



INSTITUTO DE CIÊNCIAS BIOLÓGICAS

DEPARTAMENTO DE BOTÂNICA

DOUTORADO EM BOTÂNICA

## **Brioflora Antártica: Novidades para o Gênero *Syntrichia* Brid. (Pottiaceae - Bryophyta) e para a Ilha Snow**

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Brasília

2021



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Tese de doutorado apresentada à

Programa de Pós-Graduação em Botânica da

Universidade de Brasília, como parte dos requisitos

necessários para obtenção do título de Doutor

em Botânica

Júlia Viegas Mundim

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Aprovada em 18 de março de 2021

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## **Dedicatória**

Dedico este trabalho ao meu pai, José Mundim Rios (*in memorian*), com todo o meu amor e gratidão. Sua bondade, carisma, força de vontade e resiliência foram os meus melhores exemplos e me motivaram a prosseguir. Espero ter sido merecedora do seu esforço.

## Agradecimentos

Aos meus pais, Eleuza Viegas e José Mundim por seu amor e apoio incondicionais, mesmo quando não compreendiam exatamente o caminho que estava trilhando.

Ao meu marido, Thiago Felipe Santos, por ter embarcado nesse processo comigo. Sua sinceridade, amizade e amor foram fundamentais.

Aos Meus irmãos, Juliana V. Mundim e José M. Júnior, meus exemplos de vida, que sempre me incentivaram a estudar e buscar as melhores oportunidades, etando sempre ao meu lado, e, aos irmãos de coração (Andressa Viegas, Elaine de Souza e Ambrósio) sou muito grata a vocês.

Aos meus sobrinhos (Isabela Mundim, Juliane Mundim, Joaquim Mundim, Ana Júlia Mundim e Ambrósio Júnior) pelo carinho e por entender minhas ausências.

À tia Tânia Silva por sua bondade e amor, tendo me acolhido em sua casa.

À Tamara Dantas, amiga e irmã de coração, pelo apoio e cumplicidade sempre. Não conseguia sem você.

À Osvanda Moura pela amizade e ajuda na correção dos textos.

À Ana Gabriela Duatrte pela amizade, carinho e ensinamentos.

À Carla Pereira pela amizade, vivências no laboratório e ensinamentos.

À Poliana Teixeira e Cristiely Machado pela amizade, companheirismo e ajuda em muitos momentos.

À Amanda Leal pela amizade, carinho e ajuda sempre.

À Amanda Marinho pelo carinho, amizade e companheirismo.

À Amanda Ribeiro pela amizade e incentivo desde o inicio.

À Fernanda Kucharski pela amizade, incentivo e compreensão.

À Sabrina Cajamarca pelo apoio, amizade e companheirismo nesses anos.

Ao amigo e ex-professor Dr. Lacê Breyer pela amizade, conversas e ensinamentos.

Ao Dr. Diego Knop (Pikachu) pelos ensinamentos e amizade.

Ao Diogo Pereira pelos bons momentos, conversas e amizade.

Ao Jair Faria pela amizade, apoio no trabalho e incentivo.

Ao André Moreira, pela amizade e ajuda nos estudos para ingressar na Pós-Graduação.

Aos Coelgas do Laboratório de Criptogámas-Allan Faria, Abel Soares, Eduardo Amorim, Dafne Anjos, Mateus Melo, Maria Eduarda, Leônidas Bandeira, Paulo Afonso, Maria Clara, pela ajuda, bons momentos e sorrisos.

Aos amigos e colegas do Departamento de Botânica Andressa Dantas, Eriel Amaral, Natália Bijos, Joicelene Lima, Maria Rosa Zanatta, Mayco Santos, Marlon Facco e Jéssika Viera pela amizade e conhecimentos compartilhados.

Aos amigos e companheiros de trabalho ou apoio nas pesquisas antárticas, em especial Mayara Scur, Marisol Pizarro, Rodrigo Contreras, Partrícia Jungblunth, Ingrid Hebel, Kênia Picoli, Paulo Peixoto, Camilo de Lellis.

Aos amigos briologos, Professor Denilson Peralta, Dimas do Carmo, Anne Morel e Emanuelle Santos.

Aos amigos que a UnB me presenteou Tamiris Assis, Alexandre Coutinho, Mayara Albergaria, Diego Jacome, Reneida Mendes, Aline Firmino, Flávia Souza, Fernanda Fiusa, Jesus Perez e Jazmin Magana, pelo incentivo.

Ao professor Dr. Paulo Câmara por aceitar o desafio de me orientar (desde a graduação), por seus ensinamentos, sua amizade e pela oportunidade de trabalhar com ciência antártica.

À professora Dra. Micheline Carvalho-Silva pelo acolhimento. Sua ajuda, paciência e amizade foram imprecindíveis.

À professora Dra. María Teresa Gallego pelos ensinamentos, treinamento e cordialidade, bem como a equipe do grupo Pottiaceae (Universidade de Múrcia – Espanha) pela oportunidade.

Aos professores do Departamento de Botânica, em especial as professoras Dra. Graça Machado, Cássia Munhoz e Regina de Oliveira pelo exemplo; e ao professor Lício Flávio pelos ensinamentos e empatia.

Aos professores da FUP –UnB, que me incentivaram desde o início da minha formação, em especial Maria Elizabeth Mamede da Costa, Dulce Maria Sucena, Renata Aquino, Cleilton (Mickhael) e Alex Fabiano.

Aos funcionários técnicos da Universidade de Brasília (Josemíbia Miranda, Jéssika, André Rodolfo, Dani Cemin, Sarah Lee, Cássia Cavalcante e Sílvia) por todo o auxílio prestado.

À Universidade de Brasília e ao Programa de Pós-Graduação em Botânica pela viabilização e pelo apoio para realização deste trabalho.

À CAPES. O presente trabalho foi realizado com apoio da Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Código de Financiamento 001.

À FAPDF, pelo auxílio para realização da visita técnica.

À Marinha do Brasil pelo empenho e apoio nas expedições de campo e aos programas antárticos da Polônia, Chile e Argentina (em especial a FAA) pela cooperação.

Aos Programas Antárticos Equatoriano e Uruguaios pela Acolhida.

Aos membros da banca pela paciência e por disporem do seu tempo e conhecimentos para avaliar este trabalho.

A todos, meus sinceros agradecimentos!

## Sumário

Organização da Tese .....	2
Resumo .....	3
Abstract .....	4
Introdução Geral.....	5
Referências.....	7
CAPÍTULO 1 .....	10
<b>Taxonomic Studies of <i>Syntrichia</i> Brid. in Antarctica .....</b>	10
Abstract .....	12
Introduction.....	13
Materials and Methods .....	14
Sampling .....	14
Morphological study.....	15
Molecular data analyses .....	15
Alternative Hypothesis Test - Teste SH .....	17
Results.....	17
Molecular studies .....	17
Alternative Hypothesis Test - Teste SH .....	21
Morphological investigation.....	21
Key to the species of <i>Syntrichia</i> Brid. occurring in Antarctica.....	26
Discussion .....	27
Conclusions.....	29
References .....	30
Supplementary Material .....	36
Appendix 1 .....	36
Appendix 2 .....	39
CAPÍTULO 2 .....	42
<b>Small areas and small plants: Updates on Antarctic bryophytes .....</b>	42
Abstract .....	44
Introduction.....	45
Materials and Methods .....	47
Results and Discussion.....	49
Final Considerations.....	61
Acknowledgments .....	62
Appendix 1. Voucher information of specimens collected in Snow Island in this study.....	67
Considerações Finais da Tese.....	69
Apoio e Financiamento .....	70

## **Lista de Figuras**

### **Capítulo 1 - Taxonomic Studies of the *Syntrichia* Brid. in Antarctica**

Figure 1. <i>Syntrichia magellanica</i> (Mont.) R.H. Zander – Robert Island, Antarctica.....	13
--	----

Figure 2. Figure 2 - Phylogram (consensus) generated by Bayesian Inference from the ITS2 matrix (with indel coding) [...]	20
---	----

Plate 1. Morphology of <i>Syntrichia</i> Brid. species occurring in Antarctica.....	25
---	----

### **Capítulo 2 - Small areas and small plants: Updates on Antarctic bryophytes**

Figure 1. Map of President Head, Snow Island, South Shetlands, Antarctica. The indicate region represent Snow island, and President Head.....	48
---	----

Plate 1. Morphology of some of the mosses occurring in Snow Island.....	60
---	----

## **Lista de Tabelas**

### **Capítulo 1 - Taxonomic Studies of the *Syntrichia* Brid. in Antarctica**

Table 1. List of species used in the analysis of the ITS2 region, locality where it was collected, code used in the analyzes, number of collector, herbarium where it is deposited and Genbank accession number, respectively.....	39
--	----

Table 2. Alignment characteristics and maximum parsimony tree statistics.....	18
---	----

Table 3. Results from the Shimodaira-Hasegawa (SH) tests.....	21
---	----

Table 4. Morphological characterization of <i>Syntrichia</i> species from Antarctica.....	22
---	----

### **Capítulo 2 - Small areas and small plants: Updates on Antarctic bryophytes**

Table 1. List of moss families and species collected in Snow Island during Antarctic summers of 2015 and 2018. Classification follows Goffinet <i>et al.</i> (2009). New Occurrences are indicated by an asterisk.....	49
--	----

Table 2. Distribution data of the species present on Snow Island.....	51
---	----

## **Organização da Tese**

A presente tese foi dividida em dois capítulos, antecedidos por uma introdução geral, para orientação do leitor. Os capítulos estão estruturados em formato de artigo, conforme permitido pelas diretrizes do Programa de Pós Graduação em Botânica da Universidade de Brasília.

O capítulo 1 – Taxonomic Studies of the *Syntrichia* Brid. in Antarctica – traz uma abordagem filogenética e morfológica das espécies de *Syntrichia* Brid. ocorrentes na Antártica. Para a confecção deste, foram realizados testes com os marcadores plastidiais (*rps4*, *trnL-trnF*, *trnG*) e nuclear (ITS2), sendo aqui apresentados os resultados concernentes ao marcador ITS2, que abrangeu todas as espécies. O capítulo traz novidades para as espécies do gênero, sendo o intuito publicar o manuscrito no periódico *Polar Biology*.

O capítulo 2 – Small areas and small plants: Updates on Antarctic bryophytes – traz levantamento brioflorístico realizado a partir de coletas recentes na localidade da Ilha Snow, Antártica. Dentre as novas ocorrências detectadas estão espécies do gênero *Syntrichia*, também utilizadas nos estudos moleculares do capítulo 1, além de espécies de ocorrência rara pertencentes a exemplo de exemplares do gênero *Schistidium* Bruch & Schimp., o que denota a importância do acompanhamento da localidade. O artigo foi aceito pela revista *Acta Botanica Brasilica* e aguarda publicação.

## **Resumo**

A Antártica é o ambiente mais extremo e isolado da Terra. Apresenta frio intenso, baixa precipitação (especialmente na porção continental), alto índice UV, fortes ventos, características inóspitas de um bioma singular, detentor de uma flora predominantemente criptogâmica. Atualmente são conhecidas 116 espécies de musgos distribuídas em duas zonas geobotânicas - marítima e continental, a primeira apresentando maior riqueza. Apesar da existência de uma flora para a região, trabalhos recentes denotam lacunas. Novas espécies de briófitas têm sido encontradas, bem como alterações na distribuição de alguns *taxa*. Visando contribuir para a atualização do conhecimento sobre a brioflora e a ampliação das informações disponíveis, a presente pesquisa, estruturada em dois capítulos, dedica-se no primeiro capítulo ao gênero *Syntrichia* Brid. (Pottiaceae), conhecido por sua complexidade e incongruências; as espécies deste gênero, ocorrentes na Antártica, foram submetidas a estudos moleculares com o marcador ITS2, em conjunto a revisão da morfologia, o que apresentou subsídios para a exclusão de *Syntrichia caninervis* var. *caninervis* da flora Antártica, bem como a adição de *Syntrichia sp1*. No capítulo 2, apresenta-se atualização para a distribuição de 19 espécies de musgos, dentre elas as espécies de *Syntrichia*; as espécies estão localizadas na Ilha Snow - Arquipélago das Shetland do Sul, e dentre os *taxa* estão espécies raras como *Schistidium lewis-smithii* Ochyra, endêmico da Antártica, anteriormente relatado para apenas duas localidades.

**Palavras-chave:** Bryophyta; Pottiaceae; Estudos moleculares; Flora de Musgos Antártica.

## **Abstract**

Antarctica is the most extreme and isolated environment on Earth. Presents intense cold, little precipitation (especially in continental portion), high UV index, wind strong, inhospitable characteristics of a unique biome, holder of a predominantly cryptogamic flora. Currently, 116 species of moss are known, distributed in two geobotanical zones - maritime and continental, the first presenting greater wealth. Despite the existence of a flora for the region, recent works show gaps. New species of bryophytes have been found, as well as changes in the distribution of some taxa. To contribute to updating the knowledge about bryophytes and the expansion of available information, this research, structured in two chapters, is dedicated in the first chapter to the genus *Syntrichia* Brid. (Pottiaceae), known for its complexity and incongruities; the species of this genus, occurring in Antarctica were submitted to molecular studies on ITS2 marker together review the morphology, which grants presented to the exclusion of *Syntrichia caninervis* var. *caninervis* from the Antarctic flora, as well as the addition of *Syntrichia* sp1. Chapter 2 presents an update for the distribution of 19 species of moss, among them the species of *Syntrichia*; the species are located on Snow Island - South Shetland Archipelago, and among the taxa are rare species such as *Schistidium lewis-smithii* Ochyra, endemic to Antarctica, previously reported for only two locations.

