

LILIANA ESSI

**ANÁLISE FILOGENÉTICA E TAXONÔMICA DO
COMPLEXO *BRIZA*
(POACEAE: POEAE)**

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**ANÁLISE FILOGENÉTICA E TAXONÔMICA DO
COMPLEXO *BRIZA*
(POACEAE: POEAE)**

LILIANA ESSI

Tese apresentada ao Programa de Pós-Graduação em Botânica da Universidade Federal do Rio Grande do Sul, como parte dos requisitos para a obtenção do título de Doutor em Botânica.

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EXCETO À LUZ DA EVOLUÇÃO

Theodosius Dobzhansky

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“Perdoem a cara amarrada, perdoem a falta de abraço

Perdoem a falta de espaço, os dias eram assim

Perdoem por tantos perigos, perdoem a falta de abrigo

Perdoem a falta de amigos, os dias eram assim

Perdoem a falta de folhas, perdoem a falta de ar

Perdoem a falta de escolha, os dias eram assim

E quando passarem a limpo, e quando cortarem os laços

E quando soltarem os cintos, façam a festa por mim

Quando lavarem a mágoa, quando lavarem a alma

Quando lavarem a água, lavem os olhos por mim

Quando brotarem as flores, quando crescerem as matas

Quando colherem os frutos digam o gosto pra mim”

Ivan Lins

*À minha mãe e minhas irmãs, com todo
amor, dedico esta tese.*

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INTRODUÇÃO

O gênero *Briza* (Poaceae – Poeae) foi descrito por Linnaeus (1753) com três espécies européias, *B. maxima* (Figura 1), *B. media* (Figura 2) e *B. minor*. A essas, Marschall von Bieberstein (1808) acrescentou *Briza humilis* (Figura 3), descrita para a Ucrânia, ficando, então, quatro espécies euro-asiáticas aceitas para o referido gênero. Posteriormente a Linnaeus (1753), espécies novas sul-americanas passaram a ser descritas por diferentes autores, em *Briza* ou em novos gêneros considerados relacionados (para fotos de diferentes espécies sul-americanas, vide Figuras 4 a 8). Os gêneros *Calotheca* Desv. (1810), *Chascolytrum* Desv. (1810), *Poidium* Nees (1836), *Rhombolytrum* Link (1833), *Gymnachne* Parodi (1938), *Lombardochloa* Roseng. & Arrill. (1979) e *Microbriza* Parodi ex Nicora & Rúgolo (1981) foram descritos ao longo do tempo. Entretanto, autores posteriores passaram a aceitar *Briza* e os demais gêneros acima citados com diferentes circunscrições, ora aceitando *Briza lato sensu*, com os demais gêneros como seções ou subgêneros (Parodi, 1920; Longhi-Wagner, 1987), ora aceitando *Briza* com uma circunscrição mais restrita, englobando as espécies euro-asiáticas (Matthei, 1975), ou separando parte das espécies sul-americanas em outros gêneros, mas mantendo algumas em *Briza* (Nicora & Rúgolo de Agrasar, 1981). As espécies sul-americanas até então descritas passaram a ser incluídas em um número variado de gêneros, aceitando todos ou apenas parte dos gêneros mencionados acima. Devido a tais controvérsias quanto à circunscrição genérica, o grupo passou a ser denominado por alguns autores de “Complexo *Briza*” (Matthei, 1975; Nicora & Rúgolo de Agrasar, 1981; Longhi-Wagner, 1987; Bayón, 1998).

Nas décadas de 1970/1980, um grupo de pesquisadores dos Departamentos de Genética e Botânica da Universidade Federal do Rio Grande do Sul, coordenado pelas Dras. Helga Winge e Hilda M. Longhi-Wagner, realizou uma série de trabalhos envolvendo o complexo, incluindo análises fenéticas, citotaxonômicas, quimiotaxonômicas (análise de flavonóides e isoesterases) e de parâmetros ontogenéticos, que resultaram em dissertações, uma tese e diversos artigos (Hickenbick, 1983; Leyser & Winge, 1979; Schifino-Sampaio, 1979; Schifino & Winge, 1979; Torres de Lemos, 1983). Embora nem todos os resultados obtidos pelo grupo tenham sido publicados, Longhi-Wagner (1987), ao tratar a tribo Poeae para a flora do Rio Grande do Sul, considerou o gênero *Briza lato sensu* dividido em seções [*Briza* (L.) Longhi-Wagner, *Calotheca* (Desv.) Benth. & Hook., *Chascolytrum* (Desv.) Benth. & Hook., *Poidium* (Nees) Ekman e *Lombardochloa* (Roseng. & Arrill.) Longhi-Wagner] certamente baseada nos resultados obtidos pelo grupo.

Matthei (1975) analisou o que denominou “*Briza* Komplex” e aceitou a seguinte circunscrição: *Briza stricto sensu* (apenas espécies euro-asiáticas), *Calotheca* (gênero monoespecífico), *Chascolytrum* (com seis espécies) e *Poidium* (com nove espécies).

Bayón (1998) publicou uma análise cladística do complexo, utilizando apenas dados exomorfológicos, com o objetivo de resolver a sua circunscrição. Bayón (1998) concluiu que a circunscrição mais adequada era aquela proposta por Nicora & Rúgolo de Agrasar (1981), porém adicionando *Briza ambigua* Hack. ao gênero *Poidium*, espécie que havia sido excluída do complexo pelas autoras.

Figura 1: *Briza maxima*. A. Hábito. B. Detalhe das espiguetas. C. Detalhe da lígula.



Figura 2: *Briza media*. A – B. Hábito. C. Plântulas, obtidas a partir de cariopses germinadas em laboratório. D. Espigueta.



Figura 3: *Briza humilis*. A. Hábito. B. Detalhe da lígula e de parte da bainha e lâmina foliar. C. Detalhe da espiguetas.

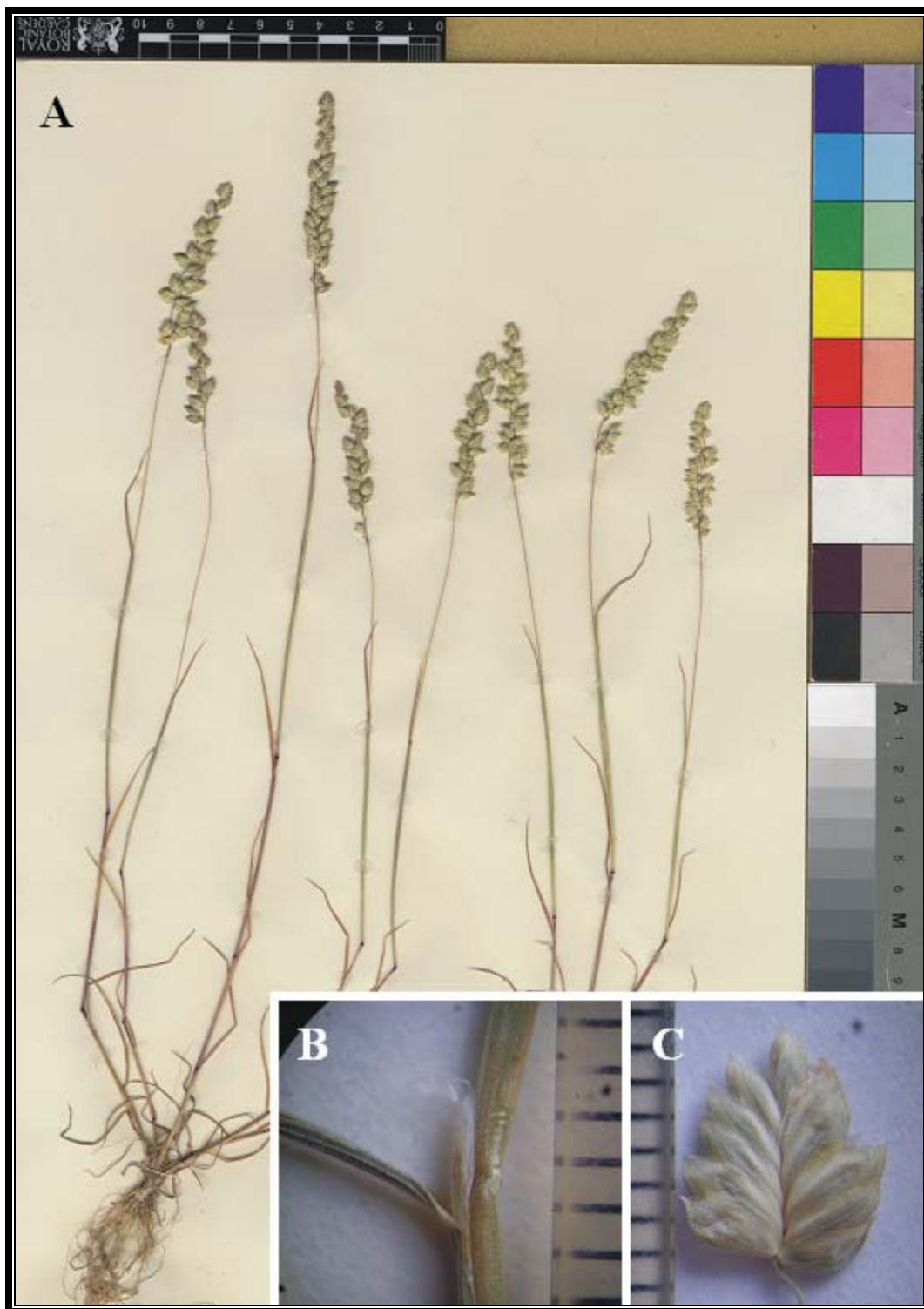


Figura 4: A – C. *Briza bidentata* Roseng., Arrill. & Izag.. A. Hábito. B. Detalhe da panícula. C. Detalhe da espiguetas. D e E. *Briza uniolae* (Nees) Nees ex Steud. D. Panícula. E. Detalhe das espiguetas.

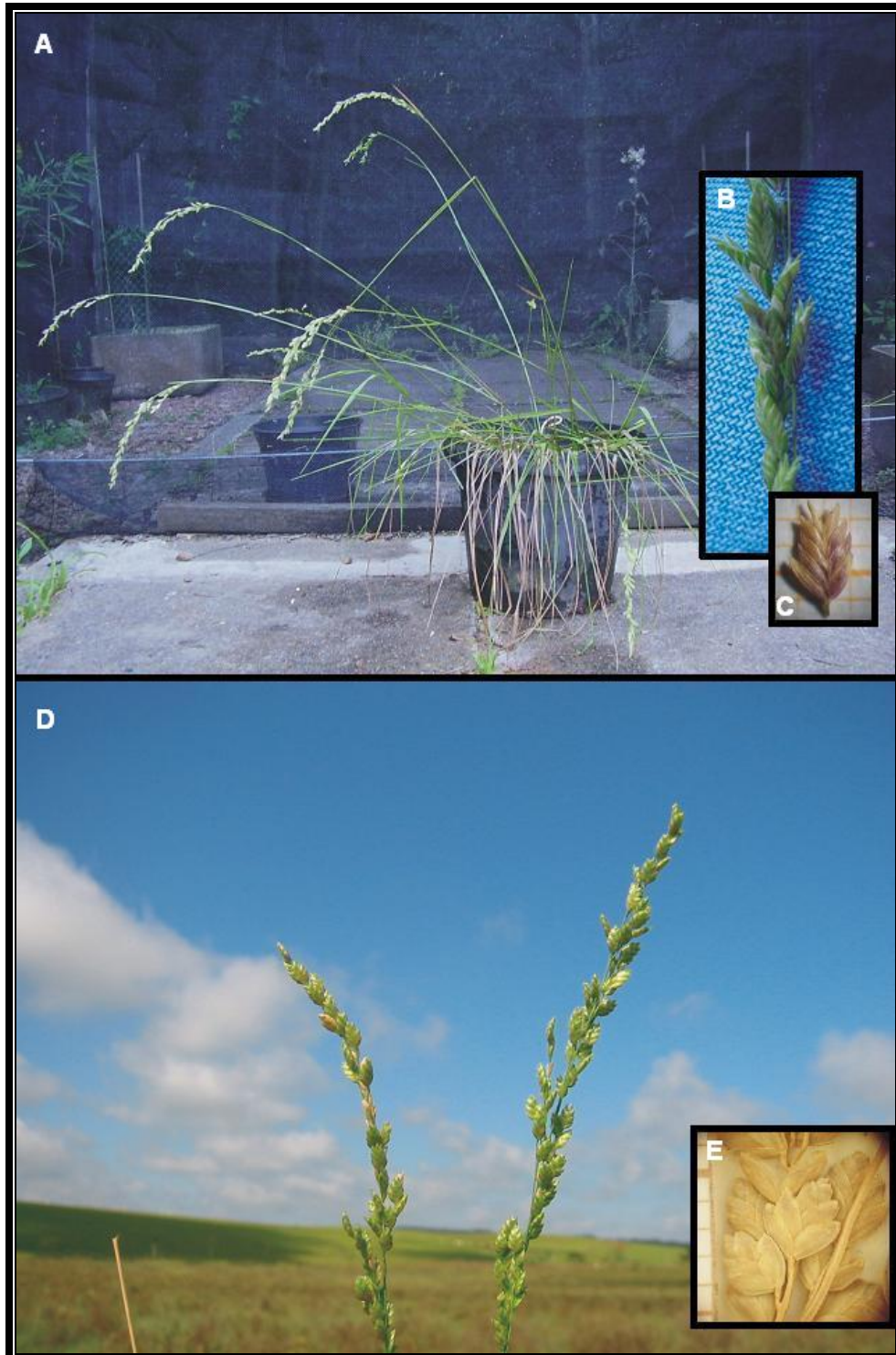


Figura 5: *Briza lamarckiana* Nees. A. Hábito. B. Detalhe das espiguetas. C. Panícula. D. Detalhe da lígula.



