{Weight of sample taken in g}} \times 100$$

The percentage yield for whole cinnamon oil estimated was 1.02%. For powdered cinnamon oil, the percentage yield was 0.91%. The whole cinnamon oil extracted from whole dried cinnamon barks was light yellow in color whereas the powdered cinnamon oil was dark brown color. The dark brown color of powdered cinnamon oil may be because powder soaks water and becomes soggy which gets settled down at the bottom of the flask. The round bottom flask is directly heated using a heating mantle, hence due to direct heating the oil color might have turned to dark brown. Upon sniffing powdered cinnamon oil, the burning smell was not identified. The color of market cinnamon oil was yellow.

GCMS Study: The GCMS identified more than 15 different compounds in whole cinnamon oil, powdered cinnamon oil, and market cinnamon oil. The identification was made based on the retention time and library search of the mass spectra. The relative amount of each compound present in all the three cinnamon oils was calculated by instrument's software using % area normalization method. Compounds identified were broadly classified into chemical classes namely, aliphatic compounds, monoterpene hydrocarbons, sesquiterpene hydrocarbon, oxygenated terpenoids, and aromatic compounds.

Table 1: Chemical constituents present in whole cinnamon, powdered cinnamon, and market cinnamon oil

Sr. no.	Name of compounds	RT	Molecular weight based on Base peak	Whole cinnamon oil (%)	Powdered cinnamon oil (%)	Market cinnamon oil (%)
1	α-Pinene	4.008	136	ND	ND	0.35
2	Camphene	4.262	136	ND	ND	0.16
3	Benzaldehyde	4.409	106	ND	ND	0.15
4	α-Phellandrene	5.217	136	ND	ND	0.30
5	p-Cymene	5.607	136	ND	ND	0.56
6	β-Phellandrene	5.724	136	ND	ND	0.44
7	Eucalyptol	5.77	154	1.26	0.16	ND
8	Linalool	7.249	154	ND	ND	1.30
9	Benzenepropanal	8.7	134	0.76	0.28	ND
10	Cis-cinnamaldehyde	10.24	132	1.28	0.35	ND
11	Saffrole	12.039	162	ND	ND	0.76
12	Trans-cinnamaldehyde	12.2	132	91.56	90.90	ND
13	Eugenol	14.302	164	ND	ND	85.31
14	α-Cubebene	14.4	204	0.46	1.45	0.45
15	Caryophyllene	15.51	204	ND	ND	2.52
16	Coumarin (2H-1-Benzopyran-2-one)	15.85	146	0.72	0.88	ND
17	Cinnamyl acetate (E)	16.00	176	1.72	1.58	0.99
18	α-Muurolene	17.5	204	0.62	1.11	ND
19	trans-cadina-1(6),4-diene	18.21	204	ND	1.62	ND
20	Eugenyl acetate	18.284	206	ND	ND	2.89
21	Benzyl benzoate	26.64	212	ND	ND	2.63
Chemical classes						
Aliphatic compounds				ND	ND	ND
Monoterpene hydrocarbons				ND	ND	1.25
Sesquiterpene hydrocarbons				1.08	4.18	2.97
Oxygenated terpenoids				1.26	0.16	1.30
Aromatic compounds				96.04	93.99	93.29

RT – Retention time, ND – Not detected

The contemplation of chemical classes indicates that whole cinnamon oil, powdered cinnamon oil, and market cinnamon oil contains aromatic compounds as its vital component. These oils also manifested presence of monoterpene hydrocarbon, sesquiterpene hydrocarbon and oxygenated terpenoids in different amount.

The GCMS profile of all the three cinnamon oils showed variations in the chemical constituents. The chromatograms of whole cinnamon oil and powdered cinnamon oil displayed good resemblance while market cinnamon oil showed non-identical chromatogram. The major compound found in the whole cinnamon oil, and powdered cinnamon oil was trans-cinnamaldehyde. 91.56% got detected in whole cinnamon oil and 90.90% in powdered cinnamon oil. In addition to trans-cinnamaldehyde, cinnamyl acetate, cis-cinnamaldehyde, eucalyptol, α-Cubebene, coumarin, and α-Muurolene were also detected in whole and powdered cinnamon oil. The descending order of compounds present in whole cinnamon oil is shown as follows: -