**Keywords:** Bryophyta; Molecular Studies; Moss Antarctic flora.

## **Introdução Geral**

As briófitas, *sensu latu*, são o segundo maior grupo de plantas terrestres, no quesito número de espécies. Apresentam geralmente pequeno porte, não possuem flores (criptogamas) e são avasculares (Goffinet & Shaw 2009; Vanderpoorten & Goffinet 2009; Costa 2010). Formam um grupo parafilético (Qiu *et al.* 2006), dividido em três linhagens – Marchantiophyta (hepáticas), Anthocerophyta (antóceros) e Bryophyta (musgos) (Goffinet & Shaw 2009; Vanderpoorten & Goffinet 2009).

Possuem um ciclo de vida em que o gametófito (haplóide) é dominante, enquanto o esporófito (diplóide) é a geração dependente e temporária (Vanderpoorten & Goffinet 2009). Ocorrem desde o nível do mar até 5.000 m de altitude e são capazes de desenvolver-se nos mais diversos ambientes, até mesmo aqueles de características extremas (como frio e baixa humidade); as briófitas são encontradas desde áreas tropicais até mesmo desertos e regiões polares, como a Antártica (Costa *et al.* 2010; Vanderpoorten & Goffinet 2009).

A Antártica abrange toda a região ao sul do paralelo 60° sul- interseção entre a porção continental e o Oceano Austral - totalizando uma extensão de aproximadamente 14 milhões de km<sup>2</sup> (Peat *et al.* 2007; Brasil 2014). Ambiente caracterizado pelos superlativos, a Antártica apresenta baixa precipitação - média anual de 20 mm na porção continental (Estação Vostok) e 1300 mm na Antártica Marítima (Shetlands do Sul); temperaturas médias anuais entre -50°C no continente e -2,8°C na porção marítima; além de ventos que podem ultrapassar 100 km/h (Brasil 2014; Cassano 2013), características adversas a muitos seres vivos, porém, bem suportadas por organismos extremófilos, ou seja, aqueles capazes de desenvolver-se em ambientes com tais especificidades (Longton & Holdgate 1979).

A flora Antártica é caracterizada pela predominância de organismos criptogâmicos, sendo 116 espécies de musgos atualmente descritas (Smith 1984; Walton 1990; Ochyra *et al.* 2008; Ellis *et al.* 2013a; 2013b; Sollman 2015; Câmara *et al.* 2019a).

O conhecimento da flora, até pouco tempo, limitava-se a dados morfológicos e baseava-se em coletas antigas como aquelas utilizadas nos levantamentos de Ochyra *et al.* (2008). Trabalhos recentes, baseados em novas amostragens (Câmara *et al.* 2017;

Henriques *et al.* 2018; Câmara *et al.* 2020) e envolvendo análises de DNA (Biersma *et al.* 2017; 2018a; 2018b; Câmara *et al.* 2019a; 2019b) põem em discussão a distribuição das espécies antárticas, que têm apresentado ocorrência mais abundante do que aquela descrita na flora Antártica, ou ainda, correspondendo, erroneamente, a outro *táxon*.

Nesse sentido, nota-se que existem lacunas nas informações disponíveis sobre a flora de briófitas da Antártica, no concernente a identidade e a distribuição das espécies, bem como conhecimento limitado referente ao inventário das floras locais (como observado em ilhas do arquipélago das Shetland do Sul).

Com intuito de contribuir para o conhecimento da brioflora Antártica, gerando atualização para distribuição das espécies, além de dados moleculares, a presente tese teve como objetivos: a) estudar o gênero *Syntrichia* (filogenia e morfologia), que apresenta um longo histórico de incongruências nomenclaturais e similaridade morfológica com outras espécies (Afonina *et al.* 2014; Zander 1993); b) realizar levantamento brioflorístico da Ilha Snow, historicamente pouco amostrada, a semelhança de outras ilhas do arquipélago (que apresentaram novas ocorrências) (Câmara *et al.* 2017; Henriques *et al.* 2018).

Para elaboração do capítulo 1- Taxonomic Studies of the *Syntrichia* Brid. in Antártica foram realizados ensaios com marcadores de DNA das regiões ITS2 (Afonina *et al.* 2014), *trnG* (Gallego *et al.* 2014), *trnL-trnF* (Gallego *et al.* 2014) e *rps4* (Werner *et al.* 2002, 2004). No entanto, como o sucesso de amplificação foi distinto entre os marcadores (*rps4* – 1 espécie amostrada; *trnG* e *trnL-trnF* – 3 espécies amostradas; ITS2 – ao menos 1 amostra de cada espécie) preconizou-se a região de melhor resolução entre as espécies (ITS2). Em adição, realizou-se revisão da morfologia.

O Capítulo 2 - Small areas and small plants: Updates on Antarctic bryophytes – traz atualizações para flora de musgos da Ilha Snow, incluindo espécies de *Syntrichia* (também utilizadas nos estudos moleculares do capítulo 1) e outros *taxa*, anteriormente não relatados para a região, promovendo um aumento descomunal do quantitativo de espécies anteriormente conhecido para a região. O capítulo traz comentários sobre distribuição, prancha com imagens das principais espécies e mapa da localidade.

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# CAPÍTULO 1

**Taxonomic Studies of *Syntrichia* Brid. in Antarctica**

## **Taxonomic Studies of *Syntrichia* Brid. in Antarctica**

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## **Abstract**

Antarctica has a unique biodiversity, adapted to extreme conditions such as intense cold, high ultraviolet radiation and low humidity. In this continent, mosses are the predominant vegetation. Among the most diverse moss families in Antarctica is Pottiaceae, with 10 described genera, the most diverse being *Syntrichia* Brid. with five species (*Syntrichia caninervis* Mitt., *S. filaris* (Müll. Hal.) R.H. Zander, *S. magellanica* (Mont.) R.H. Zander, *S. sarconeurum* (Hook. F. & Wilson) Ochyra & RH Zander and *S. saxicola* (Cardot) R.H. Zander). That genus is a taxonomically complex group, due to its morphological variability and similarities to other genera within the family, leaving a gap on the understanding of relationships between taxa, especially due to the few DNA studies. Seeking a better knowledge of the Antarctic species of *Syntrichia* and their relationships within the genus, a phylogenetic study was carried out using nuclear DNA sequences (ITS2 region). DNA was obtained from collections made in different parts of Antarctica and Subantarctica, as well as from herbarium specimens. From one to five DNA sequences were obtained for each target species and analyzed under Maximum Parsimony, Maximum Likelihood and Bayesian Inference. The results denote the presence of taxon not previously reported for Antarctica (*Syntrichia sp1*); besides as updates in the distribution of *Syntrichia sarconeurum*; and the suggestion to exclude a taxon from the list of Antarctic moss species.

**Keywords:** Bryophyta, Flora, new occurrence, Phylogeny, Pottiaceae.

## Introduction

Antarctica is the most isolated and extreme continent (Bargagli 2005; Cassano 2013). Mosses are the predominant vegetation with 116 species, distributed in 50 genera and 17 families (Ochyra *et al.* 2008; 2013a; 2013b; Sollman 2015; Câmara *et al.* 2019). According to Ochyra *et al.* (2008) Pottiaceae is the second most diverse moss family in Antarctica, with 10 genera, among which is *Syntrichia* Brid. (Fig. 1).



Figure 1. *Syntrichia magellanica* (Mont.) R.H. Zander – Robert Island, Antarctica.

*Syntrichia* is a taxonomically complex group (Afonina *et al.* 2014; Zander 1993), composed by morphologically diversified species (Gallego 2005). The majority of studies involving this genus are based on morphological characters such as traditional revisions or floristic surveys (Kramer 1980; Zander 1993; Gallego 2005; Gallego & Cano 2007a; 2007 b; 2009; Gallego *et al.* 2004; 2006; 2009; Misheler 2007; Ignatov *et al.* 2006; Goga *et al.* 2018; Henriques *et al.* 2018; Câmara *et al.* 2017; Ochyra *et al.* 2008; 2014; Mundim *et al.* 2021). A small number of studies involve phylogeny (Werner *et al.* 2002; 2004; Afonina *et al.* 2014; Gallego *et al.* 2014) or phylogeographic studies (Selkirk *et al.* 1997; Skotnicki *et al.* 1999; 2000; 2004).

Among the 80 known *Syntrichia* species in the world, five occur in Antarctica: *Syntrichia filaris* (Müll. Hal.) R.H. Zander, *Syntrichia magellanica* (Mont.) R.H. Zander, *Syntrichia saxicola* (Cardot) R.H. Zander, *Syntrichia caninervis* var. *caninervis* Mitt. (and *Syntrichia caninervis* var. *gypsophila* Ochyra) and *Syntrichia sarconeurum* (Hook. F. & Wilson) Ochyra & R.H. Zander (Ochyra *et al.* 2008).

According to Ochyra *et al.* (2008) *Syntrichia caninervis* (both varieties) is bipolar (without intermediate), that is, it occurs in both polar regions (Donoghue 2011); *Syntrichia sarconeurum* is endemic to the region (Ochyra *et al.* 2008); and *S. filaris*, *S. magellanica* and *S. saxicola* occur in Antarctic and sub-Antarctic regions (for example, South Georgia and Falkland Islands/Malvinas) (Ochyra *et al.* 2008).

There are yet no phylogenetic investigations that include Antarctic samples of *Syntrichia*, but some specimens of *Syntrichia sarconeurum* were included in a population study (Skotnicki *et al.* 1999; 2000; 2004), and apparently would represent different populations, result of different colonizations.

Based on morphological diversity, taxonomic problems and the difficulty in distinguishing *Syntrichia* from other genera of Pottiaceae, especially under harsh environments like Antarctica, the focus of this work was to clarify the morphological characters that differentiate the Antarctic species of *Syntrichia*, the genetic diversity of the species of *Syntrichia* from Antarctica and the bipolarity of *S. caninervis*.

## **Materials and Methods**

### ***Sampling***

Field expeditions were carried out to obtain fresh material with support from the Brazilian, Argentine, Chilean and Polish Antarctic Programs (in the summers of 2014/2015, 2015/2016, 2016/2017, 2017/2018 and 2018/2019), through the project Evolution and Dispersion of Bipolar Antarctic Species of Bryophytes and Lichens (MCTI/CNPq / FNDCT - Transversal Action n° 64/2013). The samples were collected according to the walking method (Filgueiras *et al.* 1994) and the herborization followed Yano (1984). The collected material is deposited in the herbarium of the University of Brasília (UB).

Herbarium materials on loan (AAS, BM H, MUB, NY and SP)<sup>1</sup> were also analyzed, with the intention to contemplate the geographic distribution of species, including *typus*.

### ***Morphological study***

The identification of the material was based on Zander (1993), Flora of North America (2007), Ochyra *et al.* (2008) and Guerra *et al.* (2006). The samples were observed in a LEICA EZ4D stereoscopic microscope and DM 750 optical microscope. Semi-permanent slides were fixed with Hoyer (Anderson 1954) to preserve the core material. A 2% solution of potassium hydroxide (KOH) was used in material identification process (Zander 1993).

A total of three hundred and forty-four (344) samples were analyzed, being Two samples of *Syntrichia caninervis* (seen in MUB); Seven samples of *Syntrichia sp* 1 (confirmed after phylogenetic analyzes); Four of *Syntrichia caninervis* var. *gypsophila* (three views in MUB and one from AAS from Antarctica); Forty-three from *Syntrichia filaris*; One hundred and thirty eight from *S. magellanica*; Fifty-two from *S. sarconeurum*; and Ninety eight from *S. saxicola* (Appendix 1). Two types of materials were borrowed: *Syntrichia caninervis* and *S. sarconeurum* (Appendix 1).

### ***Molecular data analyses***

The samples (between 2 and 6 samples<sup>2</sup>) were washed with distilled water three times to remove the substrate and other impurities found with the material. Each specimen was packed in absorbent paper, in mini-envelopes in a container containing silica gel, for about 2 to 3 days, for drying.

DNA extraction followed Doyle & Doyle (1987), with modifications by Câmara (2010). DNA from herborized specimens were extracted using DNeasy® PowerPlant® Pro Kit (50) from Quiagen.

The amplified DNA region were based in Afonina *et al.* (2014) – ITS2, with primers Bryo5.8SF (Hartmann *et al.* 2006) e 26-antR (Hedenäs 2008). The polymerase chain reaction (PCR) followed the standard protocol for 50 µl of reaction, containing 5 µl

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<sup>1</sup> Acronyms according Thiers (2016).

<sup>2</sup> Because to material availability and/or difficulty in extraction and sequencing.

of Mg buffer, 5 $\mu$ l MgCl<sub>2</sub>, 2 $\mu$ l BSA, 4 $\mu$ l dNTP mix, 0.5 $\mu$ l Taq polymerase, 2.5 $\mu$ l of each primer, 2  $\mu$ l of DNA and 26.5  $\mu$ l of Milli-Q water.

The cycle used to amplify was 35 cycles, each formed for 30 seconds at 95° C; The initial cycle of 95° C for 1 minute; annealing in 45 seconds (we used: 51°C/54°C), with an extension of 1 minute at 72° C and a final extension of 72° C for 5 minutes.

The extraction and amplification of DNA were carried out at the Laboratory of Molecular Biology of Plants, Department of Botany, University of Brasília. DNA purification and sequencing were performed by Macrogen Inc. (Korea).

The sequences were edited and assembled in Geneious v5.3.6 (Kearse *et al.* 2012) and than aligned in CLUSTALX 2.1 (Higgins & Sharp 1988) with manual correction in PhyDE (v. 0.9971) (Müller *et al.* 2006). The insertions and deletions were coded as simple indel coding (Simmons & Ochoterena 2000) using the Seqstate ver. 1.4.1 (Müller 2005).

The Maximum Parsimony (MP) analyzes were performed in PAUP v.4.0a164 for Windows (Swofford 2002), through heuristic search and tree-bisection reconnection (TBR) branch swapping, with max. 10.000 trees generated. All characters in the matrix were weighted equally.

The Maximum Likelihood (ML) in RAxML v. 8 (Stamatakis 2014) the topology generated was viewed in Figtree v.1.4.2 software (Rambaut 2012).