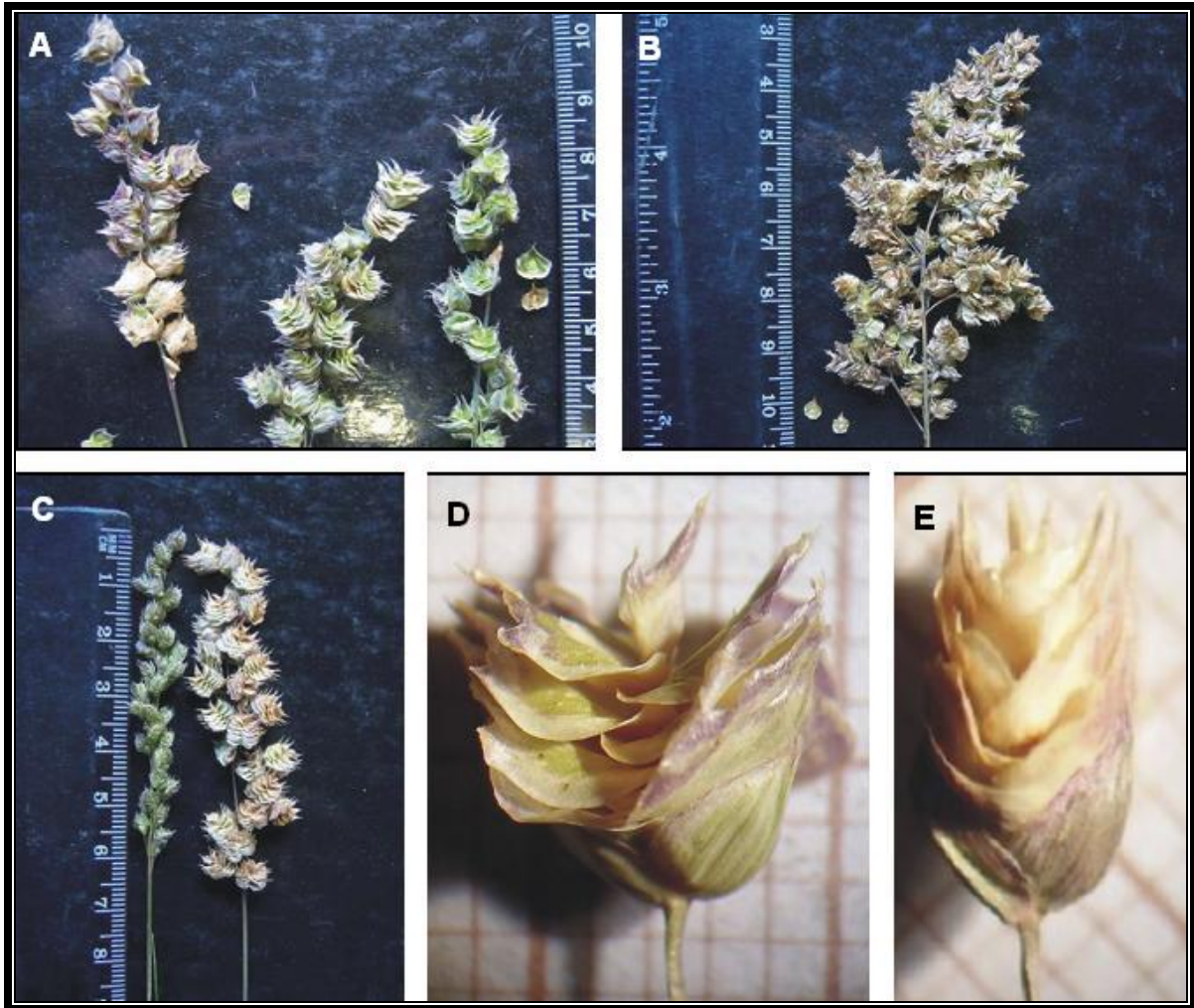
Figura 6: *Briza scabra* (Nees ex Steud.) Ekman. A. Hábito. B. Detalhe da panícula. C. Detalhe da espigueta.



Figura 7: *Briza monandra* (Hack.) Pilg. A. Parte da variação encontrada nas dimensões das espiguetas. B. Parte da variação encontrada no tamanho do antécio I.



Figura 8: *Briza subaristata* Lam. A – C. Parte da variação encontrada para as panículas. D e E. Detalhes de espiguetas com diferentes formas tridimensionais. D. espiguetas globosa. E. espiguetas cilíndrica.



Deste modo, atualmente o *Complexo Briza* abrange cerca de 272 nomes registrados na base TROPICOS (mobot.mobot.org/W3T/Search/vast.html), incluindo sinônimos, nomes ilegítimos supérfluos, categorias infra-genéricas, infra-específicas e *nomina nuda*. Os gêneros envolvidos são *Briza*, *Microbriza*, *Poidium*, *Calotheca*, *Chascolytrum*, *Rhombolytrum*, *Lombardochloa* e *Gymnachne*. Além desses, no presente

trabalho também foi incluído o gênero monoespecífico *Erianthecium* Parodi, devido às suas afinidades morfológicas com o complexo, embora nunca tenha sido tratado como membro do mesmo. Desses, cerca de 115 nomes foram considerados nesta revisão.

Diante da situação taxonômica controvertida do complexo, foi proposto um estudo sobre a evolução do grupo, utilizando dados de seqüências de DNA, para que fosse possível obter uma circunscrição em nível genérico baseada em grupos monofiléticos. Uma filogenia do grupo, baseada em dados morfológicos, já foi disponibilizada por Bayón (1998), como anteriormente referido. Entretanto, tal autor lançou mão de muitos dos mesmos caracteres que os taxonomistas vinham utilizando para produzir as mais diferentes circunscrições.

Todas as decisões taxonômicas são, em última análise, mais ou menos arbitrárias (Crawford & Dorn, 1974), do mesmo modo que a escolha dos caracteres e estados de caráter para uma matriz de dados quaisquer. Porém, análises filogenéticas baseadas em dados moleculares tendem a minimizar de modo substancial a subjetividade das análises, não apenas por permitirem automação em muitas etapas do estudo - através de inúmeros programas, baseados no mais diferentes algoritmos -, mas também por reduzirem o número de decisões complexas sobre a homologia dos caracteres. Um caráter, numa matriz de seqüências de DNA, pode apresentar, basicamente, cinco estados, A, C, T, G e inserção/deleção, sendo a homologia definida por posição no alinhamento, enquanto caracteres exo-morfológicos necessitam não apenas da definição de quantos estados aceitar, como também de quais estruturas podem ou não ser consideradas homólogas. Outra vantagem das seqüências de DNA e de outros marcadores moleculares, diante dos dados morfológicos, é o acesso direto ao genoma, que minimiza a interferência do efeito

ambiental. Salienta-se ainda que seqüências de DNA representam fontes de caracteres universais (úteis para estudos de táxons de qualquer hierarquia, desde populações até reinos), abundantes (nem todos os fragmentos são filogeneticamente informativos, mesmo assim o número de fragmentos potencialmente informativos é enorme), e apresentam propriedades estruturais, funcionais, mutacionais, seletivas, bem como taxas evolutivas diversificadas, enriquecendo a gama de abordagens possíveis.

As abordagens moleculares podem constituir uma rica fonte de informação taxonômica. Marcadores moleculares, como aqueles obtidos através das técnicas de ISSR (Inter Simple Sequence Repeats) e RAPD (Randomly Amplified Polimorphic DNA), são extremamente úteis em estudos intra e interpopulacionais. Já as seqüências de DNA são utilizadas em estudos nos mais diversos níveis hierárquicos, em todos os reinos vivos, sendo a escolha do fragmento adequado para a análise um dos fatores decisivos no sucesso de um estudo.

A escolha do fragmento de DNA depende de três fatores principais: das taxas de evolução do fragmento (se são adequadas ao nível taxonômico do grupo em estudo), do tamanho do fragmento de DNA (se é passível de amplificação e seqüenciamento para todos os táxons em questão e se oferece o número de caracteres variáveis necessários para inferir uma filogenia), e do número de cópias do fragmento por genoma (preferencialmente cópias únicas, mas famílias multigênicas com evolução em concerto ou com cópias ortólogas e parálogas facilmente discerníveis também podem ser consideradas).

Neste trabalho, dados moleculares de dois tipos foram utilizados: marcadores ISSR e seqüências de DNA. Os primeiros foram selecionados por apresentarem-se

altamente variáveis, potencialmente úteis em questões no nível populacional e interespecífico, incluindo complexos de espécies morfológicamente afins, com problemas de circunscrição, ou decisões relativas a categorias infra-específicas. Já as seqüências de DNA foram selecionadas para a análise filogenética do grupo, visando à definição de grupos monofiléticos que pudessem embasar uma circunscrição em nível genérico para o complexo.

Para a análise filogenética, foram selecionados dois fragmentos de origem nuclear (a região dos espaçadores ITS do rDNA - Internal Transcribed Spacers -, e a porção terminal do gene GBSSI – Granule Bound Starch Synthase), e um de origem plastidial (incluindo o espaçador entre os genes para DNA transportador *trnL-trnF* e o intron do gene *trnL*), todos já apontados como filogeneticamente úteis em estudo com Poaceae (Mason-Gamer, 2001; Baumel et al., 2002; Blattner, 2004; Guo & Li, 2004), sendo os ITS e o *trnL-trnL-trnF* os dois fragmentos mais utilizados em estudos filogenéticos nos últimos tempos.

As reconstruções filogenéticas foram realizadas mediante três métodos de análise filogenética distintos: Parcimônia, Máxima Verossimilhança e Análise Bayesiana. O primeiro, baseia-se no critério otimizador da parcimônia, ou seja, no princípio de que a evolução de um grupo dá-se preferencialmente pela via mais econômica possível. Apenas novidades evolutivas compartilhadas (sinapomorfias) são consideradas para a formação dos clados. Os dois últimos são métodos probabilísticos, e selecionam as árvores por suas probabilidades, conforme determinado modelo de evolução molecular (ou de outro tipo de caráter, no caso da Análise Bayesiana). A principal diferença entre os últimos métodos é o modo de implementação. Enquanto a Máxima Verossimilhança utiliza uma

abordagem estatística padrão, a Análise Bayesiana baseia-se nas probabilidades posteriores das árvores, dadas pelo teorema de Bayes. Além disso, na Análise Bayesiana, a busca é feita através do algoritmo de Monte Carlo de Cadeias de Markow (MCMC), que permite a busca também em “cadeias frias”.

A seqüência empregada na solução de problemas pelos métodos probabilísticos em questão poderia ser representada da seguinte maneira:

Máxima verossimilhança

Problema → Dados → Modelo → Análises → Conclusão

Análise Bayesiana

Problema → Dados → Modelo → Distribuição a priori → Análise → Conclusões

Todos os modelos apresentam vantagens, desvantagens e problemas. Um dos principais problemas conhecidos para a Parcimônia é a atração de longos ramos (*long-branch attraction*), que consiste no agrupamento de Unidades Evolutivas pelo simples fato de apresentarem taxas evolutivas elevadas em comum, e não necessariamente por afinidades evolutivas. Esse problema pode ocorrer também em outros métodos de análise filogenética, entretanto, é bem mais comum à Parcimônia. Por outro lado, os métodos probabilísticos podem ser hiper-parametrizados (apesar de algumas buscas com Parcimônia também serem!). A Máxima Verossimilhança apresenta igualmente a desvantagem da demora nas buscas. Já a Análise Bayesiana pode trazer dificuldades quando a estabilidade da MCMC demora a ser alcançada.

Alguns autores preferem escolher um único método de análise de sua preferência para construir árvores filogenéticas, argumentando que os métodos não são comparáveis, ou ao contrário, alegando que são comparáveis e que determinado método é superior.

Outros preferem fazer análises com múltiplos métodos. Acompanhando o segundo grupo, optou-se por explorar os dados com diferentes métodos de análise, justamente por reconhecer que todos os métodos apresentam suas mazelas, e que as metodologias podem não ser comparáveis, mas sim o produto final (as árvores, sua topologia), e grupos em comum obtidos por diferentes métodos, bem como grupos discrepantes entre os métodos, podem ser ricamente discutidos.

Objetivos

Os objetivos deste trabalho foram inferir a filogenia do chamado “Complexo *Briza*”, e, a partir da compreensão da história evolutiva do grupo, propor uma circunscrição para o mesmo no nível genérico, definir a circunscrição de espécies ou categorias infra-específicas de pequenos complexos de espécies do grupo, e contribuir para o conhecimento da flora agrostológica brasileira e da composição dos campos do sul do Brasil.

Os resultados deste trabalho estão sendo apresentados em quatro capítulos independentes, os quais foram formatados de acordo com os periódicos aos quais serão submetidos. São eles:

Capítulo I. Phylogenetic analysis of the “*Briza* Complex” (Poaceae): este capítulo consiste de uma análise filogenética molecular do Complexo *Briza*, utilizando três fragmentos de DNA distintos e três métodos de análise filogenética distintos. Teve como objetivo principal obter árvores filogenéticas que pudessem nortear uma

circunscrição mais natural para o complexo. Está formatado para ser submetido ao periódico *Molecular Phylogenetics and Evolution*.