Trans-cinnamaldehyde (91.56%) > cinnamyl acetate (1.72%) > cis-cinnamaldehyde (1.28%) > eucalyptol (1.26%)>Benzenepropanol (0.76%) > coumarin (0.72%) > α-Muurolene (0.62%) > α-Cubebene (0.46%)

Similarly, for powdered cinnamon oil the descending order is shown as follows: -

Trans-cinnamaldehyde (90.90%) > trans-cadina-1(6), 4-diene (1.62%) > cinnamyl acetate (1.58%) > α-Cubebene (1.45%) > α-Muurolene (1.11%) > coumarin (0.88%) > cis-cinnamaldehyde (0.35%) > benzenepropanol (0.28%) > eucalyptol (0.16%)

The order depicts variation in the concentration of minor compounds detected in whole and powdered cinnamon oil. trans-cadina-1(6), 4-diene was identified only in powdered cinnamon oil. Cinnamyl acetate was found in both whole and powdered cinnamon oil with a difference of 0.14% in concentration. Eucalyptol (1, 8-cineole) was found in good amount in whole cinnamon oil.

GCMS profile of market cinnamon oil showed detected eugenol as its major compound. Eugenol, a

natural antioxidant, was present accounting for 85.31% of the total constituents. The other compounds, present in less amount, were linalool (1.30%), caryophyllene (2.52%), benzyl benzoate (2.63%) and eugenyl acetate (2.89%). In addition to these compounds, compounds such as p-cymene, α-pinene, benzaldehyde, camphene, α-phellandrene, β-phellandrene, saffrole, α-cubebene, and cinnamyl acetate were detected below than 1%. The decreasing order of the compounds identified by GCMS are as follows: -

Eugenol (85.31%) > eugenyl acetate (2.89%) > Benzyl benzoate (2.63%) > caryophyllene (2.52%) > linalool (1.30%) >cinnamyl acetate (0.99%)>saffrole(0.76%) >p-cymene (0.56%) > α-cubebene (0.45%) > β-phellandrene(0.44%) > α-pinene (0.35%) > α-phellandrene (0.30%) > camphene (0.16%) > benzaldehyde (0.15%)

It is evident through the majority of researcher's work that eugenol is the main component of cinnamon leaf oil and cinnamaldehyde is the prime component of cinnamon bark oil. Pasupuleti Visweswara Rao and Siew Hua Gan had mentioned the presence of Eugenol (70 – 95%) in cinnamon leaf oil in their review article. While cinnamon bar oil has cinnamaldehyde (65 – 80%), cinnamon root bark contains camphor (60%) and fruit oil consists of trans-cinnamyl acetate (42 – 54%) as well as caryophyllene (9 – 14%) as their important constituents (Pasupuleti Visweswara Rao and Siew Hua Gan, 2014). Cinnamon oil leaf oil, collected from Nallurkadu, Palani Hills, Tamil Nadu, India, revealed the presence of eugenol (81.7%) as its major compound (Anubhav Chakraborty et al., 2015). *Cinnamomum verum* leaves and bark on distillation yielded essential oils rich in eugenol and cinnamaldehyde respectively (B R Rajeswara Rao et al., 2007). Eugenol is the major component of *Cinnamomum zeylanicum* leaves representing 84.9% of the total constituents (Felipe Queiroga Sarmen to Guerra et al., 2012). Erich Schmidt analyzed *Cinnamomum zeylanicum* leaf oil which showed the presence of eugenol (74.9%) as its main component, along with caryophyllene, benzyl benzoate, linalool and eugenyl acetate (Erich Schmidt et al., 2006). P. A. Paranagama et al. published a comparative study of *Cinnamomum zeylanicum* bark, leaf, root and fruit oil. The study conveyed that bark oil had cinnamaldehyde; leaf oil showed eugenol, benzyl benzoate, and β-caryophyllene; root oil consists of camphor, 1,8-cineole and limonene; fruit oil had δ-cadinene, γ-cadinene, T-cadinol, α-cadinene and β-caryophyllene. Richardo Dias de Castro and Edeltrudes Oliveira Lima had also mentioned their research paper that cinnamon leaf oil comprises eugenol (73.27%) as its major component, followed by benzyl benzoate and β-caryophyllene (Richardo Dias de Castro and Edeltrudes Oliveira Lima, 2013).

The chromatograms of whole cinnamon oil, powdered cinnamon oil, and market cinnamon oil are shown in figure 3, 4 and 5 respectively.