The Bayesian analyzes (BI) in Mr Bayes v. 3.2.6 (Ronquist *et al.* 2012), with two parallel runs of Markov Monte Carlo (MCMC) chains were carried out with 5 million generations, one tree was sampled every 1000 generations. The first 25% of the trees were discarded as “burn-in”.

In addition to the DNA sequences obtained via extraction, sequences available at Genbank were used in our analysis. The outgroup *Tortula muralis* Hedw. was chosen based on Werner *et al.* (2002; 2004). All material is listed in the Table 1 (Appendix 2).

The support of the phylogenetic tree was evaluated through a non-parametric bootstrap (Felsenstein 1985) with 1.000 repetitions for Maximum Parsimony (MP) and Maximum Likelihood (ML) and by the posterior probabilities for Bayesian Inference (BI). The best model of evolution for each locus was obtained based on Akaike information criterion using jModeltest 3.06 (Posada 2008).

Bootstrap values from lower than 70 was considered low; values between 70 and 85 were considered moderate and values above 85 considered high. For BI only support above 0.95 was considered as good.

### ***Alternative Hypothesis Test - Teste SH***

We used the Shimodaira- Hasegawa (SH) Test (Shimodaira & Hasegawa 1999; Goldman *et al.* 2000) to statistically compare the topologies of two phylogenetic hypotheses, in which specific taxa are resolved as monophyletic or not. The constriction tree- simulation of monophyly, was built using Mesquite v.3.03 (Maddison & Maddison 2015), then uploaded to PAUP (Swofford 2002), where likelihood analysis was performed to find the ideal tree given the oriented constraint. Then the likelihood scores between the new and the original tree were compared using the SH test through PAUP (Swofford 2002), using 10,000 replications.

We decided to test the alternative hypothesis to test a possible monophyly between *Syntrichia caninervis* (samples from the genbank, form Russia and Mongolia) and the group named here *Syntrichia sp1* – composed of samples identified as *Syntrichia caninervis* var. *astrakhanica* (from GenBank, from Mongolia and Russia) plus the material identified as *Syntrichia caninervis* from Antarctica (Seymour Island) and Kazakhstan.

## **Results**

### ***Molecular studies***

The ITS2 marker was successful. We sampled specimens of *Syntrichia sp1*, *S. filaris*, *S. magellanica*, *S. sarconeurum* and *S. saxicola*.

*S. caninervis* var. *gypsophila* has not been sequenced (but has been evaluated for morphology). Despite attempts, the only material available (herbarium material) was degraded (DNA) and the place where it occurs in Antarctica it's inaccessible (is not part of the navigation logistics of Brazilian expeditions).

The alignment characteristics and maximum parsimony tree statistics are in the Table 2. Both Maximum Parsimony (MP) and Maximum Likelihood (ML) and Bayesian Inference (BI) presented similar topology, therefore, we present Bayesian tree with the values of all analyzes (Fig. 2).

**Table 2.** Alignment characteristics and maximum parsimony tree statistics.

Taxa included	53
<b>Nucleotide sites</b>	992
<b>Constant sites</b>	512
<b>Parsimony-uninformative sites</b>	128
<b>Parsimony informative sites</b>	352
<b>Length</b>	727
<b>Consistency index (CI)</b>	0.695
<b>Retention index (RI)</b>	0.890
<b>Best parsimonious trees</b>	714

Clade I (Fig. 2) - It is formed by samples of *Bryoerythrophyllum recurvirostrum* and by samples described by Skotnicki *et al.* (2004) as *Syntrichia sarconeurum* (11 and 12), these from the Princess Elizabeth Land region. The group is monophyletic with max support and positioned externally to the group composed of the species of *Syntrichia* (87/99/1).

Clade II - *Syntrichia sarconeurum* (samples 1-10), one sample from Chile and the others from Antarctica (Black Island, Galindez Island, Danco Coast, Drone Maund Land, Ros Island and Victoria Land). With high support in Maximum Parsimony and maximum in ML and BI analysis (Fig. 2).

Clade III - *Syntrichia saxicola* composed of samples from Antarctica (Robert Island, James Ross Island, Snow Island) and South Georgia Presents maximum support in all analyzes (Fig. 2).

Clade IV - *Syntrichia magellanica* is composed of samples from Antarctica (Danco Coast, Peninsula Trinity, King George Island, Livingston Island) and South Georgia. With high support in MP Analysis: 99 and maximum support in the others analyzes (Fig. 2).

Clade V - *Syntrichia caninervis* - Presents only samples from Genbank (Russia and Mongolia), there are no materials from Antarctica - the supposed antarctic

correspondence of *S. caninervis* corresponds to another clade. High support in the analysis of MP (99) and ML (96) and maximum support for BI (1) (Fig. 2).

Clade VI - *Syntrichia sp1* - includes genbank samples (Russia and Mongolia), Antarctica sample collected as *Syntrichia caninervis*, herbarium sample (Kazakhstan) identified as *Syntrichia caninervis*. It has high support for MP (99) and ML (99) analyzes and maximum for BI (1) (Fig. 2).

Clade VII - *Syntrichia filaris* is composed of samples from Antarctica (King George Island and Snow Island) and South Georgia presented maximum support for all analyzes (Fig. 2).

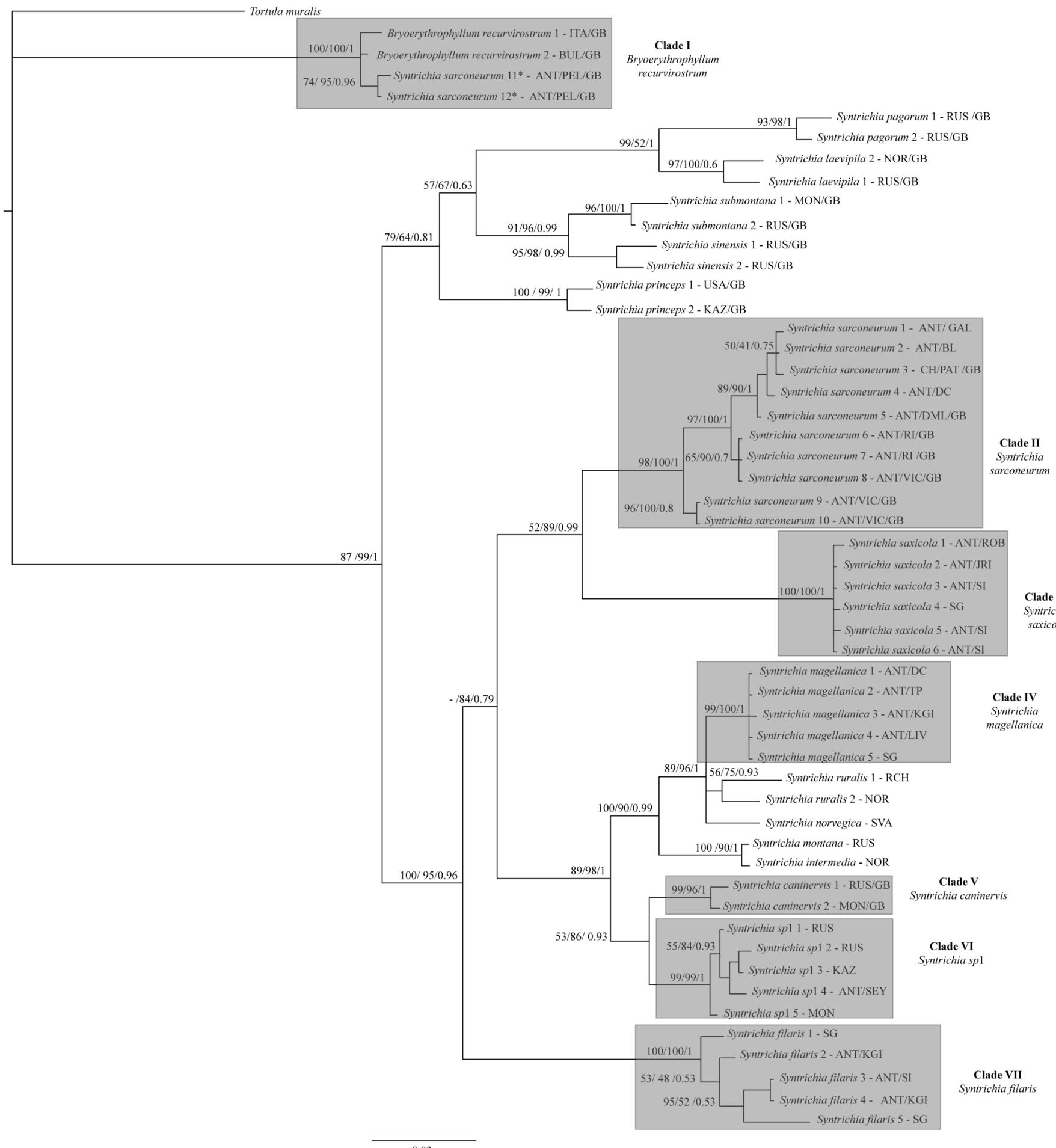


Figure 2 - Phylogram (consensus) generated by Bayesian Inference from the ITS2 matrix (with indel coding). Values at the branches indicate the support: Maximum Parsimony/Maximum Likelihood/Bayesian Inference. and (-) indicate no support. The samples analyzed are described in Appendix 2. The codes presented next to each name represent the locations of origin of the samples, separated by a bar when there is more than one specification, according to: ANT - Antarctica; BL - Black Island; BUL- Bulgaria; CH - Chile; DC - Danco Coast; DML - Dronning; Maud Land; GAL - Galindez Island; GB- GenBank; ITA- Italy; JRI - James Ross Island; KAZ - Kazakhstan; KGI- King George Island; LIV - Livingston Island; MON- Mongolia; NOR - Norway; PAT - Patagonia; PEL - Princess Elizabeth Land; PT - Trinity Peninsula; RCH- Czech Republic; RI - Ross Island; ROB - Robert Island; RUS - Russia; SVA- Svalbard; SEY - Seymour Island; SG - South Georgia; SI - Snow Island; USA - United States of America; VIC - Victoria Land. The asterisk (\*) represents the samples wrongly identified as another taxa and that in the phylogenetic reconstruction were grouped together with their correct clade.

### ***Alternative Hypothesis Test - Teste SH***

The Shimodaira-Hasegawa result was null about the monophyly hypothesis between *Syntrichia caninervis* and *Syntrichia sp1* as shown in the Table 3:

**Table 3.** Results from the Shimodaira-Hasegawa (SH) tests.

Constrained topology	Diff-InL	P
<i>Syntrichia caninervis</i> + <i>Syntrichia sp1</i>	2869.03724	0.0000*
* Statistically worse trees at $P < 0.05$ are marked with an asterisk (*).		

### ***Morphological investigation***

The species were observed for morphological characteristics, as specified in Table 4. The greatest number of information was sought to highlight the differences and/or similarities of each species and between them, reaching the characters shown in the Table 4 above - average gametophyte length, shape of the leaf, apex leaf, margin leaf, cells of basal region, marginal cells of median region, apex cells, costa, and cross section of the leaf.

**Table 4.** Morphological characterization<sup>34</sup> of *Syntrichia* species from Antarctica.

Characteristics	Species					
	<i>Syntrichia sp1</i>	<i>Syntrichia caninervis</i>	<i>Syntrichia filaris</i>	<i>Syntrichia magellanica</i>	<i>Syntrichia siccicola</i>	<i>Syntrichia sarconeurum</i>
		var. <i>gypsophila</i>				
<b>Gametophyte length</b>	0,4-1 cm	0,5- 1 cm	1,0-5 cm;	0,5-5 cm	1-4 cm	0,5 -1,5 cm
<b>Plant leaf shape</b>	Ovate (Plate 1 A, B)	Oblong- ovate to ovate- lingulate	Oblong- lanceolate to lanceolate (Plate 1 F)	Oblong to ovate- lingulate (Plate 1 K, L)	Lanceolate to oblong- lanceolate (Plate 1 T, U)	Lingulate to lanceolate (Plate 1 N, O)
<b>Leaf apex type</b>	Apex acute and	Piliferous	Acuminate.	Piliferous	Acuminate	Multiestratificat

<sup>3</sup> Characterization based on slide observations and literature descriptions (Ochyra *et al.* 2008).

<sup>4</sup> The most important characters are shown on Plate 1.

	hyaline hairpoint (Plate 1 A,B)	(short) hyaline and spiculous; Cuculate.	(Plate 1 F)	(short), reddish at the base. Apex usually dentate and cuculate (Plate 1 I)	(Plate 1 V)	e and propagulous (Plate 1 R,S)
<b>Leaf margin</b>	(Plate 1 Recurved D, E)	Recurved from the base until the apex	Plan (Plate 1 G, H)	Recurved to revolute (Plate 1 J,K,M)	Recurved to revolute (Plate 1 U, X,Y)	Plane (Plate 1 P, Q)
<b>Basal region cells</b>	Retangular	Retangular to lineares	Linear to retangular	Retangular to linear	Retangular to linear	Oblong – retangular
<b>Marginal cells median region</b>	Quadrat	Quadrat	Quadrat to retangular	Quadrat to oblute	Quadrat	Quadrat
<b>Apex cells</b>	Quadrat to hexagonal	Quadrat	Quadrat to isodiametric	Quadrat to oblute	Quadrat	Quadrat to retangular
<b>Costa</b>	Present, simple and excurrent (Plate 1 A,B);	Present and simple; Convex	Present and simple (Plate 1 F);	Present and simple (Plate 1 K);	Present and simple (Plate 1 T,U);	Present and simple (Plate 1 N);

	Convex dorsally and semi-terete (semicircular)	dorsally, present papillae (bifurcate)	Percurrent (sometimes subpercurrent ) Convex dorsally; Semi- terete to elíptica	Percurrent; Convex dorsally; Semi- terete	Percurrent to short- excurrent; Semi-terete	present papillae (bifurcate);
<b>Cross section of leaf</b>	Lamina bi- stratified in the upper half leaf (Plate 1 D, E); Presence of sub- stereids.	Lamina uni- stratified; Presence of guide cells (between 2 and 4), Substeroids or stereids and hydroid	Lamina uni- stratified (Plate 1 H); Presence of stereids, large guide cells (1-2) and Papillae.	Lamina uni- stratified (Plate 1 M); Presence of hydroid, dorsal stereids and guide cells (about 2).	Lamina uni- stratified (Plate 1 X,Y); Presence of dorsal (small) stereids, sclerimatous cells and guide cells (1-2) and Papillae.	Lamina uni- stratified; Presence of stereid or sub- stereids and 2-3 guide cells.