Capítulo II. Can ISSR markers help solve taxonomic controversies? A first approach for species of the *Briza* Complex in South America (Poaceae): este capítulo consiste de uma análise molecular, utilizando marcadores ISSR, de quatro complexos de espécies pertencentes ao Complexo *Briza*. Teve como objetivo avaliar a circunscrição de espécies afins e de duas variedades, com uma amostragem contemplando as principais variações morfológicas encontradas, bem como avaliar o potencial de tais marcadores na resolução de problemas taxonômicos encontrados no Complexo *Briza*. Está formatado para ser submetido ao periódico *Genetica*.

Capítulo III. Three new taxa of *Chascolytrum* (Poaceae, Pooideae, Poeae) from South America: este capítulo apresenta diagnoses, descrições e ilustrações de duas espécies novas e uma variedade nova, encontradas durante a revisão taxonômica do complexo. Está formatado para ser submetido ao periódico *Novon*.

Capítulo IV. Taxonomic revision of the “*Briza* Complex”: *Briza* L. and *Chascolytrum* Desv. (Poaceae- Poeae): este capítulo apresenta a revisão taxonômica do Complexo *Briza*, com uma nova circunscrição para o grupo de espécies sul-americanas, baseada nos resultados obtidos para as abordagens apresentadas nos Capítulos I e II. São apresentadas novas combinações, listas de sinônimos, chaves, descrições e ilustrações. Está formatado para ser submetido ao periódico *Kew Bulletin*.

Com base nos critérios previstos no Código Internacional de Nomenclatura Botânica (2006), o presente trabalho é considerado uma publicação não efetiva e não válida para fins de prioridade nomenclatural.

Referências Bibliográficas

- Baumel, A., Ainouche, M.L., Bayer, R.J., Ainouche, A.K. & Misset, M.T. 2002. Molecular phylogeny of hybridizing species from genus *Spartina* Schreb. (Poaceae). *Mol. Phyl. Evol.* 22(2): 303-314.
- Bayón, N.D. 1998. Cladistic analysis of the *Briza* Complex (Poaceae, Poaeae). *Cladistics* 14: 287-296.
- Blattner, F.R. 2004. Phylogenetic analysis of *Hordeum* (Poaceae) as inferred by nuclear rDNA ITS sequences. *Mol. Phyl. Evol.* 33: 289-299.
- Crawford, D.J. & Dorn, R.P. 1974. Numerical chemotaxonomy and other aspects of chemosystematics. *Taxon* 23: 331-338.
- Desvaux, A. N. 1810: Extrait d'un Mémoire sur quelques nouveaux genres de la famille des Graminées. – *Nouv. Bull. Sci. Soc. Philom. Paris* 2: 187-190.
- Guo, Z. & Li, D. 2004. Phylogenetics of the *Thamnocalamus* group and its allies (Gramineae: Bambusoideae): inference from the sequences of GBSSI gene and ITS spacer. *Mol. Phyl. Evol.* 30: 1-12.
- Hickenbick, M.C.M. 1983. Relações Fenéticas e Cladísticas entre as Espécies do Complexo *Briza* (Gramineae): Flavonóides. Tese de Doutorado. Curso de Pós-Graduação em Genética. Universidade Federal do Rio Grande do Sul. Porto Alegre.
- Leyser, V.E. & Winge, H. 1979. Análise palinológica de espécies do complexo *Briza* (Gramineae). *Ciência e Cultura* 31: 577.
- Link. 1833. *Hort. Berol.* 2: 296.
- Linnaeus, C. 1753: *Species Plantarum*. Bd. 1. – Stockholm.

- Longhi-Wagner, H.M. 1987. Flora Ilustrada do Rio Grande do Sul, Fasc. 17. Gramineae. Tribo Poeae. B. Inst. Bioc. 41: 1-191.
- Mason-Gamer, R.J. 2001. Origin of North American *Elymus* (Poaceae: Triticeae) allotetraploids based on Granule-Bound Starch Synthase gene sequences. Syst. Bot. 26(4): 757-768.
- Matthei, O. 1975. Der Briza-Komplex in Südamerika: *Briza*, *Calotheca*, *Chascolytrum*, *Poidium* (Gramineae). Willdenowia 8: 7-168.
- Nees, C. G. 1836. In Lindley (ed.): A Natural System of Botany. 2. Aufl. – London.
- Nicora, E.G. & Rúgolo de Agrasar, Z.E. 1981. Los géneros sudamericanos afines a *Briza* L. (Gramineae). Darwiniana 23(1): 279-309.
- Parodi, L.R. 1920. Notas sobre las especies de *Briza* de la Flora Argentina. Rev. Fac. Arg. Agr. y Vet. B. Aires 3: 113-137.
- Parodi, L.R. 1938. Notas Mus. La Plata, Bot. 3: 29.
- Rosengurtt, B. & Arrillaga de Maffei. B. 1979: *Lombardochloa*, nuevo genero de Gramineae. Anales Fac. Quím. 9: 255-288.
- Sampaio, M.T.S., Hickenbick, M.C.M.& Winge, H. 1979. Chromosome numbers and meiotic behavior of South American species of the *Briza* complex (Gramineae). Rev. Bras. Genet. 2(2): 125 - 134.
- Schifino-Sampaio, M. T. 1979. Citotaxonomia do complexo *Briza* (Gramineae): número cromossômico, cariótipo, quantidade de DNA nuclear, comportamento meiótico. Dissertação de Mestrado. Curso de Pós-Graduação em Genética. Universidade Federal do Rio Grande do Sul. Porto Alegre.

- Schifino, M.T. & Winge, H. 1983. Karyotypes and nuclear DNA content of species of the *Briza* complex and some other genera of Poaceae (Gramineae). Rev. Bras. Genet. (Brazil. J. Genetics) 4(2): 245 - 259.
- Torres de Lemos, C. 1983. Relações fenéticas entre as espécies do complexo *Briza* (Gramineae): isoesterases. Dissertação de Mestrado. Curso de Pós-Graduação em Genética, Universidade Federal do Rio Grande do Sul, Porto Alegre.
- Von Bieberstein, M. 1808. Fl. Taur. Caucas. 1: 66.

Capítulo I

Phylogenetic Analysis of the *Briza* Complex (Poaceae)

Abstract

A phylogenetic analysis of the *Briza* Complex was carried out using three DNA regions (ITS, GBSSI and *trnL-trnL-trnF*, a total of 2,980 bp). The searches were performed by three distinct phylogenetic methods, using plastid and nuclear data in separate matrices. All methods produced similar trees per matrix. However there were conflicts when trees resulting from distinct datasets were compared. Phylogenetic relationships found did not completely fit any circumscription previously proposed for the complex. The early divergence of the Eurasiatic species, and the monophyly of the South American group (including *Erianthecium*, *Rhombolytrum* and *Gymnachne*) support *Briza* L. *stricto sensu*, and a single genus for the American group. *Briza lato sensu* is not supported in most trees, and the American genera cannot be split due to unresolved polytomies. Conflict between chloroplast and nuclear data suggests past reticulation events, although lineage sorting or ITS paralogy cannot be ruled out. Polytomies in the American group may indicate rapid species radiation.

Keywords: Poaceae, Gramineae *Briza* Complex, ITS, *trnL-trnL-trnF*, GBSSI, *Waxy*, Phylogenetic Systematics

Introduction

The genus *Briza* (Poaceae) was described by Linnaeus in 1753, with four species, three of them – *B. media*, *B. minor* and *B. maxima* – still recognized. In 1791, Lamarck described two new South American species: *B. erecta* and *B. subaristata*. In the 19th Century, genera *Chascolytrum* Desv., *Calotheca* Desv. and *Poidium* Nees were published. Later, with new species being described, these genera were reorganized to accommodate the new taxa, or new genera, and infra-generic categories were created to place morphologically related species. Several circumscriptions were proposed for the group that was named *Briza* Complex by Matthei (1975). Matthei (1975) divided the species of the *Briza* Complex into four genera - *Briza stricto sensu* (Eurasian group), *Calotheca*, *Chascolytrum* and *Poidium* – and excluded *B. bidentata* Roseng., Arrill. & Izag. from the complex, suggesting its inclusion in the *Rhombolytrum* Link or *Gymnachne* Parodi. Matthei's work can be considered the most complete treatment of the group, not only because of its exhaustive taxonomic revision and the large number of plants examined, but also due to the cytological and anatomical approaches, the latter including studies of leaves and embryo anatomy. In 1979, the monospecific genus *Lombardochloa* Roseng. & Arrill. was described, including only *Briza rufa* (J. Presl) Steud. Nicora and Rúgolo de Agrasar (1981) followed Matthei's decision to split the group into several genera, but proposed a distinct arrangement for the genera, describing genus *Microbriza* Parodi ex Nicora & Rúgolo (see Table 1). In 1987, taking into account many approaches carried out by Winge and cols. (for a summary, see Winge et al. 1984), Longhi-Wagner renewed a *lato sensu* circumscription for the group, considering sub-generic categories previously described (Longhi-Wagner, 1987; Longhi-Wagner et al., 1987). Even Matthei (1975) had admitted

the existence of clear links between the genera, but preferred to split the group instead of accepting infra-generic categories. Bayón (1998) published the first cladistic approach for the complex, based on morphological data. He suggested accepting the circumscription proposed by Nicora and Rúgolo de Agrasar (1981), although including *B. ambigua* Hack. in *Poidium*, a species that was excluded from the complex by the latter authors. It should be emphasized that the cladistic analysis performed by Bayón (1998) included some characters already used in the taxonomy of the Complex by distinct authors producing completely different circumscriptions due to different weights assigned to each character. Besides, the parsimony method applied included weighting, in other words, some characters were in fact directed to produce a stronger impact on the phylogeny.

The group was also extensively studied using other approaches. Remarkable studies on flavonoid variation, cytology, floral biology, breeding systems and hybridization were performed by Murray and Murray & Cols. (Murray, 1974, 1975, 1976a, 1976b, 1978, 1979, 1984, 1986; Murray and Barker, 1988; William and Murray, 1972; Moss and Murray, 1990). Likewise, the series of genetic studies by Winge et al., which included chromosome counts, karyotypes, nuclear DNA content among other approaches (Sampaio, Hickenbick and Winge, 1979; Leyser and Winge 1979; Born and Winge, 1980; Schifino and Winge, 1983a, 1983b).

Although much has been done in the group, there is no agreement about the circumscription of the involved genera.

In order to provide new information that might be useful in taxonomic decisions, and to understand the evolution of this small and, at the same time, morphologically diverse group, a molecular phylogenetic approach was performed, including DNA sequence data.

Table 1: Comparison of the main classifications related to the *Briza* Complex

Authors	Parodi (1920)	Rosengurt et al. (1968)	Matthei (1975)	Nicora and Rógolo de Agrasar (1981)	Longhi-Wagner (1987)	Bayón (1998)
Species	<i>Briza l.s.</i> with 4 sub-genera	<i>Briza l.s.</i>	4 genera	4 genera	<i>Briza l.s.</i> with 5 sections	5 genera
<i>B. brizoides</i>	* <i>Calothea</i>	+	+ <i>Calothea</i>	+ <i>Calothea</i>	+ <i>Calothea</i>	+ <i>Calothea</i>
<i>B. media</i>	+ <i>Eubriza</i>	-	+ <i>Briza</i>	+ <i>Briza</i>	-	+ <i>Briza</i>
<i>B. maxima</i>	+ <i>Eubriza</i>	+	+ <i>Briza</i>	+ <i>Briza</i>	+ <i>Briza</i>	+ <i>Briza</i>
<i>B. minor</i>	+ <i>Eubriza</i>	+	+ <i>Briza</i>	+ <i>Briza</i>	+ <i>Briza</i>	+ <i>Briza</i>
<i>B. humilis</i>	-	-	+ <i>Briza</i>	-	-	-
<i>B. erecta</i>	+ <i>Chascolytrum</i>	+	+ <i>Chascolytrum</i>	+ <i>Briza</i>	+ <i>Chascolytrum</i>	+ <i>Briza</i>
<i>B. macrostachya</i>	* <i>Chascolytrum</i>	+	* <i>Chascolytrum</i>	+ <i>Briza</i>	+ <i>Chascolytrum</i>	+ <i>Briza</i>
<i>B. subaristata</i>	+ <i>Chascolytrum</i>	+	+ <i>Chascolytrum</i>	+ <i>Briza</i>	+ <i>Chascolytrum</i>	+ <i>Briza</i>
<i>B. paleapilifera</i>	+ <i>Chascolytrum</i>	-	+ <i>Chascolytrum</i>	+ <i>Briza</i>	+ <i>Chascolytrum</i>	+ <i>Briza</i>
<i>B. scabra</i>	-	+	+ <i>Chascolytrum</i>	+ <i>Briza</i>	+ <i>Chascolytrum</i>	+ <i>Briza</i>
<i>B. lamarekiana</i>	-	*	+ <i>Chascolytrum</i>	+ <i>Briza</i>	+ <i>Chascolytrum</i>	+ <i>Briza</i>
<i>B. parodiana</i>	-	+	+ <i>Chascolytrum</i>	+ <i>Briza</i>	+ <i>Chascolytrum</i>	+ <i>Briza</i>
<i>B. uniolae</i>	+ <i>Chascolytrum</i>	+	+ <i>Poidium</i>	+ <i>Briza</i>	+ <i>Poidium</i>	+ <i>Briza</i>
<i>B. calothea</i>	+ <i>Chascolytrum</i>	+	+ <i>Poidium</i>	+ <i>Briza</i>	+ <i>Poidium</i>	+ <i>Briza</i>
<i>B. rufa</i>	-	+	+ <i>Poidium</i>	+ <i>Briza</i>	+ <i>Lombardochloa</i>	+ <i>Briza</i>
<i>B. monandra</i>	* <i>Poidium</i>	+	+ <i>Poidium</i>	+ <i>Briza</i>	+ <i>Poidium</i>	+ <i>Briza</i>
<i>B. juergensii</i>	-	-	+ <i>Poidium</i>	+ <i>Briza</i>	+ <i>Poidium</i>	+ <i>Briza</i>
<i>B. brachychaete</i>	-	-	+ <i>Poidium</i>	+ <i>Microbriza</i>	+ <i>Poidium</i>	+ <i>Microbriza</i>
<i>B. poaemorpha</i>	* <i>Poidium</i>	+	+ <i>Poidium</i>	+ <i>Microbriza</i>	+ <i>Poidium</i>	+ <i>Microbriza</i>
<i>B. ambigua</i>	-	-	+ <i>Poidium</i>	! Excluded	+ <i>Poidium</i>	+ <i>Poidium</i>
<i>B. itatiaiae</i>	-	-	* <i>Poidium</i>	+ <i>Poidium</i>	+ <i>Poidium</i>	+ <i>Poidium</i>
<i>B. brasiliensis</i>	-	-	! Excluded	+ <i>Poidium</i>	+ <i>Poidium</i>	+ <i>Poidium</i>
<i>B. bidentata</i>	-	+	! Excluded	! Excluded	+ <i>Poidium</i>	+ <i>Rhombolytrum</i>