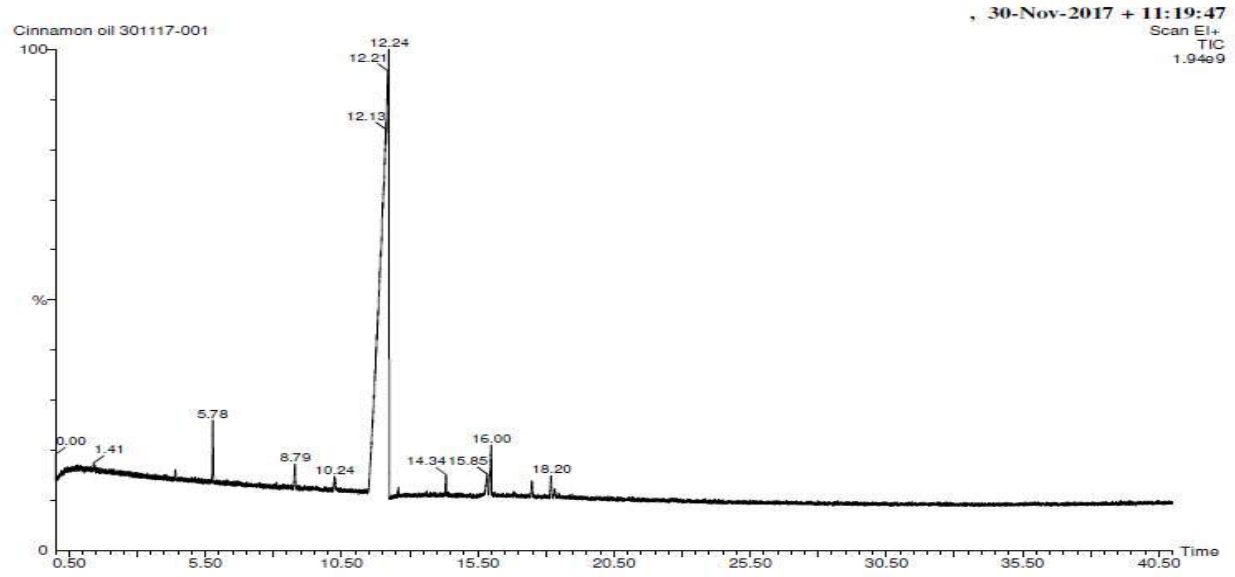

Figure 3: Chromatogram of whole cinnamon oil

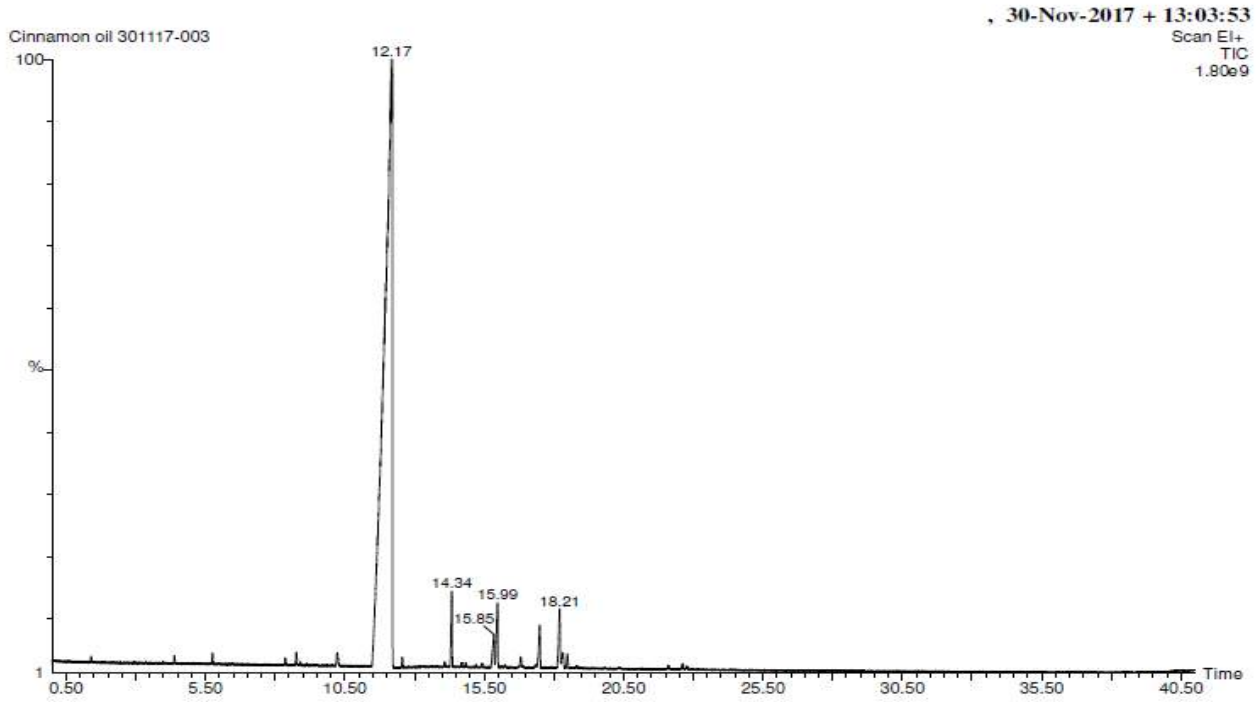

Figure 4: Chromatogram of powdered cinnamon oil

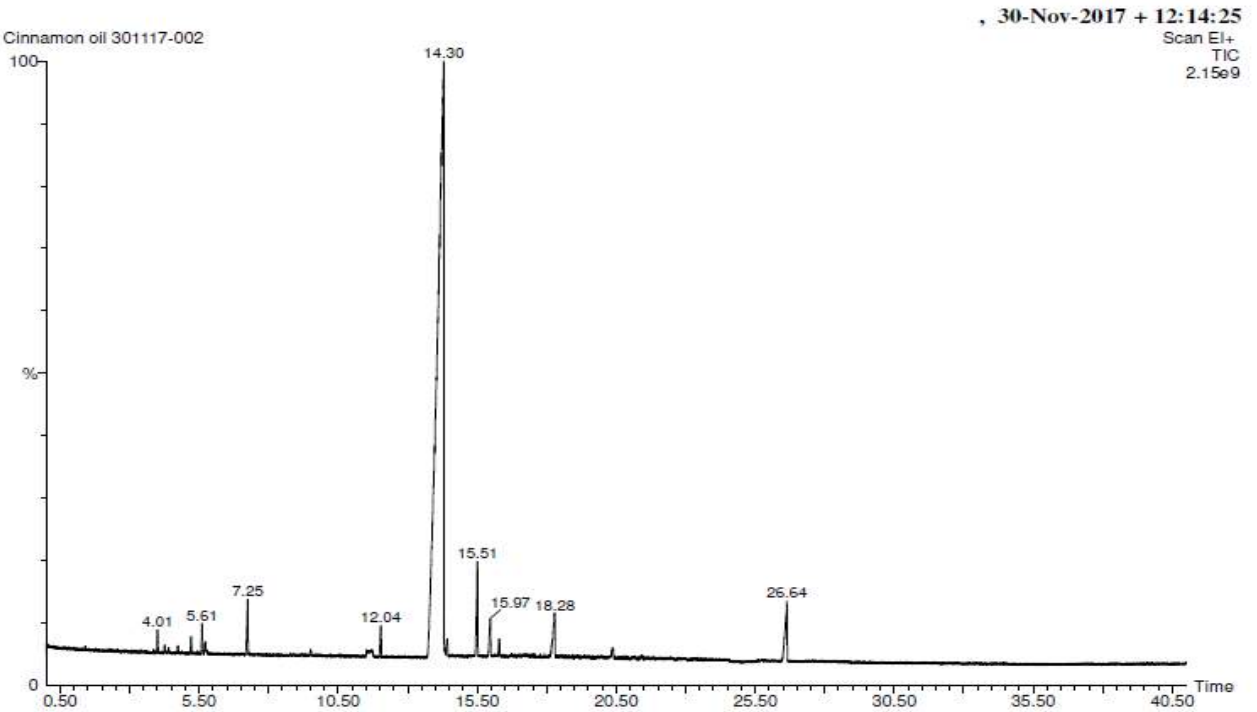

Figure 5: Chromatogram of market cinnamon oil

Comparative study of chemical composition data from five countries: - In addition to the above analysis, data, obtained from research papers, was compiled in table 2. The below table renders information of the chemical composition of cinnamon bark oil from five different countries namely, Sri Lanka, Egypt, India, Ethiopia, and Malaysia.