**Plate 1.** Morphology of *Syntrichia* Brid. species occurring in Antarctica. **A-E.** *Syntrichia* sp1: **A**, **B**. Leaf, general aspect; **C**. Detail of the transition of the cells from the region of the base of the leaf to the median region. **D, E.** Transversal section of the leaf in the upper portion (closest to the apex). **F-H.** *Syntrichia filaris*: **F**. Leaf, general aspect; **G**. Detail of the cells leaf from the median region; **H**. Transversal section of the leaf mid. **I-M.** *Syntrichia magellanica*: **I**. Showing the long and distinctive spurred hyaline awn (arrow); **J**. General aspect of leaf on the mid region; **K, L**. General aspect of leaf; **M**. Transversal section of the leaf mid. **N- S.** *Syntrichia sarconeurum*: **N, O**. General aspect of leaf; **P**. General aspect of leaf on the basal region; **Q**. General aspect of leaf on the mid region; **R, S**. General aspect of the apex (propagulous). **T-Y.** *Syntrichia saxicola*: **T**. General aspect of leaf; **U**. General aspect of the leaf (base and middle region); **V**. Detail of the apex; **W**. General leaf aspect on the middle region and margin; **X, Y**. Transversal section of the leaf. . Scales - D, E, G, M, P, Q, S: 100 µm; H, I, J, N, O, R, V, X, W: 200 µm; A, B, F, K, L, T, U: 500 µm.

### ***Key to the species of Syntrichia Brid. occurring in Antarctica***

1. Leaf apex propagulous..... ***Syntrichia sarconeurum* (Plate 1. R-S)**
1. Leaf apex not propagulous..... 2
  2. Presence of hyaline hairpoint at the leaf apex..... 3
    2. Absence of hyaline hairpoint at the leaf apex..... 5
      3. Leaf cells, in cross section, always unistratose, leaf margin revolute ..... ***Syntrichia magellanica* (Plate 1. M)**
      3. Leaf cells, presenting in cross section, some portion of bistratose..... 4
        4. Short hair point, occasionally bistratose..... ***Syntrichia caninervis* var. *gypsophila***
        4. Long hair point, bistratose in the upper portion of the leaf..... ***Syntrichia* sp1 (Plate 1. A-B, E)**
    5. Leaves short acuminate; leaf margins plane, dentate or serrate, crenate at the apex..... ***Syntrichia filaris* (Plate 1. F, H)**
    5. Leaves long acuminate; leaf margins recurved to revolute and entire..... ***Syntrichia saxicola* (Plate 1. X, Y)**

## **Discussion**

The analysis corroborated the monophyly of the genus. The Antarctic species of *Syntrichia* present good morphological and phylogenetic delimitation.

The marker ITS2 contemplated all the proposed species<sup>5</sup>, elucidating the affirmation of Afonina *et al.* (2014) to be a good region for amplification and analysis, in addition to having many substitutions that favor the distinction of species.

According to Ochyra *et al.* (2008) *Syntrichia caninervis* var. *caninervis* is reported for Antarctica, occurring on Seymour Island, but our molecular analyzes suggests that these samples (here we sequence 1 - *Syntrichia sp1* - sample 4) correspond to another *taxa*, distinct from that composed by *Syntrichia caninervis*. Still to corroborate this data, the SH test refutes the possibility of monophyly between *Syntrichia caninervis* and *Syntrichia sp1*.

Despite the *Syntrichia sp1* clade to present samples of *Syntrichia caninervis* var. *astrakhanica* has been described by Ignatov *et al.* (2002). Afonina *et al.* (2014) based on molecular data observed that *Syntrichia caninervis* var. *astrakhanica*, in fact, corresponds to a genetically differentiated taxon from *Syntrichia caninervis*, recommending the elevation of that taxon to the species level. The samples sequenced in the present study (Antarctica and Kazakhstan), have the same insertions and deletions as those of Genbank. However, the Antarctic material does not have gemmae, as described in the material type (of *Syntrichia caninervis* var. *astrakhanica*).

Unfortunately we didn't have access to the type. According to Ignatov *et al.* (2002), the type material would be MHA and the isotype MW, VOLG, LE. We tried to contact the institutions, getting feedback from LE, who informed us that he did not have the material. We believe that after the end of the pandemic period, we will be able to access the other herbaria for analysis of the material.

Despite this, we know that it is a different species of *Syntrichia caninervis* var. *caninervis*, and may be a new species yet to be described. We recommend that the variety

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<sup>5</sup> Except *Syntrichia caninervis* var. *gypsophila*, because the difficulty in accessing the place of origin and the age of the material available in herbarium.

*caninervis* is removed from the list of species in Antarctica, based on molecular data showing that it is another taxa.

As for *Syntrichia sarconeurum* (Fig. 2), the samples sequenced in this study (from Galindez Island, Black Island and Danco Coast ), are all grouped within the same clade, along with part of the samples used by Skotnicki *et al.* (2004) (Chile [Patagonia], Ross Island, Maund Land and, three from Victoria Land) - Clade II (Fig. 2), and separated from the others samples, also used by Skotnicki *et al.* (2004), here called Clade I, corresponding to *Bryoerythrophyllum recurvirostrum* (Fig. 2), composed of samples from Pricens Elizabeth Land.

Reviewing the distribution, according to Ochyra *et al.* (2008), *Bryoerythrophyllum recurvirostrum* occurs in Pricens Elizabeth Land, the same region where *Syntrichia sarconeurum* would also occur. We believe that the mistake is due to misidentification or DNA contamination.

We did not have access to the vouchers of Skotnicki *et al.* (2004), we tried all contacts with the author without success. According to Ochyra et al (2008) he also did not have access to the material during the elaboration of the Antarctic Moss Flora. We also did not find which herbarium the samples were deposited.

According to Ochyra *et al.* (2008) *Syntrichia sarconeurum* would be an endemic species to Antarctica. However, based on the analyzes performed here, the presence of a specimen from Patagonia (Fig. 2) is observed, which gives indications that the species is of Antarctic and sub-Antarctic occurrence.

With respect to morphology, it was already known that the species of *Syntrichia*, occurring in Antarctica, presents wide morphological variation among themselves (as shown in the table 4 and on the plate 1). Despite being so distinct from each other, the species actually correspond to the genus. The morphology corroborates the delimitation pointed at clades.

*Syntrichia sarconeurum* (Clade II, Fig. 2) is distinguished by the leaf shape (Lingulate to lanceolate - Plate 1 N-Q), in addition to the Multiestratificate and propagulous apex. *Syntrichia saxicola* (Clade III, Fig. 2) is distinguished by the leaf shape (Lanceolate to oblong-lanceolate) and acuminate apex (Plate 1 V). *Syntrichia*

*magellanica* (Clade IV, Fig. 2) despite presenting similarity to *Syntrichia sp 1* (Clade VI, Fig. 2), presents uni-stratified costa and reddish hair point at the base (Plate 1 K, M), as opposed to *Syntrichia sp 1* that presents Lamina bi-stratified in the upper half leaf (Plate 1 A, B and D). *Syntrichia filaris* presents plane margin and acuminate apex (Plate 1 F).

## Conclusions

We present new data and suggestions for the genus *Syntrichia*, aiming to contribute to the studies of the genus.

We also support updating the status of *Syntrichia sarconeurum*. We propose that this species, previously considered endemic to Antarctica, now has a broader distribution, including the Chilean Patagonia region, therefore being of Antarctic and Sub-Antarctic distribution, based on molecular data, revision of samples, including material type.

We propose that *Syntrichia caninervis* var. *caninervis* is removed from the Antarctic moss flora, based on molecular data and revision material type.

No changes are suggested for *Syntrichia filaris*, *Syntrichia magellanica* and *Syntrichia saxicola*. Therefore occur in Antarctica, corroborated by morphological and molecular data, the following species of *Syntrichia* are: *Syntrichia filaris*, *Syntrichia magellanica*, *Syntrichia saxicola* and *Syntrichia sarconeurum*.

We suggest that *Syntrichia sp1* be included in the list of Antarctic moss species, as a bipolar species, pending visualization of the material type.

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## **Supplementary Material**

### *Appendix I. Analyzed Material*

*Syntrichia caninervis* Mitt.

Specimen examined – **TIBET** - Thomson 174 (BM) –Type. **SPAIN** – Cano 10977 (MUB).

*Syntrichia sp 1.*

Specimens examined – **ANTARCTIC: EAST OF THE ANTARCTIC PENINSULA** - **Seymour Island** – R. I. L. Smith 07886A (AAS), 07892(AAS), 07893B (AAS), 07891 (AAS), 07889A (AAS); J. Bordin 2312 (SP). **KAZAKHSTAN** – Magill 10705 (H).

*Syntrichia caninervis* var. *gypsophila* Mitt.(G. Roth) Ochyra

Specimen examined – **ANTARCTICA: ANTARCTIC PENINSULA, Trinity Peninsula** - G. Branding 00010 (AAS). **SPAIN** – Cano 10976 (MUB); Guerra 3826 (MUB), 3827 (MUB).

*Syntrichia filaris* (Müll. Hal.) R.H. Zander

Specimens examined – **ANTARCTICA: ARCHIPELAGO SOUTH SHETLANDS** - **Deception Island** - B.J. Taylor 243 (BM), J. Bordin 2530 (UB), P.E.A.S Câmara 3612 (UB), R.I.L. Smith 3621 (BM), 11346 (AAS), 11351(AAS), 11407 (AAS), R. Ochyra 2802/80 (NY); **King George Island** - B. Jablonski M186 (NY), C.M. Bernardo 123 (UB), G.T. Prance 28607 (NY), 28608 (NY), P.P.U. Aquino 52 (UB), R. Ochyra 2455/80 (NY), No 458/80 (NY), 4860/79 (NY), 5062/79 (NY); **Elephant Island** - J. Bordin 2931 (UB), 3009 (UB), J.S. Allison 104 (BM); **Livingston Island** - R.I.L. Smith 3775 (BM); **Robert Island** - J. Bordin 2692 (UB), R.I.L. Smith 916 (BM); **Snow Island** – P.P.U. Aquino 184 (UB). **EAST OF THE ANTARCTIC PENINSULA** - **Vega Island**- R.I.L. Smith 07831 (AAS); **ARCHIPELAGO Southern Orkney** - **Coronation Island** - R.I.L. Smith 08021 (AAS), 08024A(AAS); **Ilha Laurie** - N. Bellisio 11755 (NY), 11786 (NY), 11789 (NY), N. Bellisio 11691 (NY), 11694 (NY), 11796 (NY), 11697 (NY); **Signy Island**- M.L. Holdgate 143 (BM), 765b (BM), 767d (BM), R.I.L. Smith 646b (BM), R.N. Heywood 62 (BM). **SOUTH GEORGIA ISLAND**: B.G. Bell 871 (NY), N.V. Jones 3579A (BM), S.W. Greene 637 (NY), 2268 (NY), 3459 (BM), W.J.L. Sladen 47 (BM).

*Syntrichia magellanica* (Mont.) R.H. Zander

Specimens examined – **ANTARCTICA: ARCHIPELAGO SOUTH SHETLANDS** - **Ardley Island**- H.C. Oliveira 2496Pop.3 (UB); **Cormorant Island**- A. Scott 10 (BM), 41 (BM); **Cuverville Island** - R.I.L. Smith 11950 (AAS); **Deception Island** - H.C. Oliveira 2560c (UB), 2561c (UB), J. Bordin 2507 (UB), 2524 (UB), J.V. Mundim 636 (UB), P.E.A.S Câmara 3651 (UB), 3585 (UB), 3586 (UB), 3671 (UB), R.I.L. Smith 11523B (AAS), 11919B (AAS), 11917 (AAS); **Greenwich Island**- D.C. Lindsay 705 (BM); **Geologist Island** - P.E.A.S Câmara 4024 (UB), 4030 (UB); **Livingston Island** - D.C. Lindsay 170 (BM), 289 (BM), 322 (BM), 348 (BM), 359 (BM), H.C. Oliveira 2567b (UB), 2567e (UB), 2570a (UB), 2571c (UB), J. Bordin 2551 (UB), 2553 (UB), 2558 (UB); **Pinguin Island** – J. Bordin 2454 (UB) 2460 (UB); **King George Island** - C.M. Bernardo 54 (UB), 115 (UB), 128 (UB), 132