Legend: + Accepted by the author; - Not cited by the author; * Cited by the author under another name or as a synonym; ! Excluded from the complex. Ps.: *Briza brasiliensis sensu* Ekman is excluded by Matthei (1975), but is maintained as *sensu* Nees in *Poidium*; *B. bidentata* is excluded from the complex by Nicora and Rógolo de Agrasar (1981) and treated as *Rhombolytrum* in the same work (thus, five genera can be considered in this classification).

Phylogenetic studies based on molecular data have been a powerful tool to help in solving several distinct taxonomic problems, with special contribution to the groups whose morphological classifications are confuse, where there is too much disagreement, or for which the morphological characters available are not enough to produce congruent or

biologically sense-rich classifications. Nowadays, phylogenetic classifications are increasingly required, since they help identify true homologies in a group. Predictability is certainly a necessity, not a whim, and even when a new classification based on [good] DNA data looks very unlikely or unhelpful, sooner or later some new or understudied characters will appear and achieve the much-needed predictability.

The better understanding of the evolution of the so called *Briza* Complex, which can involve a single *lato sensu* genus, or even eight small genera (*Briza*, *Calotheca*, *Chascolytrum*, *Erianthecium* Parodi, *Gymnachne*, *Microbriza*, *Poidium* and *Rhombolytrum*), will help solve some morphological dilemmas.

This work aims to infer the phylogeny of the *Briza* Complex based on molecular data, and to propose genera circumscription based on monophyletic groups.

Three distinct phylogenetic analysis methods were applied, with a data set constituted by three separated DNA fragments. Confidence tests were also performed, and the supported groups are discussed, considering a few morphological aspects and previous works on the complex.

Materials and Methods

This study includes a total of 44 accessions, including the 19 species belonging to *Briza Complex sensu* Matthei (1975), tree new taxa, accessions of the species included in synonymy or from related genera - *Briza bidentata* Roseng., Arrill. & Izag. (transferred for some authors to *Rhombolytrum*), *Briza itatiaiae* Ekman (included in the synonymy of *B. brasiliensis sensu* Matthei), *Gymnachne koelerioides* (Trin.) Parodi, *Erianthecium bulbosum* Parodi – and four other species from the tribes Poeae and Bromeae used as

outgroup. All the species already cited from the *Briza* Complex or considered closely related were included in this study, except *B. humilis* M. Bieb. and *Rhombolytrum rhomboideum* Link. For purposes of simplification, throughout this paper a *lato sensu* circumscription of *Briza* is considered. Species with high morphological variability were sampled more than once, and their sequences were included as consensus, where sites with multiple possible bases were coded as following: R=A/G, Y=C/T, M=A/C, K=C/T, S=C/G, W=A/T, H=A/C/T, B=C/G/T, V=A/C/G, D=A/G/T and N=A/C/G/T. Some extra accessions with extreme morphological features were not included as consensus sequences, but as separate evolutionary units (EU). The outgroup was constituted by one (Bayesian analysis) to four species, belonging to tribe Poeae *lato sensu* (i. e., Aveneae included) and Bromeae. The list of samples with authorities, localities, herbarium vouchers, and GENBANK accession numbers is shown in Table 2.

Leaves of fresh, silica gel dried or herbarium vouchers were collected for this molecular survey. Sequences already available at the GENBANK (<http://www.ncbi.nlm.nih.gov>) for *Poa annua* L., *Bromus catharticus* Vahl and *Briza minor* L. were incorporated into the data matrix and used for the phylogenetic analyses. All the sequences obtained were deposited in GENBANK (Table 2). DNA isolation followed a CTAB protocol adapted for minipreparations (Doyle and Doyle, 1987); DNA from herbarium vouchers was extracted with the same protocol, but changing the overnight precipitation period in isopropanol at -20°C for one week in isopropanol at -80°C . Herbarium samples from which only degraded DNA was obtained (*B. itatiaiae*, *Briza. aff. lamarckiana*, *Briza* sp. 2) were improved amplifying total DNA with the DNA Amplification Kit GenomiPhi (Amersham Biosciences). Amplification of the ribosomal ITS region (ITS1 - 5.8S - ITS2) was performed in 35 PCR cycles with hot-start (45°C -

1' 58°C-2' 72°C) followed by three minutes of extension at 72°C, using the external primers 92 and 75 described by Desfeux and Lejeune (1996). The reactions were prepared in a final volume of 25ul, following standard proportions of reagents, with the addition of 1ul of DMSO (2%) and 30-75ng of DNA. PCR amplification of the *trnL-trnL-trnF* region was carried out in 35 PCR cycles (1' 94°C- 1' 55°C- 2' 72°C) followed by three minutes of extension at 72°C, using the external primers C and F described by Taberlet et al. (1991). A total volume of 25ul standard mixes was prepared, adding 50-100ng of DNA. PCR amplification of the 3' region (exons 9-14 and introns) of the GBSSI gene (Granule-Bound Starch Synthase, or *Waxy*) was performed in five pre-PCR cycles (45'' 94°- 2' 65°-1' 72°) followed by 30 PCR cycles (30'' 94°C- 40'' 65°C-40'' 72°C) and seven minutes of extension at 72°C. The primers used were F-for and M-bac described by Mason-Gamer et al. (1998). PCR mixes in 25ul were prepared with the following composition: 2.5ul PCR Buffer 10x, 2.3ul MgCl₂ 25mM, 2.5ul dNTP mixture 40mM (10mM each dNTP), 0.2ul each primer (25pmole), 0.3ul *Platinum* Taq DNA Polymerase (Invitrogen), 1.5ul DMSO (3%), 50ng DNA, sterile milli-Q purified water to complete the volume.

Volumes with 5ul of PCR products were pre-treated with 3.3U SAP (Shrimp Alkaline Phosphatase) and 0.66U of Exonuclease I, except the GBSSI fragment for *B. parodiana* Roseng., Arill. & Izag., which was purified from 1.4% agarose gel using the *Purelink* Quick Gel Extraction Kit. The purified products were sequenced directly, in the ACTGene Laboratory (Centro de Biotecnologia, UFRGS, Porto Alegre, RS, Brazil) using the automatic sequencer ABI-PRISM 3100, with the same primers used for amplification, except some ITS samples, which were also sequenced with the internal primers ITS3 and 74, described by Desfeux and Lejeune (1996), and some *TrnL-trnL-trnF* samples, which were also sequenced with the primers D and E, described by Taberlet et al. (1991).

TrnL-trnL-trnF sequences were obtained for all EUs, and ITS sequences were obtained for all samples included except one. Most samples were sequenced for GBSSI (Table 2). The EUs without GBSSI sequences were included in the combined matrix as missing data. A multicode for DNA bases was applied for both consensus sequences and heterozygote positions (see first paragraph).

At first sequences obtained for every fragment were aligned by Clustal X 1.81 software (Thompson et al., 1997). Then sequences were manually added to the initial alignments with GeneDoc software (Nicholas, Nicholas and Deerfield, 1997). Gaps in the *trnL-trnL-trnF* matrix were coded as binary characters by their presence/absence; only gaps that were unambiguous, longer than 5 bp, and potentially informative were added and considered for the parsimony-based analyses. All other gaps were treated as missing data.

To test the phylogenetic signal of the DNA fragments, the G1 statistics were carried out for every fragment separately, through the generation of 10,000 random trees. In order to test the significance of the incongruence between the phylogenetic signals of the DNA fragments, the Partition Homogeneity Test (=Incongruence Length Difference – ILD; Farris et al., 1994) was carried out, implemented by the PAUP* 4.0b10 software (Swofford, 2002). Two comparisons were run: the GBSSI data set against the ITS data set, and then the nuclear dataset (GBSSI and ITS combined in a single matrix) against the chloroplast matrix (*trnL-trnF*). Each test was performed through heuristic searches with TBR as a swapping algorithm, 500 random-addition replications and MULPARS option ON.

Considering the result of the PHT performed, the phylogenetic analyses were run as two separate datasets (nuclear dataset, including GBSSI and ITS sequences, and chloroplast dataset, with only *trnL-trnL-trnF* sequences) with PAUP*, based on Maximum

Parsimony and Maximum Likelihood, and with MRBAYES v. 3.1.2 (Huelsenbeck and Ronquist, 2002), based on Bayesian inference.

Parsimony analysis was conducted on each data matrix, nuclear and chloroplast matrix, without weighting. Each data set was subjected to heuristic searches with TBR as a swapping algorithm, MULPARS option ON, and trees rooted with the outgroup (*Bromus catharticus*, *Amphibromus scarbrivalvis*, *Poa annua* and *P. bradei* Pilg.). Random-addition was applied to nuclear data (1000 replicates, one tree held each step), but simple addition with 10,000 Maxtrees was set to chloroplast matrix, due to tree overflow. All parsimonious trees found in every search were used to compute 50% majority-rule consensus trees. Branch support for the optimal trees found under the parsimony criterion was estimated through 1,000 bootstrap replicates (Felsenstein, 1985) using the fast stepwise addition option. The level of homologous data in the trees was estimated by Consistency Index (CI), Retention Index (RI) and Rescaled Consistency Index (RC), implemented by PAUP*.

The Bayesian inference and likelihood searches were performed with the models of DNA evolution chosen with the help of the ModelTest 3.06 software (Posada and Crandall, 1998). The models chosen were calculated through AIC (Akaike Information Criterion).

Likelihood analyses were performed with heuristic searches with simple-addition and addition of taxa type as-is. The models applied were: K81uf+G for chloroplast matrix, indicated through hLTR test too, and TrN+G for nuclear matrix. Branch support for the optimal trees found under the likelihood criterion was estimated through 150 bootstrap replicates (Felsenstein, 1985) with a fast stepwise addition option. The outgroup was the same as described above for Parsimony.

Bayesian inference used the same substitution models for both data matrixes (GTR+G) and it was performed through 1,000,000 generations by the Markov Chain Monte Carlo (MCMC) sampling trees every 100 generations. *Bromus catharticus* was chosen as the outgroup. Phylogenies sampled from their posterior probability distribution were analyzed in order to observe the number of generations of trees needed to converge to a stable likelihood value for each separate data set (Huelsenbeck and Ronquist, 2002). Stationarity was achieved when the loglikelihood scores of sample points plotted against generation time reached a stable equilibrium value. Sampled points from generations before stationarity were discarded manually, comparing the values of the logfiles and the tree files. All remaining trees were used to construct the respective 50% majority-rule consensus trees, where the percentage of times that a clade is recovered is interpreted as an estimation of robustness. The consensus trees were calculated with the Consense software, implemented by the Phylip (PHYLogeny Inference Package) version 3.66 software package (Felsenstein, 2006).

All the consensus trees presented were edited with the TreeView version 1.6.6 software (Page, 2001).