Table 2: Chemical composition of cinnamon bark oil obtained from five different countries

Sr. no.	Compounds identified (area percentage)	Country	References
1	α-pinene (3.34%), Myrcene (2.70%), α-Terpenene (1.30%), Limonene (1.20%), p-cymene (1.91%), Linalool (3.70%), 1,8-cineole (4.60%), Cinnamaldehyde (50.5%), z-cinnamyl acetate (8.78%), Eugenol (4.15%), β-caryophyllene (8.00%), α-Humulene (1.30%)	Sri Lanka	P.A. Paranagama et al., 2001
2	α-pinene (1.12%), Limonene (1.48%), 1,8-cineole (1.01%), Nerol (1.06%), Neral (1.16%), Geranial (1.79%), Cinnamaldehyde (45.13%), Cinnamyl alcohol (5.13%), Eugenol (7.47%), Dihydroeugenol (3.31%), cis-ethyl cinnamate (3.86%), t-methyl cinnamate (2.19%), Methyl eugenol (5.23%), Isoeugenol (1.59%)	Egypt	G. S. El-Baroty et al., 2010
3	Borneol (1.03%), trans-cinnamaldehyde (84.97%), Ethanone (1.11%), 1,2-Naphthalenedione (9.03%)	Malaysia	Nur Nasulhah Kasim et al., 2014
4	Ethyl benzene (1.50%), trans-cinnamaldehyde (64.84%), 1,2,4-Metheno-1H-indene, octahydro-1,7α-dimethyl-5-(1-methylethyl) (1.90%), Copaene (8.48%), Eugenol (6.72%), Caryophyllene (2.01%), Cadina-4,9-diene (3.09%), Cadina-3,9-diene (4.46%), 17-Pentatriacontene (3.19%)	Egypt	Eman M. Elgendy et al., 2017
5	Trans-cinnamaldehyde (87.013%), Eugenol (9.317%) and other compounds were present in less than 1%	Ethiopia	Bizuneh Adinew, 2014
6	Cinnamaldehyde (91.82%), 1,8-cineole (1.5%), Ylangene (1.49%), E-Phellendrene (1.23%), α-Murrolene (1.92%), Selinene (1.84%), Geraniol (1.31%), E-Cinnamic acid (1.62%)	India	Pooja Arora et al., 2013

From table 2, it becomes crystal clear that no matter from where the cinnamon bark oil originates it contains cinnamaldehyde as its prime compound. The percentage of cinnamaldehyde detected is less Sri Lanka and Egypt cinnamon bark oil. Whereas the cinnamon bark oil obtained in Malaysia, Ethiopia and India are rich in cinnamaldehyde which accounts for more than 80% of the total oil constituents. Along with cinnamaldehyde, other compounds were also reported. Sri Lanka cinnamon bark oil showed the presence of z-cinnamyl acetate, β-caryophyllene, eugenol, and 1,8-cineole in the range of 4% - 9%. Cinnamon bark oils from Egypt was extracted from cinnamon bark purchased from regional, local store/market. Both these oils showed variation in cinnamaldehyde concentration with a difference of 19.71%. G. S. El-Bartoy et al. mentioned the presence of α-Pinene, Limonene, neral, methyl eugenol, dihydroeugenol, cinnamyl alcohol,1,8-cineole, t-methyl cinnamate, and cis-ethyl cinnamate. The common compound identified in both Egypt cinnamon oil trans-cinnamaldehyde and eugenol. Indian cinnamon oil exhibited the presence of cinnamaldehyde, along with ylangene and selinene.

IV. Conclusion

From the current study, it may be concluded that cinnamon bark oil comprises cinnamaldehyde as its major compound. Hence cinnamaldehyde may also be considered as a marker compound of cinnamon bark oil. The composition of whole cinnamon oil and powdered cinnamon oil showed good similitude. The market cinnamon oil, found rich in eugenol content, may be extracted from cinnamon leaves, according to the literature survey. On the basis of literature survey, it may be concluded that eugenol is the marker compound of cinnamon leaf oil. Hence identification of these marker compounds is essential, as they reflect the crucial information from which part of the plant the oil is extracted. In general, an inference may be drawn that these marker compounds shall be used as an indicator to evaluate the quality or authenticity of essential oils.

The disparity in the chemical composition of cinnamon bark oil found may be attributed due to the variation in cultivation practices, plantation season, number and time of cuts, plant development stage and the climatic conditions. The present work also provides an evident that apart from climatic, plant varieties and agricultural practices, the chemical composition also varies according to the parts of plants (whole, flowers, buds, leaf, stem, root, etc.).

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