(UB), 183 (UB), 193 (UB), 203 (UB), 214 (UB), *D.C. Lindsay* 692b (BM), 667 (BM), *D.K. Henriques* 187 (UB), 256 (UB), *J. Bordin* 2419 (UB), 2421 (UB), 2481 (UB), 2485 (UB), 2487 (UB), *J.V. Mundim* 158 (UB), 204 (UB), 409 (UB), 432 (UB), 474 (UB), *P.E.A.S Câmara* 3312A (UB), 3314A (UB), 3339B (UB), 3345 (UB), 3352 (UB), 3354 (UB), 3356 (UB), 3358 (UB), 3396 (UB), 3407 (UB), 3445 (UB), 3450 (UB), 3519 (UB), 3524 (UB), 3528 (UB), 3541 (UB), 3551 (UB), 3555 (UB), 3570 (UB), *P.P.U. Aquino* 41 (UB), 45 (UB), 46 (UB), 65 (UB), 67 (UB), *M. Carvalho-Silva* 2055 (UB), 2060a (UB), 2065 (UB), 2095 (UB), 2096 (UB), 2102A (UB), 2104 (UB), 2109 (UB), *T.S. Dantas* 129 (UB), 130 (UB), 131 (UB), 618 (UB), *R. Ochyra* 1556/80 (AAS, NY), 4990/79 (AAS, NY), 2128/80 (NY); **Robert Island** – *J.V. Mundim* 1338 (UB); **Snow Island** – *P.P.U. Aquino* 177 (UB), 186 (UB). **ELLSWORTH LAND - Thurston Peninsula-** *R. Stan* 84 (NY). **QUEEN MARY LAND - Costa Knox - G.A. Llano** 25 (NY). **EAST OF THE ANTARCTIC PENINSULA - Hope Bay - C.G. Brading** 30b (BM); **James Ross Island - M. Kitaura** 2844 (UB), 2860 (UB), 2872 (UB), 2873 (UB), 2874 (UB), 2987 (UB), 3202 (UB). **WEST OF THE ANTARCTIC PENINSULA- ARCHIPELAGO MELCHIOR-** *Alpha Island- R.I.L. Smith* 4106 (BM); *P.A. Siple* 345.1 (NY), *M. Lamb* 7897 (NY); **Danco Coast - Primavera Base-** *J. V. Mundim* 720 (UB), 1083 (UB), *T.S. Dantas* 544 (UB); **Fallieres Coast, Adelaide Island - R.I.L. Smith** 2259 (BM); **Dion Island- R.I.L. Smith** 587 (BM); **COSTA GRAHAM- Argentine Island- R.I.L. Smith** 3319 (BM), **Petermann Island- A. Scott** 105b (BM). **SOUTH GEORGIA ISLAND:** *B.G. Bell* 648 (NY), 3434 (NY), *G.C.S. Clarke* 588 (BM), 620 (BM), *R.E. Longton* 313 (NY), *L.W. Greene* 537 (NY), 1648b (NY), 3461c (NY), *S.W. Greene* 528(BM), 639 (BM), 646 (NY), 780 (BM), 2176 (NY), 3261B (NY), 3438 (BM). **KERGUELEN ISLAND:** 181 (BM);

*Syntrichia sarconeurum* Ochyra & R.H. Zander

Specimens examined – **ANTARCTICA: ARCHIPELAGO SOUTH SHETLANDS - Deception-** *R.E. Longton* 00024 (AAS). **EAST OF THE ANTARCTIC PENINSULA - Bunger Hills-** *E.S. Korotkivicz* 26 (NY), *M.M. Hollerbach* 11/312 (NY); **Cordilheira Shackleton – R.I.L. Smith** 10000 (AAS); **Cockburn Island-** Cockburn nº 4 (NY-Mitten) - Type; **James Ross Island - R.I.L. Smith** 07573(AAS); **Ross Island- C.R. Lewis** hp-10 (NY), *E. Schofield AA-10* (NY), *G.A. Llano* 18 (NY), 2217a (NY), 2123a (NY), 2168 (NY), 2169 (NY), 2170 (NY), 2172 (NY), 2173 (NY), 2256 (NY); **Nunatak west of Laserman-** *K.K. Markov* 1 (NY); **MARIE BRYD LAND - Lichen Peak - P.A. Siple** G1 (NY), 7a (NY), 43 (NY), 46 (NY), 51 (NY). **WEST OF THE ANTARCTIC PENINSULA- Danco Coast-** *R.I.L. Smith* 1945 (BM), 1967 (BM); **Graham Coast –R.W.M. Corner** 00783 (AAS); **Wilhelm Archipelago - Uruguay Island-** *R.I.L. Smith* 04271 (AAS), *R.W.M. Corner* 602 (BM); **Galindez Island-** *R.E. Longton* 1329 (BM), *R.W.M. Corner* 404a (NY), 466a (BM); **Black Island-** *R.W.M. Corner* 832 (BM). **QUEEN MAUNDS LAND (St. 46 F)-** NY 3105257 (NY), NY 3105258 (NY), NY 3105260 (NY), NY 3105261 (NY), NY 3105259 (NY), *G.T. Bowra* Z92a (NY). **VICTORIA LAND-** *G.A. Llano* 2196 (NY), 2198a (NY), *E. Schofield AA-5* (NY), AA-8 (NY), *K.A.J. Wise sn* (NY), *W.C. Steere* 65/55 (NY), 65/16 (NY); **Shackleton Glacier-** *K.A.J. Wise* 18 (NY); **Barrett Glacier (Shackleton Glacier region) -K.A.J. Wise** 16 (NY). **ILHAS ÓRCADAS DO SUL: Signy Island - R.I.L. Smith** 00658 (AAS), 08055 (AAS);

*Syntrichia saxicola* (Cardot) R.H. Zander

Specimens examined – **ARGENTINA:** Puerto Valdez – *H. Roivainen* s.n. (H). **ANTARCTICA: SOUTH SHETLANDS ARCHIPELAGO-** Ardley Island- *H.C. Oliveira* 2496 Pop.4 (UB); Deception Island- *R.I.L. Smith* 11269 (AAS), 11424 (AAS), *J. Bordin* 2492 (UB), 2502 (UB), 2515 (UB), 2528 (UB), 2528 (UB), 2532 (UB), 2533 (UB), *H.C. Oliveira* 2558a (UB), 2558b (UB), 2560a (UB), 2560d (UB), 2560e (UB), *J.V. Mundim* 647 (UB), 654 (UB), *P.E.A.S. Câmara* 3585 (UB), 3620 (UB), 3646 (UB), *P.P.U. Aquino* 144 (UB); Elephant Island- *J. Bordin* 2903 (UB), 2908 (UB), 2965 (UB), 2969 (UB), 3023 (UB), 3096 (UB); Geologist Islands - *P.E.A.S Câmara* 4024 (UB), 4030 (UB), 4051 (UB), 4054a (UB), 4061 (UB), 4062 (UB), 4064 (UB); James Ross Island - *R.I.L. Smith* 07400B (AAS), 07633 (AAS), 07336D (AAS), 07824A (AAS), *M. Kitaura* 2844 (UB); Livingston Island - *D.C. Lindsay* 321 (BM), *R.I.L. Smith* 3804 (BM); King George Island - *C.M. Bernardo* 239 (UB), *D.C. Lindsay* 823 (BM), *D.K. Henriques* 227 (UB), *J.V. Mundim* 108 (UB), 121 (UB), 165 (UB), 170 (UB), 171 (UB), 346 (UB), 462 (UB), 472 (UB), *P.E.A.S Câmara* 3339b (UB), 3420 (UB) 3407 (UB), 3584 (UB), *J. Bordin* 2446 (UB), *M. Carvalho-Silva* 2047 (UB), 2089 (UB), 2095 (UB), 2069 (UB), 2102a (UB), *P.P.U. Aquino* 45 (UB), 47 (UB), *H.C. Oliveira* 2590b (UB), 2590e (UB); Gibbs Island- *J.P. Baylis* 175a (BM), 216 (BM); Ilha Robert- *H.C. Oliveira* 2535b (UB), 2535c (UB), 2535d (UB); Snow Island – *D.V. Valente* 1653 (UB), 1657 (UB), 1684 (UB), 1686 (UB), 1687 (UB), *P.P.U. Aquino* 185 (UB), 187 (UB), 188 (UB). **GRAHAM LAND - O'Brian Island-** *J.P. Baylis* 11a (BM), 18 (BM). **WEST OF THE ANTARCTIC PENINSULA- Danco Coast-** *V. Komarkova* B-112803 (NY). **VICTORIA LAND-** *W.C. Steere* 65/46 (NY). **ORKNEYS ARCHIPELAGO:** Signy Island- *M.W. Holdgate* 155b (BM), *R.E. Longton* 1145 (BM). **SOUTH GEORGIA ISLAND:** *B.G. Bell* 976 (NY), 1163 (H), *C.J. Barrow* 180 (NY), 181 (H), *S.W. Greene* 627 (H, NY), 638 (H, NY), 956 (BM), 2324 (H, BM, NY), 2420 (BM), 2764c (BM), 3502a (BM), *R.I.L. Smith* 4973 (BM), 4975(BM).

## Appendix 2

Table 1. List of species used in the analysis of the ITS2 region, locality where it was collected, code used in the analyzes, number of collector, herbarium where it is deposited and Genbank accession number, respectively. The sequences obtained in this study have not yet been submitted to Genbank, but it is being provided.

Species	Code	Locality	Collector number or herbarium record	Herbarium	Genbank ITS2	Note
<i>Bryoerythrophyllum recurvirostrum</i>	<b>1</b>		<i>MUB 15334</i>	MUB	AY437131	
	<b>2</b>	Bulgaria	<i>Kucera 13997</i>		KY406826	
(Hedw.) PC Chen	<i>Syntrichia sarconeurum</i> 11*	Antarctica - Vestfold Hills, Princess Elizabeth Land			AY616857	Although identified as <i>Syntrichia sarconeurum</i> corresponds to <i>Bryoerythrophyllum recurvirostrum</i>
	<i>Syntrichia sarconeurum</i> 12*	Antarctica - Mossel Lake, Princess Elizabeth Land			AY616855	Although identified as <i>Syntrichia sarconeurum</i> corresponds to <i>Bryoerythrophyllum recurvirostrum</i>
<i>Syntrichia caninervis</i> Mitt	<b>1</b>	Russia	<i>Ignatov &amp; Abakarova 11-102</i>	MHA	KM381996	
	<b>2</b>	Mongolia	<i>Ignatov 01-663</i>	MHA	KM381997	
<i>Syntrichia sp1</i>	1	Russia	<i>Fedosov 05-547</i>	MW	KM382037	
	2	Russia			KM381998	
	3	Kazakhstan	<i>Magill 10705</i>	AAS		
	4	Antarctica – Seymour	<i>Bordin 2312</i>	SP		
	5	Mongolia	<i>Ignatov 01-669</i>	MHA	KM381999	
<i>S. filaris</i> (Müll. Hal.) R.H. Zander	1	South Georgia	<i>Greene 2268</i>	NY		
	2	Antarctica	<i>Aquino 52</i>	UB		
	3	Antarctica- Snow Island	<i>Valente 1684</i>	UB		
	4	Antarctica	<i>Henriques 926</i>	UB		
	5	South Georgia	<i>Greene 3459</i>	BM		

<i>S. intermedia</i> Brid		Norway	TRH-672989, <i>Jordal</i>	TRH	KC333244
<i>S. laevipila</i> Brid.	1	Russia	<i>Ignatov &amp; Ignatova</i> 05-598	MW	KM382000
	2	Norway	<i>TRH-671896,</i> <i>Johnsen, J. I.;</i> <i>Jordal, J. B.</i>	TRH	KC333245
<i>Syntrichia magellanica</i> (Mont.)	1	Antarctica – Danco Coast	<i>Mundim 789</i>	UB	
R.H. Zander	2	Antarctica- Península Trinity	<i>Oliveira 2645d</i>	UB	
	3	Antarctica – King George Island	<i>Carvalho-Silva 2060<sup>a</sup></i>	UB	
	4	Antarctica – Livingston Island	<i>Oliveira 2567b</i>	UB	
	5	South Georgia	<i>Longton 313</i>	NY	
<i>Syntrichia montana</i> Nees		Russia	<i>Fedosov 10-2-335</i>	MW	KM382003
<i>Syntrichia norwegica</i>		Svalbard	<i>Frisvoll</i>		KC333246
<i>Syntrichia pagorum</i> (Milde) J.J. Amann	1	Russia	<i>Ignatov et al. 05-1540</i>	MW	KM382005
	2	Russia	<i>Afonina s.n.</i>	LE, MHA	KM382009
<i>Syntrichia princeps</i> (De Not) Mitt.	1	USA	<i>Ignatov s.n.</i>	MHA	KM382013
	2	Kazakhstan	<i>Neshataeva s.n.</i>	MHA	KM382014
<i>Syntrichia ruralis</i> (Hedw.) F. Weber & D. Mohr	1	Czech Republic	<i>Kosnar 1035</i>	CBFS	KP307511
	2	Norway	<i>TRH-673036</i> <i>Reiso</i>	TRH	KC333249
<i>Syntrichia sarconeurum</i> Ochyra & R.H. Zander	1	Antarctica - Galindez Island			
	2	Antarctica – Black Island			
	3	Chile - Patagonia			AY616859
	4	Antarctica- Danco Coast- Liechfield Island	<i>R.I.L. Smith 1967</i>		
	5	Antarctica - Henriksenskjera, Dronning	<i>10066a</i>	AAS	AY616858

Maud Land							
6	Antarctica - Ross Island			AY616852			
7	Antarctica - Ross Island			AY613332			
8	Antarctica - Victoria Land			AY616851			
9	Antarctica - Victoria Land	09973	AAS	AY616853			
10	Antarctica - Victoria Land	<i>R.I.L. Smith</i> 9766		AY613333			
<b><i>Syntrichia saxicola</i> (Cardot.) R.H. Zander</b>							
1	Antarctica- Robert Island	<i>Oliveira</i> 2535a	UB				
2	Antarctica- James Ross	<i>Kitaura</i> 2860	UB				
3	Antarctica- Snow Island	<i>Valente</i> 1653	UB				
4	South Georgia	<i>Greene</i> 2324	H, BM, NY				
5	Antarctica - Snow Island	<i>Valente</i> 1657	UB				
6	Antarctica - Snow Island	<i>Valente</i> 1658	UB				
<b><i>S. sinensis</i> (Müll. Hal.) Ochyra</b>							
1	Russia	<i>Afonina</i> 5306	LE	KM382023			
2	Russia	<i>Chandolaz, Ignatov et al.</i> 06-2825	MHA	KM382022			
<b><i>Syntrichia submontana</i> (Caldo.) Ochyra</b>							
1	Mongolia	<i>Ignatov</i> 01-658	MHA	KM382039			
2	Russia	<i>Abakarova &amp; Ignatov</i> 11-477	MHA	KM382028			
<b><i>Tortula muralis</i> Hedw.</b>		Bosnia and Herzegovina	<i>Košnar</i> 1363	CBFS	JN544813		
					<b>Outgroup</b>		

## CAPÍTULO 2

### **Small areas and small plants: Updates on Antarctic bryophytes**

Paper accepted for publication in Acta Botanica Brasilica.