Table 2: Source and GENBANK accession for included species

Species	Source (Geographical origin, collector number and voucher)	GENBANK accession (<i>trnL</i> - <i>trnL-trnF</i> , ITS and GBSSI, respectively)
Outgroup		
<i>Amphibromus scabrivalves</i> (Trin.) Swallen	S7015, MO	
<i>Bromus catharticus</i> Vahl	Brazil, RS, Porto Alegre Li20, ICN 132.519	- -AF521898 -DQ157055
<i>Poa bradei</i> Pilg.	HI8714, ICN	
<i>Poa annua</i> L.	Brazil, RS, Porto Alegre Li23, ICN 132.522	- -AF521901 -
<i>Ingroup</i>		
<i>Briza aff. juergensii</i>	Brazil, SC, São Joaquim HI8759, ICN 136.601	
<i>Briza calotheca</i> Hack.	Brazil, RS, Tainhas/Terra de Areia ZrHI1452, ICN 98.925	
<i>Briza ambigua</i> Hack.	Brazil, SC, Urubici Li190, ICN 132.689	
<i>Briza bidentata</i> Roseng., Arrill. & Izag.	Brazil, RS, Piratini Li55, ICN 132.554	

<i>Briza brachychaete</i> Ekman	Brazil, RS, São José dos Ausentes, Li151, ICN 132.650	
<i>Briza brasiliensis</i> (Nees ex Steud.) Ekman	Brazil, RJ, Itatiaia Rm357, ICN 149.151	
<i>Briza brizoides</i> (Lam.) Kuntze	Chile, Bio Bio S7014	
* <i>Briza calotheca</i> (Trin.) Hack.	Brazil, RS, São Francisco de Paula HILi8062, ICN 135.199	
* <i>Briza calotheca</i> (Trin.) Hack.	Brazil, MG, Serra da Caraça HI9662, ICN 136.997	
<i>Briza erecta</i> Lam.	Uruguay, Carrasco HI5056, ICN	
<i>Briza itatiaiae</i> Ekman	Brazil, RJ, Macieiras HI1548, ICN 49.093	
<i>Briza juergensii</i> Hack.	Brazil, SC, São Joaquim HI8713, ICN 136.595	
<i>Briza juergensii</i> Hack.	Brazil, SC, Urubici Li186, ICN 132.685	
<i>Briza lamarckiana</i> Nees	Brazil, RS, São Francisco de Paula HILi8060, ICN 135.197	
<i>Briza lamarckiana</i> Nees [lemma back not typical]	Brazil, RS, São Francisco de Paula	

	HILi8058, ICN 135.195	
* <i>Briza macrostachya</i> (J. Presl) Steud.	Brazil, RS, São Pedro do Sul Li13, ICN 132.512	
* <i>Briza macrostachya</i> (J. Presl) Steud.	Brazil, RS, São Pedro do Sul Li18, ICN 132.517	
* <i>Briza macrostachya</i> (J. Presl) Steud.	Brazil, RS, Bagé/Minas de Camaquã HI5038, ICN 131.396	
<i>Briza maxima</i> L.	Brazil, RS, São Lourenço do Sul Li45, ICN 132.544	
<i>Briza media</i> L.	Cultivated (caryopses from Austria, Tirolia) Li256, ICN 134.853	
<i>Briza minor</i> L.	Brazil, RS, Porto Alegre Li1, ICN 132.500	- -L36510 -
<i>Briza monandra</i> (Hack.) Pilg.	Bolivia, La Paz, P.D. Murillo, Unduavi Sb30397, LPB, ICN 149.157	
<i>Briza monandra</i> (Hack.) Pilg. * <i>Briza paleopilifera</i> Parodi	Brazil, SC, Urubici, Ib1356, ICN 132.576 Argentina, Misiones, Concepción de la Sierra, Concepción de la Sierra/Puerto Concepción Z8119, BAA	

* <i>Briza paleopilifera</i> Parodi	Argentina, Misiones, Cainguás, Salto Golondrina Z8173, BAA	
<i>Briza parodiana</i> Roseng., Arrill. & Izag.	Brazil, RS, Piratini Li290, ICN 134.887	
* <i>Briza poaemorpha</i> (J.Presl) Henr.	Brazil, RS, Camaquã HI5012, ICN 131.371	
* <i>Briza poaemorpha</i> (J.Presl) Henr.	Brazil, RS, São Francisco de Paula HILi8056, ICN	
* <i>Briza rufa</i> var. <i>rufa</i> (J.Presl) Steud.	Brazil, RS, Caxias do Sul Rt 125, ICN 149.156	
* <i>Briza rufa</i> var. <i>sparsipilosa</i> Roseng., Arrill. & Izag.	Brazil, RS, São Francisco de Paula HILi8061, ICN 135.198	
<i>Briza scabra</i> (Nees ex Steud.) Ekman	Brazil, RS, Soledade Li101, ICN 132.600	
* <i>Briza subaristata</i> Lam.	Brazil, RS, Porto Alegre Li8, ICN 132.507	
* <i>Briza subaristata</i> Lam.	Brazil, RS, Bagé/Minas de Camaquã HI5040, ICN 131.398	
* <i>Briza subaristata</i> var. <i>interrupta</i> (Hack. ex Stuck.) Roseng., Arrill. & Izag.	Brazil, RS, Porto Alegre Li19, ICN 132.518	

* <i>Briza uniolae</i> (Nees) Steud.	Brazil, RS, São Francisco de Paula HILi8059, ICN 135.196	
* <i>Briza uniolae</i> (Nees) Steud.	Brazil, RS, São Francisco de Paula HILi8057, ICN 135.194	
<i>Briza</i> sp. 1	Brazil, SC, Urubici Li201, ICN 132.700	
<i>Briza</i> sp. 2	Bolivia, Chuquisaca, Oropeza W10768, LPB	
<i>Erianthecium bulbosum</i> Parodi	Brazil, RS, Piratini Li60, ICN 132.559	
<i>Gymnachne koelerioides</i> (Trin.) Parodi	Chile S7035	

Accessions with asterisk indicate EUs included as species-consensus at the matrixes. Abbreviations for collector names: Hl= H.M.

Longhi-Wagner, Ib= I. Boldrini, Li= L. Essi, Rm= R. Moura, Rt= R. Trevisan, S= R. Soreng, Sb=S. Beck, W= J.R. Wood, Z= F. Zuloaga,

Zr= Z. Rúgolo.

Results

Sequence Analysis

The entire ITS region, including both spacers and the 5.8S subunit, of 43 accessions ranged from 586 bp in *Briza brasiliensis* and *B. itatiaiae* to 621 bp in *B. subaristata* and this length variation is similar to other published sequences for Poaceae (Grebenstein et al., 1998; Hsiao et al., 1999; Hodkinson et al., 2000; Catalán et al., 2004; Souza-Chies et al.,

2006; Essi and Souza-Chies, 2006). The ITS1 region ranged from 211 bp in *B. brasiliensis* and *B. itatiaiae* to 236 bp in *B. bidentata*. The ITS2 region ranged from 213 bp in *Amphibromus scabrivalvis*, *Poa bradei* and *B. brachychaete* Ekman to 228 bp in *B. subaristata*. The 5.8S subunit sequence was the most conserved region and was 157-158 bp long in all species sequenced. Most indels in the sequences are small (1-4 bp), small inversions (1-2 bp) also being frequent. Alignment of 34 sequences resulted in a matrix of 642 alignment positions with the introduction of indels. No evidence of obvious ITS length variants, multiple rDNA repeat types, was observed in any of the accessions analyzed. Many sites presented more than one possible base, which were included through a special coding (see Material and Methods). The addition of multiple accessions for some species amplified the ingroup, promoted the increase of homoplasious data. Intending to minimize this problem without excluding the information on intra-specific sequence variability, some accessions belonging to the same species were included as consensus, with good results. Accessions corresponding to extreme morphological variation, which were under taxonomic evaluation, were kept separate, to avoid mistaken combinations of distinct taxa. Accessions included as consensus were submitted separately at GENBANK, and are indicated with an asterisk in Table 2. The same accessions were included as consensus into the *trnL-trnL-trnF* matrix, but the intra-specific variation at molecular level was insignificant, sometimes inexistent.

The sequenced plastid region for 35 accessions, including the *trnL* intron and the *trnL-trnF* spacer, ranged from 826 bp in *Briza media* to 987 bp in *Poa bradei*. The indels presented several dimensions, normally from a single base up to 7-bp indels, but there were also two gaps extrapolating this pattern, one 20-bp gap, unique to *P. bradei*, and another 133-bp deletion, shared by the ingroup. The sequences presented several regions with

repetitive bases, but none of the indels could be attributed to a clear sequence reversion or duplication. The alignment resulted in a matrix of 1,028 alignment positions with introduction of the indels.

The sequenced GBSSI region included five introns and six exons. The size of the indels ranged from one bp to 30 bp, with the exception of a 103-bp indel exclusive for *Bromus catharticus*. All the sequenced species presented a short intron ten (11 bp), *B. catharticus* being responsible for its enlargement due to the 103-bp indel in this region. Davis and Soreng (2004) referred to the loss of GBSSI intron ten as a synapomorphy of Poae *sensu lato*. The alignment of 28 sequences resulted in a matrix of 1,301 alignment positions with introduction of indels. Considering the two matrixes, a total of 2,980 characters were included in this study. All the alignments are available upon request from the corresponding author.

Phylogenetic Analysis

The Partition Homogeneity Test resulted in a value of 0.13 for the ITS X GBSSI comparison, which meant a non-significant incongruence, but resulted in a value of 0.01 for the following nuclear X chloroplast comparison, indicating a significant level of incongruence. High levels of incongruence can result in polytomization or loss of phylogenetic information regarding one or more data sets, when these data are combined. In this case, running separate analyses of the incongruent data sets can improve the information available in the trees. Therefore it we chose to run the analyses in two different matrices.

Parsimony searches resulted in 10,000 equally parsimonious trees to chloroplast matrix, built based on 36 Parsimony-informative characters (CI=0.95, RI=0.89, RC=0.85; steps=180), and 1,483 equally parsimonious trees to nuclear matrix, based on 293

parsimony-informative characters (CI=0.76, RI=0.63, RC=0.48; steps=1,178). Ten thousand equally parsimonious chloroplast trees were built, based on 42 parsimony-informative characters, including coded gaps, each of them 190 steps long (CI=0.94, RI=0.89, RC=0.84).

The Bayesian analyses reached the likelihood stationarity in distinct generations. In chloroplast analysis, likelihood stationarity was reached approximately at generation 517,100. The likelihood stationarity for the nuclear matrix was reached earlier, approximately at generation 12,200.

Figures 1 and 2 show phylogenetic trees produced based on the nuclear matrix, and figures 3 and 4 present phylogenetic trees produced based on the chloroplast matrix. Likelihood and Bayesian trees are presented combined in the same pictures, since the topologies are similar. Branch lengths correspond to the Maximum Likelihood searches and numbers above and below branches indicate majority rule consensus greater than 95 for Bayesian, and bootstrap values for maximum likelihood searches greater than 50, respectively.

The three phylogenetic methods applied generated similar consensus trees. Groups with bootstrap support were also similar among the different methods, which diminishes the possibility of some relationships being due to method bias, but there were several conflicts for the trees generated based on distinct data sets.

Two main clades can be identified in all chloroplast analyses: a group constituted by Eurasian species and another group constituted by all the South American species. The clades can also be identified in the nuclear analysis, however with *Briza media* placed inside the South American group instead of placed with the Eurasian species. Thus,

considering the nuclear data set, there is a clade of annual species (*B. minor* and *B. maxima*) and another of perennial ones.

The conflicting placement of *Briza media* is most evident, but other conflicts inside the South American group deserve to be analyzed: the placement of *Erianthecium bulbosum* grouped with *B. brizoides* in chloroplast trees, or with *B. monandra* in nuclear trees, the relationship of *B. poaemorpha*, linked to an accession of *B. juergensii* in nuclear trees, but linked to *B. parodiana* in chloroplast trees, and the position of *B. paleapilifera*, placed with *B. erecta* in nuclear trees, and with an accession of *B. calotheca* in chloroplast trees.

In the South American/Perennial clade, several other relationships should be emphasized: 1) Genus *Erianthecium*, sometimes considered related to the complex but never included in it, is placed inside it in all trees; 2) Genus *Gymnachne*, which is morphologically related to the complex through *Briza bidentata*, is placed inside the South American clade, but its relationships with the other species have not been not well resolved; 3) *Briza ambigua* and *B. bidentata*, excluded from the complex by Nicora and Rúgolo de Agrasar (1981), are included in the South American group.

Discussion

Phylogenetic relationships and possible circumscriptions

The monophyly of the South American group, according to the plastid matrix, or consisting of a perennial group considering a nuclear matrix, is supported for all analyses. However, the immediate common origin for the two main clades is not supported by Bayesian or Parsimony analysis of nuclear matrices, and is weakly supported by likelihood

analysis of nuclear data. It is supported by chloroplast data in parsimony and likelihood analyses, but not in Bayesian analysis. This situation should discourage authors from accepting a single *lato sensu* genus for the complex. A preferable solution would be to consider a small genus *Briza* including only Eurasian species, and another larger genus, from South America. This is in agreement with the results obtained by King (1986), based on the taxometric analysis of seed proteins, by Savchenko et al. (1998), based on the study of phytoecdysteroids distribution, and by Williams and Murray (1972), based on the study of flavonoid variation.

However, the ambiguous position of *Briza media* requires some reflection on the common history of the groups. Many authors already recognized the morphological relationships between Eurasian and American species, even when preferring to split the group into distinct genera (Matthei, 1975; Nicora and Rógolo de Agrasar, 1981).

Previous studies suggested the involvement of *Briza media* both in the origin of the Eurasian (Matthei, 1975) and the South American group (Murray, 1976; Harborne, 1977). The participation of *B. media* in the origin of the Eurasian group can be ruled out here, since the early-diverging species of the Eurasian group is *B. maxima*. The involvement of *B. media* as an early-diverging element for the South American group can also be refuted, both because of its clear evolutionary placement within the Eurasian clade (chloroplast trees) and due to its grouping with *B. bidentata* (nuclear trees), instead of a sister position in relation to other South American species.