## **Small areas and small plants: Updates on Antarctic bryophytes**

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## **Abstract**

Snow Island is part of the South Shetland Archipelago in Antarctica. Most of its surface is permanently covered by snow, yet it has an important paleobotanical. There are no protected areas on the island and no recent data regarding its vegetation. This study aimed to collect and identify fresh samples of bryophytes from President Head Peninsula of Snow Island. Samples were collected during the summers of 2015 and 2018. Among the 24 bryophyte species identified in this work, 19 are new occurrences for Snow Island, bringing the total known for the island to 29 species. The most diverse family is Pottiaceae, with four species of two genera, followed by Bryaceae and Polytrichaceae, with three species each. The results show that the diversity of mosses on Snow Island is greater than previously reported. This study has demonstrated the region's importance and the 190% increase in Snow Island species number. This study also updated the Antarctic distribution of some species.

**Keywords:** Bryophyta, Flora, Maritime Antarctica, new occurrence, South Shetlands.

## **Introduction**

Antarctica is the most distant and isolated continent, and is considered the driest and coldest region on the planet (Bargagli 2005; Cassano 2013). It is inhabited by extremophile organisms that are able to establish themselves and survive the harsh conditions (Longton & Holdgate 1979). These characteristics make Antarctica a unique place for the development of scientific research in diverse areas such as environmental science and biodiversity (Câmara *et al.* 2017). The continent itself has an area of over 14 million km<sup>2</sup>, of which only 0.3% is ice free during the summer season and representing potential substrata for local vegetation (Fox & Cooper 1994). The South Shetlands is an archipelago that covers an area of 3687 km<sup>2</sup>. It is comprised of 11 main islands and is located north of the Antarctic Peninsula. According to Lewis-Smith (1984) and Ochyra *et al.* (2008), the archipelago belongs to the northern Maritime Antarctica geobotanical zone characterized by a cold moist climate with milder temperatures during summer and annual precipitation ranging 350–550 mm, mostly concentrated as rain during the warmer season. This zone also holds greater botanical diversity than other Antarctic regions zones (Ochyra *et al.* 2008).

Antarctic vegetation is predominantly cryptogamic, with 116 species of bryophytes, 500 species of lichens and three species of flowering plants having been reported for the continent (Ochyra *et al.* 2008; Ellis *et al.* 2013a; 2013b; Sollman 2015; Câmara *et al.* 2019a). Although the bryophyte flora of Antarctica has been the focus of various researchers in the past, and many aspects of its composition have been elucidated (Putzke & Pereira 1990; Bednarek-Ochyra *et al.* 2000; Ochyra *et al.* 2008), there have been few efforts to investigate the genetic diversity and true identity of many of the taxa occurring on the frozen continent.

Recent studies regarding moss diversity and biogeography integrating a molecular approach have shown that bryophytes, especially mosses, have much yet to be discovered

(Biersma *et al.* 2017; 2018a; 2018b; Câmara *et al.* 2018; 2019a; 2019b). Recent sampling and site-oriented investigations have also demonstrated that moss species distributions remain a subject of discussion and that the South Shetland Archipelago, which bears most of the ice-free area in Maritime Antarctica, may hold greater diversity than that described in traditional literature (Câmara *et al.* 2017; Henriques *et al.* 2018; Câmara *et al.* 2020). Although the flora of Antarctica has been published, new occurrences continue to be found. In addition, treatments of local floras are almost non-existent, and are known to be fundamental, as they allow for better monitoring of changes that vegetation may be experiencing in the face of climate change.

Climate change has a strong impact in Antarctica, such as changes in the original composition of organisms (Amesbury *et al.* 2017; Robinson *et al.* 2018), disappearance of species, competition with new species and decreased availability of habitats and microhabitats (Hogg *et al.* 2006, Bokhorst *et al.* 2008, Glime 2017).

According to Turner *et al.* (2009), climate change is more perceptible in the region of the South Shetland Archipelago and the Antarctic Peninsula, which are areas of high concentrations of mosses and lichens (Ochyra *et al.* 2008). Even so, studies that monitor vegetation and local floras are scarce.

Snow Island is part of the South Shetland Archipelago. It is located southwest of Livingston Island and separated from it to the northeast by the Morton Strait. Snow Island has an area of approximately 162.57 km<sup>2</sup>, with its surface almost completely covered by snow during most of the year, and its highest point is 305 m a.s.l. (López-Martínez *et al.* 2016). President Head Peninsula is located in the northeast of Snow Island and is an extremely rich geological and paleobotanical site — it is considered an important source of data for understanding the expansion and paleobotany of Antarctica (Phillipe *et al.* 1995; Cantrill

1997; Cantrill *et al.* 1998). Studies have characterized the paleoenvironment and the past flora of the Peninsula, especially regarding the Mesozoic, demonstrating the Island's connections through palynological and biostratigraphical data with the Byers Peninsula group of Livingston Island (Duane 1996; Torres *et al.* 1997; Cantrill *et al.* 1998). Other studies have also demonstrated the Early Cretaceous flora connection among Australia, South America and Antarctica through paleopalynological data, highlighting the scientific importance of Snow Island (Cantrill 1997; Cantrill *et al.* 1998).

Although Snow Island has its scientific relevance, it has no protected areas, not even an Antarctic Specially Protected Area (ASPA) or an Antarctic Specially Managed Area (ASMA). Even though several studies have revealed the fossil flora (Cantrill 1997; Torres *et al.* 1997; Cantrill *et al.* 1998), there are no current projects focusing on the Island's extant flora, especially the current bryoflora. The only available information regarding liverworts from Snow Island is a list produced by Bednarek-Ochyra *et al.* (2000). The mosses of the Island were treated by Ochyra *et al.* (2008), who reported only ten species collected on President Head Peninsula.

Due to its scientific relevance, and knowing that recent studies reported greater diversity than other areas already surveyed in Antarctica, we collected and studied fresh material from President Head Peninsula with the aim of increasing and updating knowledge of the botanical diversity of Snow Island. Thus, we provide an updated list of species along with an identification key and illustrations.

## **Materials and Methods**

Field expeditions to Snow Island occurred during the Antarctic summers of 2015 and 2018, in snow free areas on President Head Peninsula (Fig. 1). Collections were made

according to Yano (1984), and vouchers were deposited in herbarium UB (University of Brasilia). Samples were observed and studied under a stereomicroscope (Leica EZ4 HD) and an optical microscope (Leica DM 750). Semi-permanent slides were made using Hoyer solution (Anderson 1954) to investigate taxonomically useful structures. Images of important structures and details were taken with a camera (Leica 170 HD) coupled to an optical microscope (Leica DM 750). Identifications followed Ochyra *et al.* (2008) and the classification system adopted was that of Goffinet *et al.* (2009). All terminology followed Magill (1990). Voucher information is available in Appendix 1. The list presented here combines the data resulting from our collections with the list of Ochyra *et al.* (2008).

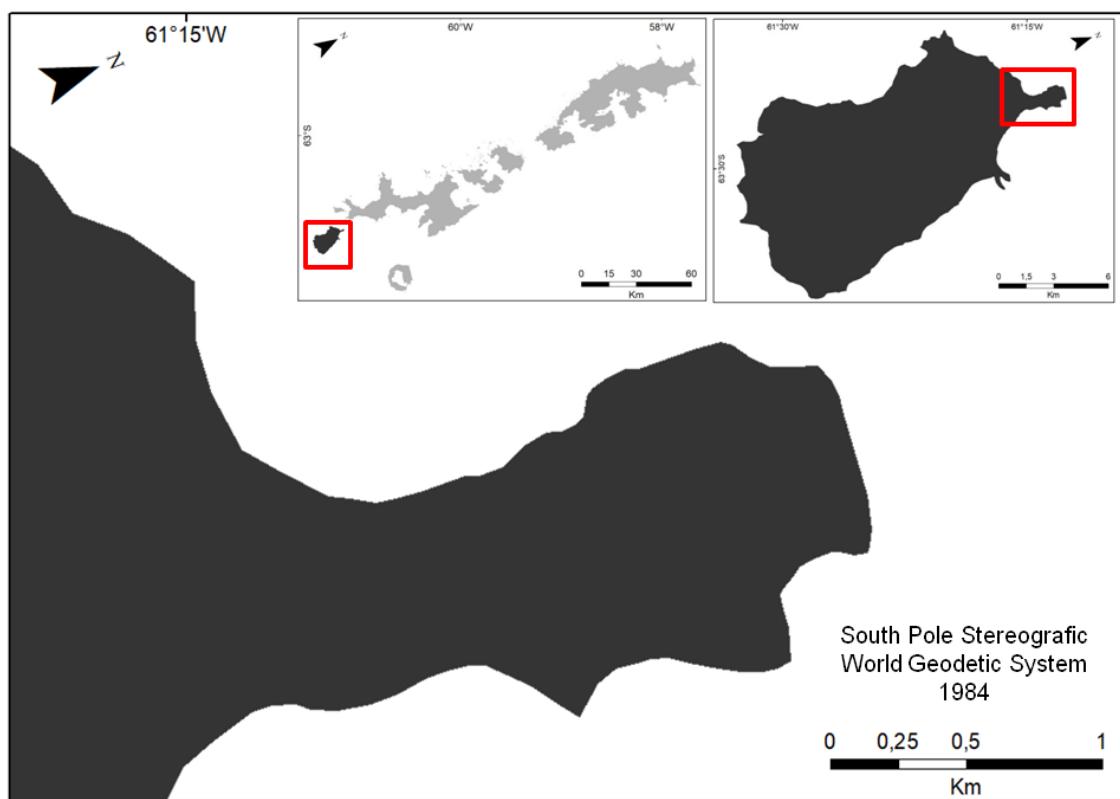


Figure 1. Map of President Head, Snow Island, South Shetlands, Antarctica. The indicated region represents Snow Island, and President Head.

## Results and Discussion

One hundred and eighty-five samples were collected on Snow Island during the Antarctic summers of 2015 and 2018, representing a total of 24 species of bryophytes distributed among 16 genera and 12 families. Among the 24 species (Table 1), 19 are new occurrences for Snow Island. The most diverse family is Pottiaceae, with four species in two genera, followed by Bryaceae and Polytrichaceae, with three species each.

**Table 1.** List of moss families and species collected in Snow Island during Antarctic summers of 2015 and 2018. Classification follows Goffinet *et al.* (2009). New Occurrences are indicated by an asterisk.

Families	Species
Amblystegiaceae	<i>Drepanocladus polygamus</i> (Schimp.) Hedenäs *
	<i>Sanionia uncinata</i> (Hedw.) Loeske
Andreaeaceae	<i>Andreaea regularis</i> Müll. Hal.
Bartramiaceae	<i>Bartramia patens</i> Brid.
Brachytheciaceae	<i>Brachythecium austroglareosum</i> (Müll. Hal.) Kindb. * <i>Brachythecium austrosalebrosum</i> (Müll. Hal.) Kindb. *
Bryaceae	<i>Bryum argenteum</i> var. <i>argenteum</i> Hedw. * <i>Bryum orbiculatifolium</i> Cardot & Broth. <i>Bryum pseudotriquetrum</i> (Hedw.) G. Gaertn., B. Mey. & Scherb. *
Calliergonaceae	<i>Warnstorffia fontinaliopsis</i> (Müll. Hal.) Ochyra *
	<i>Warnstorffia sarmentosa</i> (Wahlenb.) Hedenäs *
Ditrichaceae	<i>Distichium capillaceum</i> (Hedw.) Bruch & Schimp. * <i>Ceratodon purpureus</i> (Hedw.) Brid. *
Grimmiaceae	<i>Schistidium lewis-smithii</i> Ochyra *

	<i>Schistidium praemorsum</i> (Müll. Hal.) Herzog *
Mniaceae	<i>Pohlia cruda</i> (Hedw.) Lindb. *
	<i>Polytrichastrum alpinum</i> (Hedw.) G.L. Sm.
Polytrichaceae	<i>Polytrichum juniperinum</i> Willd. ex Hedw.*
	<i>Polytrichum piliferum</i> Hedw.
	<i>Bryoerythrophyllum recurvirostrum</i> (Hedw.) P.C. Chen *
Pottiaceae	<i>Syntrichia filaris</i> (Müll. Hal.) R.H. Zander *
	<i>Syntrichia magellanica</i> (Mont.) R.H. Zander *
	<i>Syntrichia saxicola</i> (Cardot) R.H. Zander *
Rhabdoweisiaceae	<i>Hymenoloma grimmiaeum</i> (Müll. Hal.) <i>Ochyra</i> *

The considerable number of new moss occurrences found by the present study can be explained by the lack of detailed studies focused on Snow Island. President Head Peninsula is the main ice-free area of the Island and past studies mainly focused on just part of it. The present research surveyed the peninsula and its surrounding areas, wherever soil and rocks were available for examination. Even though the focus of our work was to perform a broader sampling of the Island, we were limited by logistics and weather conditions. Nevertheless, 19 new occurrences were found.

Our results are congruent with recent local studies of Antarctic flora (Câmara *et al.* 2017; Henriques *et al.* 2018) and revealed that the number of species occurring in Snow Island was underestimated in previous works. The diversity of local bryophytes in Antarctica is still worth investigating. Table 2 provides information on the distribution of all species of bryophytes found to date on Snow Island, including those cited by Ochyra *et al.* (2008), but not collected by the present study.

**Table 2.** Distribution data of the species present on Snow Island.

Species	Distribution
<i>Drepanocladus polygamus</i> (Schimp.) Hedenäs	Occurring in few localities in Antarctica. In the South Shetland Archipelago it is cited only for King George and Livingston Islands (Ochyra <i>et al.</i> 2008) and in this study it was found in Snow Island.
<i>Sanionia uncinata</i> (Hedw.) Loeske	Is widely distributed and very common in Antarctica and it had been already reported for Snow Island in President Head Plateau (Ochyra <i>et al.</i> 2008).
<i>Andreaea regularis</i> Müll. Hal.	Widely distributed in the Antarctic, especially in South Shetland Archipelago and was reported for Snow Island in President Head (Ochyra <i>et al.</i> 2008).
<i>Bartramia patens</i> Brid.	Widely distributed in maritime Antarctica and common in the South Shetland Archipelago. Was reported for Snow Island in President Head (Ochyra <i>et al.</i> 2008).
<i>Brachythecium austroglareosum</i> (Müll. Hal.) Kindb.	Widely distributed in Antarctic, including the South Shetland archipelago, but in this region it was only reported in the Livingston and Deception Islands (Ochyra <i>et al.</i> 2008). This species is a new occurrence for Snow Island.
<i>Brachythecium austrosalebrosum</i> (Müll. Hal.) Kindb.	Widely distributed in Antarctica, including in the South Shetland archipelago (Ochyra <i>et al.</i> 2008). This species is a new occurrence for Snow Island.
<i>Bryum argenteum</i> var. <i>argenteum</i> Hedw.	Widely distributed in Antarctica, but rare in South Shetland Island occurring only in King George and Deception Islands (Ochyra <i>et</i>

	<i>al.</i> 2008) and now reported for Snow Island.
<i>Bryum orbiculatifolium</i> Cardot & Broth.	Has primarily distribution on some volcanic islands in South Sandwich and South Shetland Islands (Ochyra <i>et al.</i> 2008). Considering the South Shetland Archipelago, this species occur in Deception Island (Ochyra <i>et al.</i> 2008) and King George Island (Câmara <i>et al.</i> 2017) and it was reported in Snow Island in the present study.
<i>Bryum pseudotriquetrum</i> (Hedw.) G. Gaertn., B. Mey. & Scherb.	Is one of the most common species and widely distributed in Antarctica. It is present in South Shetland Archipelago, reported for King George, Livingston, Deception, Nelson and Greenwich Islands (Ochyra <i>et al.</i> 2008) and now it was found in Snow Island.
<i>Warnstorffia fontinaliopsis</i> (Müll. Hal.) Ochyra	Is widespread and common in the South Shetland Archipelago where it is reported for the King George, Livingston, Nelson and Robert Islands (Ochyra <i>et al.</i> 2008) but it is not reported for Snow Island until this moment.
<i>Warnstorffia sarmentosa</i> (Wahlenb.) Hedenäs	Is widely distributed and locally common only in the South Orkney and South Shetland Islands. In the South Shetland Archipelago, this species is reported in several islands (Elephant, King George, Robert and Livingston Islands) but it was not reported for Snow Island (Ochyra <i>et al.</i> 2008).
<i>Distichium capillaceum</i> (Hedw.) Bruch & Schimp.	Is present in several localities of Antarctica. In the South Shetland Islands it is reported to the King George, Livingston, Deception and Robert Islands (Ochyra <i>et al.</i> 2008) and now also being reported for Snow Island.
<i>Ceratodon purpureus</i> (Hedw.)	Is widely reported for the South

Brid.	Shetland Islands (Ochyra <i>et al.</i> 2008), however, it had not been cited for Snow Island.
<i>Schistidium lewis-smithii</i> Ochyra	Is endemic from Antarctica and reported only for South Shetlands Archipelago in King George and Livingston Islands (Ochyra <i>et al.</i> 2008; Câmara <i>et al.</i> 2017) and now it is reported to one more island in this archipelago, the Snow Island.
<i>Schistidium praemorsum</i> (Müll. Hal.) Herzog	Is known only in three localities in Antarctica. In the West Antarctica Peninsula it is registered in Danco Coast and Fallières Coast and in South Shetlands Archipelago is reported only for Deception Island (Ochyra <i>et al.</i> 2008) and this study found this species in Snow Island too.
<i>Pohlia cruda</i> (Hedw.) Lindb.	Is widely distributed in the South Shetland Islands and in the other localities of maritime Antarctic (Ochyra <i>et al.</i> 2008) but had been not reported yet for Snow Island.
<i>Polytrichastrum alpinum</i> (Hedw.) G.L. Sm.	Is widely distributed in maritime Antarctica and present in the Antarctic Peninsula. This species had been already reported to Snow Island in President Head Plateau (Ochyra <i>et al.</i> 2008).
<i>Polytrichum juniperinum</i> Willd. ex Hedw.	According to Ochyra <i>et al.</i> (2008) this species of more limited occurrence and least common of all species of <i>Polytrichum</i> that occur in Antarctica. This species is locally frequent in tree islands of northern maritime Antarctica (Candlemas and Bellingshausen Islands (South Sandwich Archipelago) and King George Island (South Shetlands Archipelago)). It is scarce elsewhere of South Sandwich Archipelago (Ochyra <i>et al.</i> 2008) and now it is reported for Snow

	Island too.
<i>Polytrichum piliferum</i> Hedw.	Is widely distributed in Antarctica, especially in the South Shetland Archipelago and it is reported to Snow Island in President Head Plateau (Ochyra <i>et al.</i> 2008).
<i>Bryoerythrophyllum recurvirostrum</i> (Hedw.) P.C. Chen	Is widely distributed, but scattered in Antarctica. In South Shetland Archipelago was reported only for Livingston Island (Ochyra <i>et al.</i> 2008), but now it was found in Snow Island.
<i>Syntrichia filaris</i> (Müll. Hal.) R.H. Zander	Is widely distributed in Antarctica and it is most common in South Orkneys and South Shetland Islands, but until this moment this species is not had been reported for Snow Island (Ochyra <i>et al.</i> 2008).
<i>Syntrichia magellanica</i> (Mont.) R.H. Zander	Is present in several regions of Antarctica. In the maritime Antarctica, this species is widely distributed and one of the most frequent species of moss in the South Shetlands Archipelago, but it is not known for Snow Island (Ochyra <i>et al.</i> 2008).
<i>Syntrichia saxicola</i> (Cardot) R.H. Zander	Is widely distributed in South Shetland Island and had not been yet reported for Snow Island (Ochyra <i>et al.</i> 2008).
<i>Hymenoloma grimmiaeum</i> (Müll. Hal.) Ochyra	Is well distributed in several localities of Antarctica including the South Shetland Islands and until this moment it was not reported for Snow Island (Ochyra <i>et al.</i> 2008).

The following is an identification key based on the updated list of species occurring on Snow Island. Included in the key are 19 newly recorded species found by the present study and the 10 other species already mentioned by Ochyra *et al.* (2008).

### **Key to the mosses of Snow Island**

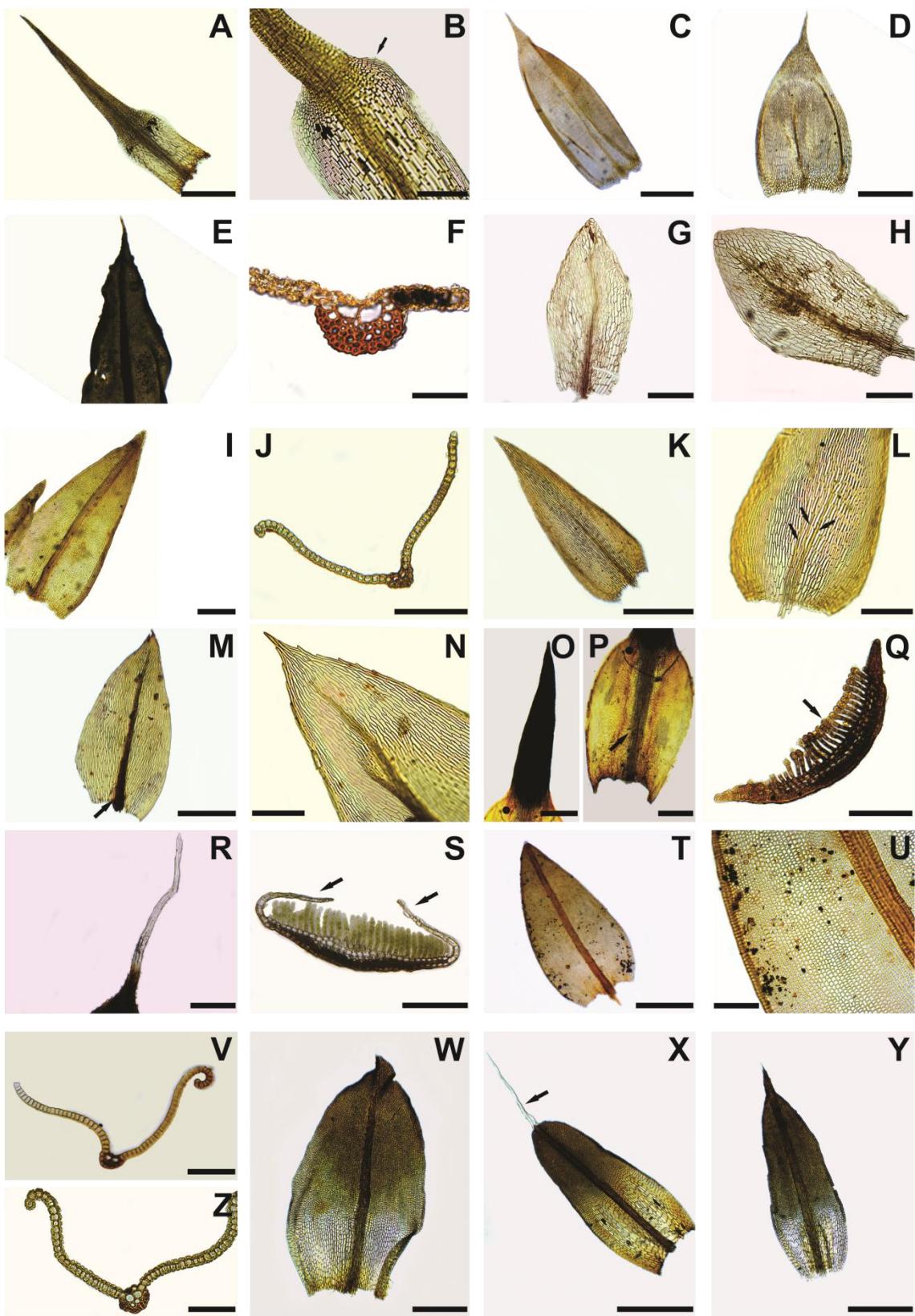
1. Plants pleurocarpous, stems prostrate, forming wefts or mats..... 2
1. Plants acrocarpous, stems erect, forming tufts or cushions..... 8
2. Costa extending into apex, leaves strongly falcate-secund..... 3
2. Costa ending below apex, leaves not falcate-secund..... 4
3. Leaves with alar cells forming an isodiametric group..... *Sanionia georgicouncinata*
3. Leaves with alar cells forming a triangular group..... *Sanionia uncinata*
4. Costa frequently short and branched near the leaf base  
..... *Drepanocladus polygamus* (Plate 1K-L)
4. Costa extending for  $\frac{1}{2}$  to  $\frac{3}{4}$  of leaf length, not branched..... 5
5. Leaves plane, non plicate..... 6
5. Leaves strongly concave, plicate..... 7
6. Alar region undifferentiated..... *Warnstorffia fontinaliopsis*

6. Alar region differentiated.....*Warnstorffia sarmentosa*
7. Leaf margins irregularly; apex long  
acuminate.....*Brachythecium austroglareosum* (Plate 1C)
7. Leaf margins plane; apex short,  
acuminate.....*Brachythecium austrosalebrosum* (Plate 1D)
8. Leaf costa absent.....9
8. Leaf costa present .....10
9. Leaves lanceolate to ovate-lanceolate; leaf margins entire.....*Andreaea regularis*
9. Leaves panduriform to ovate-spatulate or oblong, leaf margins crenate or dentate at the base.....*Andreaea gainii*
10. Leaves subulate.....11
10. Leaves not subulate .....16
11. Subula never tubulose, shoulder well marked.....*Bartramia patens* (Plate 1A-B)
11. Subula subtubulose, shoulder not marked.....12
12. Leaves distichous.....*Distichium capillaceum*
12. Leaves never distichous .....13
13. Large central strand in transverse cross section of the stem, leaves abruptly tapering to a subula .....*Ditrichum hyalinum*

13. Small central strand in transverse cross section of the stem, leaves gradually tapering to a subula ..... 14
14. Costa broad, occupying 1/3 or more of the leaf base, excurrent ..... *Chorisodontium aciphyllum*
14. Costa narrower, occupying ¼ or less of the leaf base, subpercurrent or percurrent..... 15
15. Costa percurrent; cross section of costa showing a strong dorsal stereid band and a weaker (or absent) ventral one; ventral epidermal cells long, rectangular to linear in the subula..... *Hymenoloma antarcticum*
15. Costa subpercurrent; cross section of costa showing weak stereid bands on both surfaces; ventral epidermal cells short, rectangular to rounded in the subula ..... *Hymenoloma grimmiaeum*
16. Cell walls thickened or ornamented with papillae or mamillae..... 17
16. Cell walls never thickened or ornamented with papillae or mamillae ..... 20
17. Leaves without C-shaped papillae, lanceolate to long lanceolate; costa with two stereid bands in cross section..... *Bryoerythrophyllum recurvirostrum* (Plate 1E-F)
17. Leaves with C-shaped papillae, leaves ovate, oblong-lanceolate; costa with one or more stereid bands in cross section ..... 18
18. Leaves ovate-elliptic to ovate-lingulate; costa excurrent, ending in a hyaline hair-point..... *Syntrichia magellanica* (Plate 1X,Z)