It appears more likely that the contribution of *Briza media* to the evolution of the South American species is related to past hybridization events. Interestingly, all the American species with chromosome counts available are tetraploid ($2n=28$, $X=7$), while the Eurasian species are diploid ($2n=14$, $X=7$ or $2n=10$, $X=5$ for *B. minor*) (Matthei, 1975;

Sampaio, Hickenbick and Winge, 1979; Schifino and Winge, 1983). A major exception is *B. media*, which presents both diploid ($2n=14$, $X=7$) and tetraploid ($2n=28$, $X=7$) races.

Briza media is an outbreeding species, self-incompatible, but able to produce hybrids with other self-compatible species, like *B. maxima* and *B. minor*. The American species are mainly self-compatible (Murray, 1974), but usually cleistogamous and particularly difficult to use in hybridization experiments, but some results show that many of the American species are compatible with some European species, and it may be possible to produce hybrids between species from both areas (Murray and Barker, 1988).

Although the majority of both Eurasian and South American species presents a common chromosome base count ($X=7$), the difference in ploidy level is added to the difference in DNA content among the clades. Schifino and Winge (1983) found out that the Eurasian species present a smaller number of chromosomes, but a higher DNA content per chromosome. In this case, if the South American species had originated from the Eurasian ones, as suggested by other authors, one possibility is that they lost a considerable amount of DNA during their evolution (Schifino and Winge, 1983). The topology of the trees contradict both the autopolyploid (Harborne, 1977) and allotetraploid (Murray, 1976a) origin of the South American group starting from *B. media*, but agree with possible past reticulation events involving *B. media* and other [probable] South American species. The disjunct distribution of the *Briza* Complex between America and Eurasia is also a non negligible argument for recognition of at least two distinct genera.

The circumscription of the South American genera is beyond doubt the most controversial. Based on morphology, some authors proposed very distinct circumscriptions, from a single genus, including the Eurasian group - with or without infra-generic categories (Parodi, 1920; Rosengurtt et al., 1968; Longhi-Wagner, 1987), up to several genera,

separating (Matthei, 1975) or not (Nicora and Rùgolo de Agrasar, 1981; Clayton and Renvoize, 1986) the Eurasian species. Other approaches, however, support *lato sensu* circumscriptions. Cytotaxonomic data did not permit any clear division of the South American species (Schifino and Winge, 1983), as well as taxometric analysis of seed proteins (King, 1986) and phytoectosteroids analysis (Savchenko et al., 1998). Based on numerical taxonomy, cytotoxic, chemiotaxonomic, pallinologic and some ontogenetic data, Longhi-Wagner et al. (1987) proposed a *lato sensu* taxonomic arrangement for genus *Briza*, recognizing five sections, instead of several genera, arguing that the sections were not supported for all analyses and the level of variation is lower than usually accepted to circumscribe other genera in the same Poaeae tribe. Analyzing all trees, genus *Poidium sensu* Matthei is the only one which could be weakly circumscribed within the South American group, but with some important problems: 1) The genus is separated throughout the nuclear trees, forming four small clades, one early-diverging, two without resolved basal relationships, and including representatives of other genera. 2) In the chloroplast trees, almost all *Poidium* species group in a single clade, but excluding *B. poaemorpha* and *B. monandra*, and including *B. paleapilifera*, the last a *Chascolytrum* representative. 3) The recognition of a *Poidium* genus based on a chloroplast tree would force the acceptance of paraphyletic groups for the remaining species, and genus *Poidium* would be clearly polyphyletic based on nuclear trees.

Small genera, like *Microbriza*, *Calotheca*, *Erianthecium*, *Gymnachne*, *Rhombolytrum* and *Lombardochloa*, cannot be considered monophyletic groups, some because of ambiguous placement, and some due to basal polytomies, which do not allow clear decisions. Genus *Microbriza*, for instance, should not be accepted, because its two

species are never together. Morphologically, *B. brachychaete* is more similar to *B. itatiaiae* than to *B. poaemorpha*, which is unique in several spikelet characters.

The initially surprising link between *B. brizoides* and *Erianthecium*, according to chloroplast data, can be morphologically explained by the shared presence of lemma awn and densely pilose paleas. Considering the nuclear data, the relationships of both species inside the American clade are still unclear.

The relationships among some accessions of multiple-sampled species were better studied using an additional molecular approach, an ISSR analysis, more appropriate to the species level. The DNA sequences appeared excessively conserved for species-level resolution (chloroplast matrix) or poorly resolved (ITS data), slightly increasing the level of homoplasious data in the trees. Despite these disadvantages, the multiple accessions guaranteed a sample of intra-specific diversity, which is important to avoid overestimating the three branches. Taxon sampling has less impact on phylogenetic accuracy than sequence sampling (Rokas and Carrol, 2005), but broad taxonomic sampling helps to avoid long-branch attraction, a problem common to all phylogenetic methods, but with a stronger effect in parsimony analysis.

Although the species-level circumscription problems will be discussed more appropriately using another approach, it is possible to comment on some relationships here.

Two new species, *Briza* sp. 1 and *Briza* sp. 2 (in preparation) appeared to be genetically clearly distinct EUs. One variety presented a placement, which denoted a genetic distance good enough to consider it as a new species, better than a new variety. However, morphologically there are only a few elements for such a decision, and a placement at variety level appears to be morphologically more appropriate.

The utility of DNA fragments

Regarding the utility of the selected DNA fragments to study the evolution of this complex, the chloroplast fragment appeared as highly conserved inside the South American group, being unable to resolve several relationships. Its high degree of conservation concerning this clade and the clear and supported distinction from the Eurasian sequences encourage us to accept the Eurasian clade as a distinct genus, taking *B. media* as an original EU for the Eurasian clade which is more likely than for the South American one, to which it seem to be linked due to past hybridization events. The Eurasian group is supported by several base substitutions and conserved indels of the chloroplast data. As a species-level marker, the chloroplast sequences contributed by grouping some accessions of the same species, even when they are from a disjunct distribution (*B. monandra*). Unfortunately, the conservation of the fragment did not allow resolution in most cases.

On the other hand, ITS sequences concentrated the highest number of changes. They did not appear to be a good species-level marker, but considerably improved the resolution for all clades of the trees. The regions with more variation were ITS1 and ITS2, but they were also the most homoplasious. Although the 5.8S was short, it also contributed parsimony-informative characters.

The inclusion of GBSSI sequences was extremely important, even if it was not available for all taxa, both to confirm some surprising relationships detected by ITS sequences (the conflicting placement of *B. media*, for instance), and to improve the resolution of nuclear trees. In terms of level of information, the GBSSI appeared as intermediate between the *trnL-trnL-trnF* and the ITS sequences.

ITS and *trnL-trnL-trnF* sequences are certainly the most popular and frequently the first choice of a great number of researchers. *TrnL-trnL-trnF* is certainly easy to amplify

and to sequence, even from partially degraded DNA, and has become especially popular due to the existence of universal primers described (Taberlet et al., 1991). The use of ITS sequences is more contradictory, but all the same, it is still one of the best options for nuclear representation and covers most phylogenetic studies (Álvarez and Wendel, 2003). They are relatively easy to amplify, and present a sequence homogenization process called concerted evolution, which allows the sequences amplified to be treated as a single one. Another point that makes their use attractive is the high number of nucleotide substitutions, which are quickly accumulated, making it useful for low-level taxonomic studies, like phylogeny of genera and species-complexes (Buckler and Holtsford, 1996; Torrecilla and Catalán, 2002; Guo et al., 2002; Barker et al., 2003; Blattner, 2004). The disadvantages are linked to its multi-copy nature: sometimes the concerted evolution is not completely effective, leaving a few partially homogenized copies, allowing a certain level of lineage sorting, putting the researchers at risk of comparing paralogous copies; the concerted evolution can homogenize copies in a certain direction, which is not necessarily the same of the species phylogeny (for a review of concerted evolution, see Liao, 1999). Another aspect is that the high number of substitutions accumulated can generate homoplasious data and phylogenetic noise.

The GBSSI has been very useful due to its low copy number. For grasses, it has been described as a single copy gene (Mason-Gamer et al., 1998; Mason-Gamer, 2001; Baumel et al., 2002; Mathews et al., 2002; Guo and Li, 2004), enabling sequencing without cloning.

The inclusion of both chloroplast and nuclear sequences in a phylogenetic analysis is extremely important, especially in groups where reticulation is frequent, like the grasses.

Incongruence between chloroplast and nuclear sequences is relatively common (Kellogg and Mason-Gamer, 1996; Mason-Gamer and Kellogg, 1996; Catalán et al., 2004; Fehrer et al., 2007), but usually difficult to interpret. Two main explanations are usually considered: one (or both) fragment is not phylogenetically useful (due to lineage sorting, chloroplast capture, high levels of homoplasy, gene paralogy, etc) or reticulation has played an important role in the evolution of the group.

Considering that all fragments included in this study have already been recognized as phylogenetically useful, including for Poaceae genera (Mason-Gamer, 2001; Baumel et al., 2002; Blattner, 2004; Guo and Li, 2004), the first explanation is unlikely to be true here.

There is some risk in using ITS sequences, due to their complex and unpredictable behaviour, so that a certain level of lineage sorting cannot be ruled out. However, the ITS here are not the only representative of the biparental inheritance, and major relationships drawn by ITS were confirmed by GBSSI data.

The second explanation should be seriously considered. Hybridization plays a major role in angiosperm speciation (Stebbins, 1971; Arnold, 1992; Rieseberg and Carney, 1998). This is especially true for Poaceae, which has a high number of polyploids. Several examples of reticulate past events have been reported for Poaceae, besides for Pooideae (Soreng, 1990, Soreng and Davis, 2000). Although natural hybrids were not reported for the genus *Briza*, which is mostly cleistogamous in South America, intermediate forms among species are common, and the incongruence between chloroplast and nuclear trees seems to indicate hybridization events. These events are not necessarily recent. The ambiguous placement of *B. media* could indicate, for instance, an hybridization event involving an individual in the coalescent lineage of *B. media* and the common ancestor of

the South American group, or indicate any past horizontal transfer between the two lineages. The long branches of the Eurasian group could be explained by the group's normally shorter life cycle so that the mutations are fixed faster, or by an earlier divergence. In the latter case, a descendant of the *B. media* lineage could have interacted with a recently diverging element of the South American group. It is tempting to explain the origin of the South American group through allotetraploidization from *B. media*, as proposed by Murray (1976a), but this would require a next step of introgression with the unknown parental line to explain the non-basal position of *B. media* in the American clade. We believe that the network is much more complicated than this, and therefore it is better to admit simply that reticulated evolution has to be considered in this group, with a major role for *B. media*, but this process should be better explored with other approaches, including FISH (Fluorescent In Situ Hybridization).

Conclusion and future perspectives

It was impossible to trace the course of possible hybridization events, due to basal polytomies inside the South American clade. The inclusion of more DNA fragments does not assure better resolution, since some groups with rapid diversification may never be fully resolved. The South American group, however, deserves further studies, with special emphasis on the comprehension of relationships of species like *Erianthecium bulbosum* and *Briza brizoides*, which seem to be morphologically early-diverging, but appeared non resolved, and species with conflicting placement between nuclear and chloroplast trees.

The results obtained so far indicate a separation of the Eurasian group into a *stricto sensu* genus, as already proposed by Matthei (1975), and a single genus to the South American group, a circumscription that has not yet been published and which will require several new combinations.

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Figures

Figure 1: Majority-rule consensus trees of 1,483 equally parsimonious trees from nuclear matrix (ITS+GBSSI). Steps=1,178, CI=0.76, RI=0.63, RC=0.48. Numbers above branches: majority-rule values >50%. Numbers below branches: bootstrap support. Asterisks indicate consensus sequences for ITS.

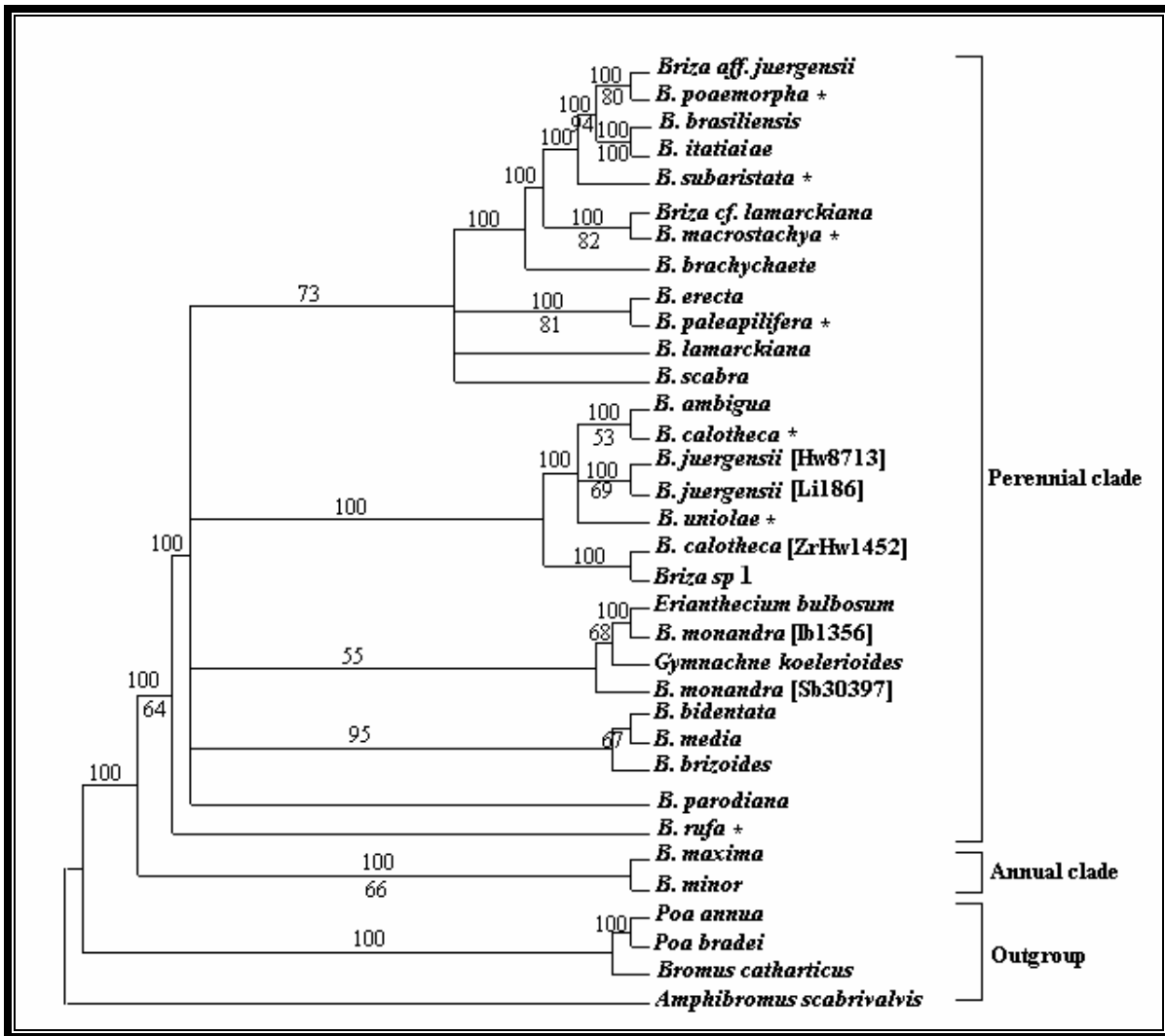


Figure 2: Likelihood phylogram inferred based on nuclear matrix (ITS+GBSSI) and TrN+G model. Likelihood=-9472.64979. Numbers above branches: posterior probabilities >95% for Bayesian analysis (same tree topology, branch-lengths not considered). Numbers below branches: bootstrap support for likelihood tree. Bars indicate the values for short branches. Asterisks indicate consensus sequences for ITS.

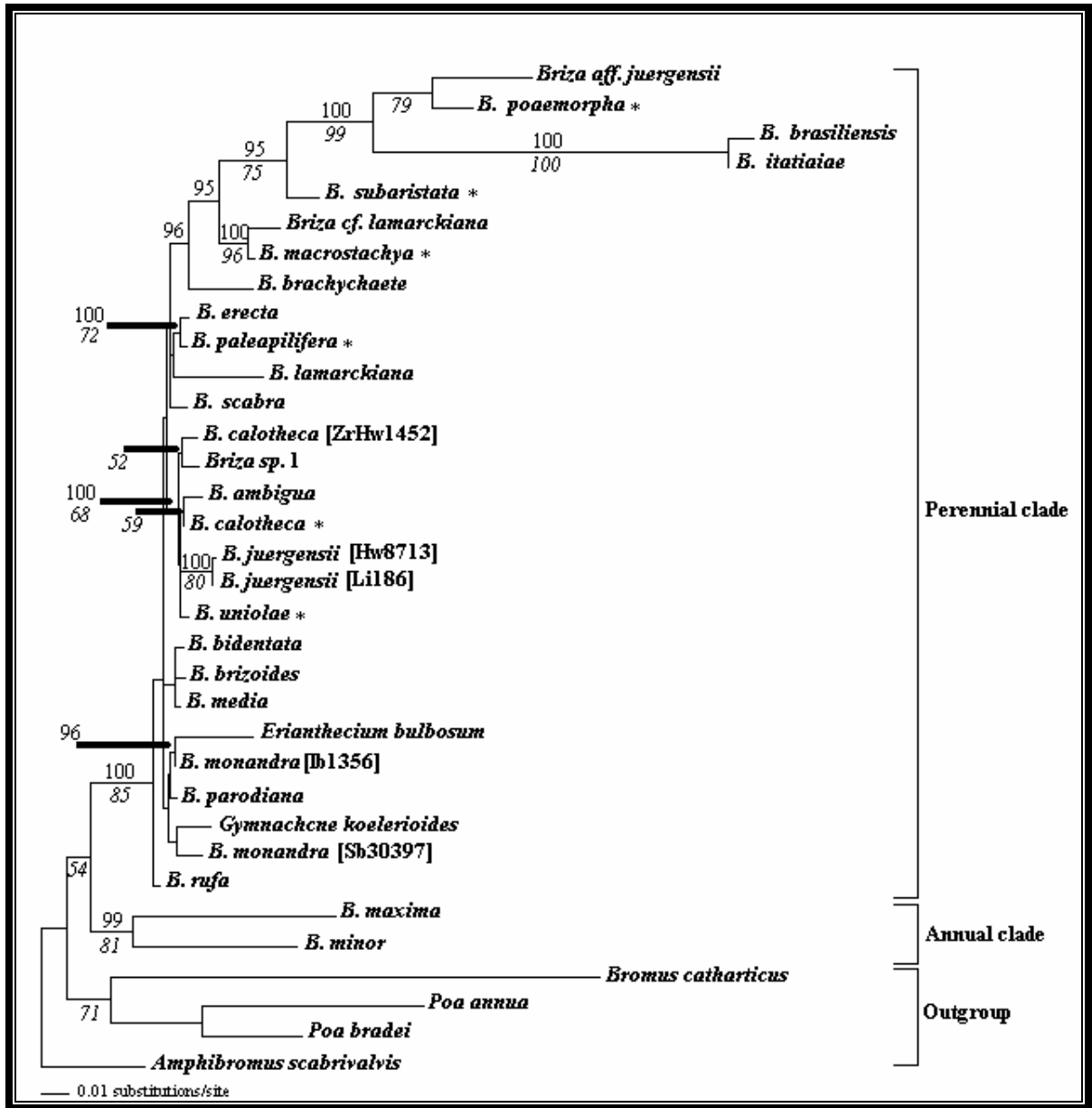


Figure 3: Majority-rule consensus tree of 10,000 equally parsimonious trees from chloroplast matrix, indels not coded (indels treated as missing data). Steps=180, CI=0.95, RI=0.89, RC=0.85. Numbers above branches: majority-rule values >50%. Numbers below branches: bootstrap support. Asterisks indicate consensus sequences.

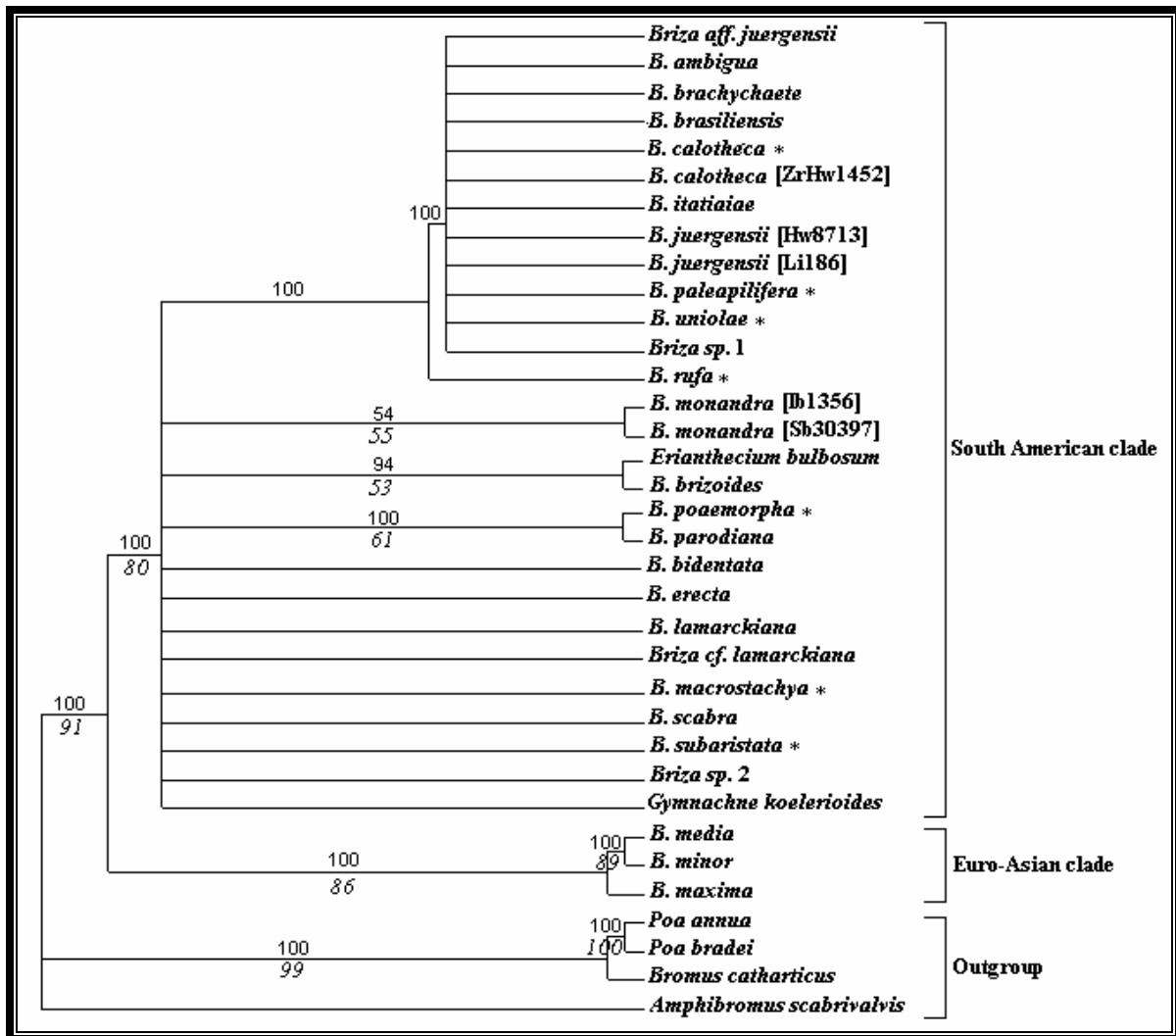
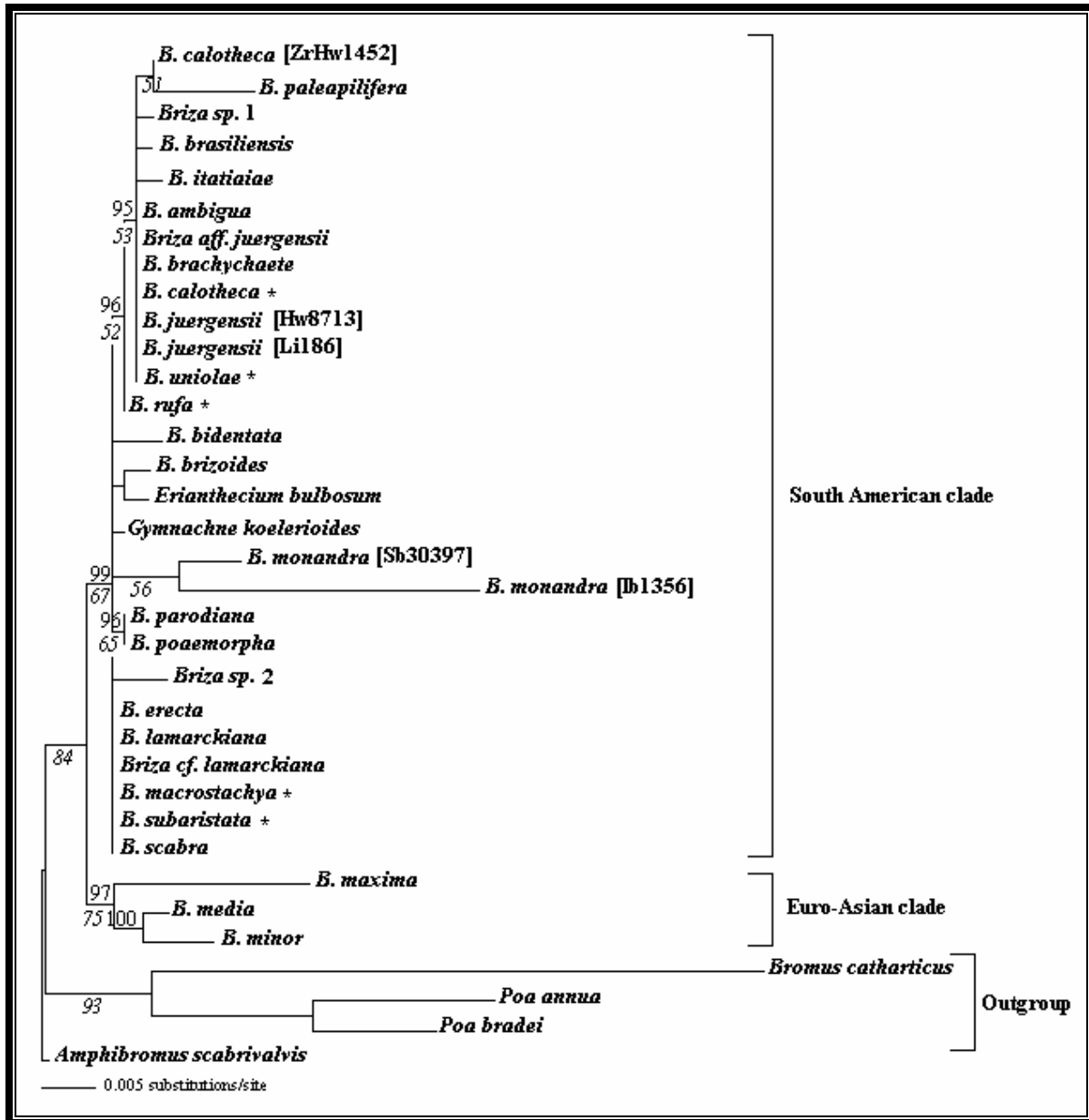


Figure 4: Likelihood phylogram inferred based on chloroplast matrix and K81uf+G model. Likelihood=-2496.80173. Numbers above branches: majority-rule consensus values >95% for Bayesian analysis (same tree topology, branch-lengths not considered). Numbers below branches: bootstrap support for likelihood tree. Asterisks indicate consensus sequences.