18. Leaves oblong-lanceolate to lingulate-spathulate; costa percurrent, rarely short-excurrent, hyaline hair-point absent ..... 19
19. Leaves short acuminate; leaf margins dentate or serrate, crenate at the apex ..... *Syntrichia filaris* (Plate 1W)
19. Leaves long acuminate; leaf margins entire throughout ..... *Syntrichia saxicola* (Plate 1Z)
20. Ventral lamellae present in leaf cross section ..... 21
20. Ventral lamellae absent in leaf cross section ..... 23
21. Leaf margins entire; apex hyaline and piliferous ..... 22
21. Leaf margins serrate; apex never piliferous ..... *Polytrichastrum alpinum* (Plate 1O-Q)
22. Leaves crowded in upper part of the stem; leaves ending in a short hair-point ..... *Polytrichum juniperinum*
22. Leaves not crowded, distributed throughout the stem; leaves ending in a long, piliferous hair-point ..... *Polytrichum piliferum* (Plate 1R-S)
23. Apical cells hexagonal, rhomboid, linear or somewhat vermicular ..... 24
23. Apical cells rounded, elliptic, oblate to short rectangular ..... 27
24. Leaf border present; laminal cells broad and short, rhomboidal to hexagonal ..... 25

24. Leaf border always absent, laminal cells oblong, linear-hexagonal or linear or somewhat vermicular.....*Pohlia cruda* (Plate 1M-N)
25. Leaves reddish and decurrent at the base, apex acuminate, acute to broadly acute, often cuspidate.....*Bryum pseudotriquetrum* (Plate 1G-H)
25. Leaves greenish to pale brown, never decurrent at the base, apex rounded and obtuse to apiculate.....26
26. Plants whitish due to colorless cells in the apex; apex acute.....*Bryum argenteum*
26. Plants greenish, never whitish; apex obtuse.....*Bryum orbiculatifolium*
27. Margins plane to incurved near the apex; costa homogeneous in cross section.....28
27. Margins recurved to revolute near the apex; costa heterogeneous in cross section.....*Ceratodon purpureus* (Plate 1I-J)
28. Leaves U-shaped in cross section.....*Schistidium lewis-smithii*
28. Leaves keeled to V-shaped in cross section.....*Schistidium praemorsum* (Plate 1T-V)



**Plate 1.** Morphology of some of the mosses occurring in Snow Island. **A-B** *Bartramia patens*: **A.** Leaf, general aspect; **B.** Detail of the shoulder region (arrow). **C.** *Brachythecium austroglareosum*: leaf general aspect; **D.** *Brachythecium austrosalebrosum*: Leaf, general aspect. **E-F** *Bryoerythrophyllum recurvirostre*: **E.** General aspect of leaf tip and mid region. **G-H:** *Bryum pseudotrichetrum*: **G.** Branch leaf, general aspect; **H.** Branch leaf showing variation in shape. **I-J:** *Ceratodon purpureus*: **I.** Leaf, general aspect; **J.** Transversal section of the leaf mid region. **K-L:** *Drepanocladus polygamus*: **K.** Leaf, general aspect; **J.** Detail of the base showing the branched costa (arrows). **M-N** *Pohlia cruda*: **M.** Leaf, general aspect; **N.** Detail of the sturdy cells and the reddish costa near the base (arrow). **O-Q:** *Polytrichastrum alpinum*: **O.** Detail of the apex showing the thick costa covering whole region; **P.** Detail of the leaf base showing slender costa and the limb (arrow); **Q.** Cross section of the leaf showing the lammelae in the costa (arrow). **R-S** *Polytrichum piliferum*: **R.** Detail of the apex bearing long and hyaline awn; **S.** Transversal section of the leaf showing the lamellae partially covered by the limb (arrows). **T-V** *Schistidium praeomorsum*: **T.** Leaf, general aspect; **U.** Detail of the mid region showing cells with thickened walls; **V.** Transversal “V-shaped” section of the leaf. **W.** *Syntrichia filaris*: Leaf, general aspect. **X, Z:** *Syntrichia magellanica*: **X.** Leaf, general aspect showing the long and distinctive spurred hyaline awn (arrow); **Z.** Leaf cross section. **Y.** *Syntrichia saxicola*: leaf, general aspect. Scales - A, C, D, E, K, M, O, P, T, W, X, Y: 500 µm; B, G, H, I, L, N, Q, R, S, U: 200 µm; V, Z: 100 µm; F, J, 50 µm.

## Final Considerations

Mosses are the most abundant plants in Antarctica (Ochyra *et al.* 2008), unlike other places in the world. Snow Island has high species richness of mosses, representing approximately 26% of the known diversity of Antarctica, of which about 65% are new occurrences for the Island found by the present study.

Snow Island has important species of mosses, such as *Schistidium lewis-smithii* Ochyra, an endemic species of Antarctica, which is now registered for Snow Island. *Schistidium praeomorsum* (Müll. Hal.) Herzog had been cited for only three localities in Antarctica, but was found on Snow Island by the present study.

The present study increased the knowledge of moss diversity on Snow Island by 190%. This indicates the need and importance for further studies to obtain further knowledge and aid the preservation of local biodiversity.

We believe that the considerable increase in the number of species documented for Snow Island can be explained by climate change, in particular warming in the region. As discussed by Turner *et al.* (2009), the South Shetland Archipelago is among the regions of Antarctica most affected by the climate. This has been evidenced by other studies, such as Sancho *et al.* (2017), who revealed changes in the lichen flora, Brabyn *et al.* (2006). This demonstrates changes in the distribution of moss and algae in Antarctica, in addition to Câmara *et al.* (2017), Henriques *et al.* (2018) and Câmara *et al.* (2020) who showed substantial changes in local floras of the South Shetland Archipelago.

We emphasize the importance of monitoring the local moss flora of Snow Island, due to the establishment of new and rare species. We believe that the creation of a protected area within President Head would be interesting considering the moss flora and the paleobotanical specimens that have been uncovered there.

## Acknowledgments

The authors are grateful to the Brazilian Antarctic Program (PROANTAR), the Ministry of Science Technology Innovation and Communication (MCTIC), the Brazilian Navy and the Brazilian Air Force for providing important logistic support. The authors are also grateful to Bárbara Guedes Costa Silva for helping with the maps.

The authors thank PROANTAR and CNPq (National Council for Scientific and Technological Development - Finance Code 64/2013) for providing funds. The first and second authors thank CAPES (Coordenação de Aperfeiçoamento de Pessoal de Nível Superior) for partial funding of the study (CAPES Finance Code 001).

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## **Appendix I. Voucher information of specimens collected in Snow Island in this study.**

*Andreaea regularis* Müll. Hal. – *Henriques D.K.* 2129; *Valente D.V.* 1691. *Bartramia patens* Brid. – *Henriques D.K.* 2131; *Valente, D.V.* 1651. *Brachythecium austroglareosum* (Müll. Hal.) Paris – *Carvalho-Silva, M.* 2820; *Valente, D.V.* 1654, 1595. *Brachythecium austrosalebrosum* (Müll. Hal.) Kindb. – *Valente D.V.* 1593, 1603. *Bryoerythrophyllum recurvirostrum* (Hedw.) P.C. Chen – *Bordin J.* 2540. *Bryum argenteum* var. *argenteum* Hedw. – *Valente D.V.* 1601, 1612. *Bryum orbiculatifolium* Cardot & Broth. – *Aquino P.P.U.* 183; *Valente D.V.* 1681. *Bryum pseudotriquetrum* (Hedw.) G. Gaertn., B. Mey. & Scherb. – *Aquino P.P.U.* 178, 183, 191, 193, 194; *Bordin J.* 2544, 2545, 2546; *Carvalho-Silva, M.* 2762, 2766, 2767, 2773, 2777, 2783, 2785, 2786, 2787, 2788, 2791, 2795, 2798, 2800, 2801, 2804, 2807, 2808, 2809, 2811, 2815; *Valente, D.V.* 1597, 1602, 1606, 1612, 1620, 1622, 1627, 1645, 1648, 1661, 1663, 1664, 1669, 1670, 1677, 1679, 1680, 1681, 1685, 1688. *Ceratodon purpureus* (Hedw.) Brid. – *Aquino P.P.U.* 186; *Bordin J.* 2541, 2542, 2543; *Carvalho-Silva, M.* 2763, 2765, 2769, 2778, 2794, 2796, 2802, 2805, 2809, 2812, 2813, 2814, 2816, 2817, 2818; *Valente D.V.* 1629. *Distichium capillaceum* (Hedw.) Bruch & Schimp. – *Aquino P.P.U.* 192; *Valente D.V.* 1652, 1654, 1669, 1690. *Drepanocladus polygamus* (Schimp.) Hedenäs – *Valente D.V.* 1641. *Hymenoloma grimmiaeum* (Müll. Hal.) Ochyra – *Valente D.V.* 1604, 1659, 1692. *Pohlia cruda* (Hedw.) Lindb. – *Valente D.V.* 1614. *Polytrichastrum alpinum* (Hedw.) G.L. Sm. – *Aquino P.P.U.* 190; *Carvalho-Silva M.* 2768, 2810, *Valente D.V.* 1596, 1607, 1620, 1623, 1626, 1633, 1637, 1638, 1647, 1655, 1666, 1672, 1676. *Polytrichum juniperinum* Willd. ex Hedw. – *Carvalho-Silva M.* 2806. *Polytrichum piliferum* Hedw. – *Henriques D.K.* 2133, 3627; *Carvalho-Silva M.* 2763, 2774, 2819; *Valente D.V.* 1671. *Sanionia uncinata* (Hedw.) Loeske – *Aquino P.P.U.* 189; *Bordin J.* 2539; *Carvalho-Silva M.* 2765, 2779, 2787, 2788, 2789, 2798, 2801, 2803, 2805, 2814, 2821, 2822; *Valente D.V.* 1598, 1601, 1602, 1603, 1605, 1607, 1609, 1610, 1611, 1613, 1614, 1617, 1618, 1619, 1621, 1622, 1624, 1628, 1630, 1632, 1634, 1635, 1637, 1639, 1640, 1641, 1642, 1643, 11645, 1646, 1649, 1651, 1656, 1660, 1661, 1663, 1664, 1665, 1667, 1668, 1669, 1674, 1675, 1676, 1677, 1678, 1683, 1687. *Schistidium lewis-smithii* Ochyra – *Aquino P.P.U.* 181; *Carvalho-Silva M.* 2764; *Valente D.V.* 1693. *Schistidium praemorsum* (Müll. Hal.) Herzog – *Aquino, P.P.U.* 180. *Syntrichia filaris* (Müll. Hal.)

R.H. Zander – *Aquino P.P.U* 187; *Valente D.V.* 1606, 1617, 1643, 1663, 1684. *Syntrichia magellanica* (Mont.) R.H. Zander – *Aquino P.P.U* 177; *Bordin J.* 2547, 2548; *Carvalho-Silva M.* 2770, 2771, 2772, 2773 2797; *Henriques D.K.* 2130, 2131, 2134; *Valente D.V.* 1594, 1598, 1599, 1601, 1610, 1611, 1621, 1624, 1625, 1628, 1644, 1646, 1650, 1662, 1669, 1689. *Syntrichia saxicola* (Cardot) R.H. Zander – *Aquino P.P.U* 187, 188; *Carvalho-Silva M.* 2775, 2776 2780; *Henriques D.K.* 2132; *Valente D.V.* 1651, 1653, 1657, 1658, 1684, 1686, 1687. *Warnstorffia fontinaliopsis* (Müll. Hal.) Ochyra – *Carvalho-Silva M.* 2784, 2789, 2790, 2793, 2821; *Valente D.V.* 1605, 1610, 1615, 1618, 1627, 1631, 1636. *Warnstorffia sarmentosa* (Wahlenb.) Hedenäs – *Carvalho-Silva M.* 2787, 2789.

## **Considerações Finais da Tese**

Foram geradas 21 novas sequências (ITS2) (mais 25 platidiais) para as espécies de *Syntrichia* Brid. ocorrentes na Antartica. Os estudos filogenéticos e a morfologia corroboram para a circunscrição das espécies - *Syntrichia filaris*, *S. magellanica*, *S. saxicola* e *S. sarconeurum*, bem como evidenciam que o *Syntrichia caninervis* var. *caninervis* não ocorre na localidade, tratando-se de um equívoco taxonômico, evidenciado nas análises moleculares, que mostram a distância entre estas.

As análises filogenéticas denotam a existência de uma espécie anteriormente não relatada para a Antártica, de distribuição bipolar – *Syntrichia sp1*. Com a pendência da revisão do material *typus* para confirmação do nome e tratamento taxonômico.

*Syntrichia sarconeurum*, por sua vez, que seria produto de distintas colonizações (Skotnicki *et al.* 1999; 2000; 2004), na realidade apresenta-se como espécie bem definida clado II (Fig. 2 - capítulo 1), que agrupa as amostras sequenciadas neste estudo e outras oriundas do Genbank (provenientes de diversas localidades), correspondentes morfologicamente ao *typus* da espécie, enquanto o clado I, externo ao grupo de *Syntrichia* (vide Figura 2), agrupa amostras de *Bryoerythrophyllum recurvirostrum*, o que indica erro de identificação ou no processo de extração de DNA, provavelmente.

Além disso, o presente estudo proporcionou dados concernentes à distribuição das espécies, trazendo atualizações a Flora da Ilha Snow, com incremento no quantitativo de espécies conhecidas, que anteriormente estava restrita a 10 espécies (Ochyra *et al.* 2008). A partir de novas coletas foram identificadas 19 novas espécies ocorrentes na região, dentre elas exemplares de *Syntrichia* utilizadas nos estudos moleculares do capítulo 1, além de *taxa*, raros, a exemplo de - *Schistidium praemorsum* (Grimmiaceae), anteriormente citado para apenas três localidades da Antártica (Ochyra *et al.* 2008); e *Schistidium lewis-smithii*, endêmica da Antártica, e, antes deste estudo era reportada para para duas ilhas do arquipélago das Shetlands do Sul.

## **Apoio e Financiamento**

Este estudo é parte do projeto Evolução e Dispersão de Espécies Antárticas Bipolares de Briófitas e Líquens, aprovado pelo edital 64/2013 Chamada MCTI/CNPq/FNDCT contado com o apoio financeiro do mesmo.

A Fundação de Apoio e Pesquisa do Distrito Federal – FAPDF auxiliou na realização de visita técnica e na participação no XIX Congresso Internacional de Botânica na cidade de Quito Ecuador.

O presente trabalho foi realizado com apoio da Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) - Código de Financiamento 001.