References

- Álvarez, I., Wendel, I.F. 2003. Ribosomal ITS sequences and phylogenetic inference. *Mol. Phyl. Evol.* 29, 417-424.
- Arnold, M.L. 1992. Natural hybridization as an evolutionary process. *Ann. Rev. Ecol. Syst.* 23, 237-261.
- Barker, N.P., Linder, H.P., Morton, C.M., Lyle, M. 2003. The paraphyly of *Cortaderia* (Danthonioideae: Poaceae): evidence from morphology and chloroplast and nuclear DNA sequence data. *Ann. Missouri Bot. Gard.* 90, 1-24.
- Baumel, A., Ainouche, M.L., Bayer, R.J., Ainouche, A.K., Misset, M.T. 2002. Molecular phylogeny of hybridizing species from genus *Spartina* Schreb. (Poaceae). *Mol. Phyl. Evol.* 22(2), 303-314.
- Bayón, N.D. 1998. Cladistic analysis of the *Briza* Complex (Poaceae, Poaeae). *Cladistics* 14, 287-296.
- Blattner, F.R. 2004. Phylogenetic analysis of *Hordeum* (Poaceae) as inferred by nuclear rDNA ITS sequences. *Mol. Phyl. Evol.* 33, 289-299.
- Born, I.F., Winge, H. 1980. Ontogenia comparada de espécies do complexo *Briza* (Gramineae). *Ciência e Cultura* 32, 683-684.
- Buckler, E.S., Holtsford, T.P. 1996. *Zea* Systematics: ribosomal ITS evidence. *Mol. Biol. Evol.* 13(4), 612-622.
- Catalán, P., Torrecilla, P., Rodríguez, J.A.L., Olmstead, R.G. 2004. Phylogeny of the festucoid grasses of subtribe Loliinae and allies (Poeae, Pooideae) inferred from ITS and *trnL*-F sequences. *Mol. Phyl. Evol.* 31, 517-541.

- Clayton, W.D., Renvoize, S.A. 1986. Genera graminum: grasses of the world. Royal Botanic Gardens, London.
- Davis, J., Soreng, R.J. 2004. A phylogenetic analysis of the grass subfamily Pooideae (Poaceae), with focus on the Poeae/Aveneae/Hainardieae complex, and the evolution of structural features and nuclear genomes. In: The Annual Botany Conference. The Botanical Society of America. Abstract ID: 346.
- Doyle, J.D., Doyle, J.L. 1987. A rapid DNA isolation procedure for small quantities of fresh leaf tissue. Phytochem. Bull. 19, 11-15.
- Desfeaux, C.; Lejeune, B. 1996. Systematics of Euromediterranean *Silene* (Caryophyllaceae): evidence from a phylogenetic analysis using ITS sequences. Compt. Rend. Acad. Sci. Paris 319, 351-358.
- Essi, L.; Souza-Chies, T.T. 2006. Phylogeny of Linearia and Notata groups of *Paspalum* L. (Poaceae, Panicoideae, Paniceae) and related species. Genet. Res. Crop Evol. DOI 10.1007/s10722-006-9148-7.
- Farris, J.S., Källersjö, M., Kluge, A.G., Bult, C. 1994. Testing significance of incongruence. Cladistics 10, 315-319.
- Fehrer, J., Gemeinholzer, B., Chrtek, J.Jr., Bräutigam, S. 2007. Incongruent plastid and nuclear DNA phylogenies reveal ancient intergenic hybridization in *Pilosella* hawkweeds (*Hieracium*, Cichorieae, Asteraceae). Mol. Phyl. Evol. 42, 347-361.
- Felsenstein, J. 1985. Confidence limits on phylogenies: an approach using the bootstrap. Evolution 39, 783-791.
- Felsenstein, J. 2006. PHYLIP (Phylogeny Inference Package) version 3.66. Distributed by the author. Department of Genome Sciences, University of Washington, Seattle.

Guo, Z., Chen, Y., Li, D. 2002. Phylogenetic studies on the *Thamnocalamus* group and its allies (Gramineae: Bambusoideae) based on ITS sequence data. *Mol. Phyl. Evol.* 22(1), 20-30.

Guo, Z., Li, D. 2004. Phylogenetics of the *Thamnocalamus* group and its allies (Gramineae: Bambusoideae): inference from the sequences of GBSSI gene and ITS spacer. *Mol. Phyl. Evol.* 30, 1-12.

Grebenstein, B., Röser, M., Sauer, W., Hemleben, V. 1998. Molecular phylogenetic relationships in Aveneae (Poaceae) species and other grasses as inferred from ITS1 and ITS2 rDNA sequences. *Pl. Syst. Evol.* 213, 233-250.

Harborne, J.B. 1977. Flavonoids and the evolution of the Angiosperms. *Biochem. Syst. Ecol.* 5, 7-22.

Hsiao, C., Jacobs, S.W.L., Chatterion, N.J., Asay, K.H. 1999. A molecular phylogeny of the grass family (Poaceae) based on the sequences of nuclear ribosomal DNA (ITS). *Austral. Syst. Bot.* 11, 667-688.

Hodkinson, T.R., Renvoize, S.A., Chonghaile, G.N., Stapleton, C.M.A., Chase, M.W. 2000. A comparison of ITS nuclear rDNA sequence data and AFLP markers for *Phyllostachys* (Bambusoideae, Poaceae). *J. Plant Res.* 113, 259-269.

Huelsenbeck, J.P., Ronquist, F. 2002. MRBAYES: Bayesian inference of phylogenetic trees. *Bioinformatics* 17(8), 754-755.

Kellogg, E.A., Mason-Gamer, R. 1996. When gene trees tell different stories: the diploid genera of Triticeae (Gramineae). *Syst. Bot.* 21, 321-347.

King, G.J. 1986. A taxometric analysis of seed proteins in the genus *Briza* s.l. (Poaceae). *Pl. Syst. Evol.* 151, 163-174.

- Leyser, V.E., Winge, H. 1979. Análise palinológica de espécies do complexo *Briza* (Gramineae). *Ciência e Cultura* 31, 577.
- Liao, D. 1999. Molecular Evolution '99 – Concerted evolution: Molecular Mechanism and Biological Implications. *Am. J. Hum. Genet.* 64, 24-30.
- Longhi-Wagner, H.M. 1987. Flora Ilustrada do Rio Grande do Sul, Fasc. 17. Gramineae. Tribo Poeae. *B. Inst. Bioc.* 41, 1-191.
- Longhi-Wagner, H.M., Winge, H., Hickenbick, M.C.M., Schifino, M.T., Torres de Lemos, C., Fonseca, I.A., Freitas-Sacchet, A.M.O., Passaglia, L.M.P. 1987. Sistemática e origem das espécies neotropicais de *Briza* L. (Gramineae). In: *Anales del IV Congreso Latinoamericano de Botánica, III. Simposio: Citología y citotaxonomía. Bogotá.*
- Mason-Gamer, R.J. 2001. Origin of North American *Elymus* (Poaceae: Triticeae) allotetraploids based on Granule-Bound Starch Synthase gene sequences. *Syst. Bot.* 26(4), 757-768.
- Mason-Gamer, R., Kellogg, E.A. 1996. Testing for phylogenetic conflict among molecular data sets in the tribe Triticeae (Gramineae). *Syst. Biol.* 45, 524-545.
- Mason-Gamer, R.J., Weil, C.F., Kellogg, E.A. 1998. Granule-Bound Starch Synthase: structure, function, and phylogenetic utility. *Mol. Biol. Evol.* 15(12), 1658-1673.
- Mathews, S., Spangler, R.E., Mason-Gamer, R.I., Kellogg, E.A. 2002. Phylogeny of Andropogoneae inferred from Phytochrome B, GBSSI, and *NDHF*. *J. Plant Sci.* 163(3), 441-450.
- Matthei, O. 1975. Der *Briza*-Komplex in Südamerika: *Briza*, *Calotheca*, *Chascolytrum*, *Poidium* (Gramineae). *Willdenowia* 8, 7-168.

- Moss, J.M., Murray, B.G. 1990. The three-dimensional arrangement of chromosomes at meiotic metaphase I in normal and interchange heterozygotes of *Briza humilis*. *J. Cell Sci.* 97, 565-570.
- Murray, B.G. 1974. Breeding systems and floral biology in the genus *Briza*. *Heredity* 33(2), 285-292.
- Murray, B.G. 1975. The cytology of the genus *Briza* L. (Gramineae). I. Chromosome numbers, karyotypes and nuclear DNA variation. *Chromosoma (Berl.)* 49, 299-308.
- Murray, B.G. 1976a. The cytology of the genus *Briza* L. (Gramineae). II. Chiasma frequency, polyploidy and interchange heterozygosity. *Chromosoma (Berl.)* 57, 81-93.
- Murray, B.G. 1976b. The cytology of the genus *Briza* L. (Gramineae). III. B Chromosomes. *Chromosoma (Berl.)* 59, 73-81.
- Murray, B.G. 1978. B Chromosomes and multivalent formation in tetraploid hybrids between *Briza media* and *Briza elatior*. *Heredity* 41, 227-231.
- Murray, B.G. 1979. Unusual chromosome pairing and B chromosomes in *Briza spicata* (Poaceae). *Pl. Syst. Evol.* 132, 245-253.
- Murray, B.G. 1984. The structure, meiotic behaviour and effects of B chromosomes in *Briza humilis* Bieb. (Gramineae). *Genetica* 63, 213-219.
- Murray, B.G. 1986. Interchange quadrivalents and chromosome order at meiotic metaphase I in *Briza* L. (Gramineae). *Chromosoma (Berl.)* 94, 293-296.
- Murray, B.G., Barker, N.R.N. 1988. Pollen/stigma interactions and hybridization in the genus *Briza* L. (Gramineae). *Evol. Trends Pl.* 2(2), 107-110.
- Nicholas, K.B, Nicholas, H.B.Jr., Deerfield, D.W. 1997. GeneDoc: Analysis and visualization of genetic variation. *EMBNEW.NEWS* 4, 14.

- Nicora, E.G., Rúgolo de Agrasar, Z.E. 1981. Los géneros sudamericanos afines a *Briza* L. (Gramineae). Darwiniana 23(1), 279-309.
- Page, R.D.M. 2001. TREEVIEW: An application to display phylogenetic trees on personal computers. Comput. Appl. Biosci. 12, 357-358.
- Parodi, L.R. 1920. Notas sobre las especies de *Briza* de la Flora Argentina. Rev. Fac. Arg. Agr. y Vet. B. Aires 3, 113-137.
- Posada, D., Crandall, K.A. 1998. MODELTEST: testing the model of DNA substitution. Bioinformatics applications note. 14(9), 817-818.
- Rieseberg, L.H., Carney, S.E. 1998. Plant hybridization. New Phytol. 140, 599-624.
- Rokas, A., Carroll, S.B. 2005. More genes or more taxa? The relative contribution of gene number and taxon number to phylogenetic accuracy. Mol. Biol. Evol. 22(5), 1337-1344.
- Rosengurtt, B., Arrillaga de Maffei, B., Izaguirre de Artucio, P. 1968. Sinopsis de *Briza* (Gramineae) del Uruguay y notas taxonomicas sobre otras especies de este genero. Bol. Fac. Agr. Montevideo 105, 1-35.
- Rosengurtt, B., Arrillaga de Maffei, B. 1979: *Lombardochloa*, nuevo genero de Gramineae. Anales Fac. Quím. 9, 255-288.
- Sampaio, M.T.S., Hickenbick, M.C.M., Winge, H. 1979. Chromosome numbers and meiotic behavior of South American species of the *Briza* complex (Gramineae). Rev. Bras. Genet. 2(2), 125-134.
- Savchenko, T., Whiting, P., Šik, V., Underwood, E., Sarker, S.D., Dinan, L. 1998. Distribution and identities of phytoecdysteroids in the genus *Briza* (Gramineae). Biochem. Syst. & Ecol. 26, 781-791.
- Schifino, M.T., Winge, H. 1983a. Circadian rhythms of mitotic divisions in seedling meristem of *Briza uniola* (Nees) Steud. (Gramineae). Rev. Bras. Genet. 6 (3), 575-578.

- Schifino, M.T., Winge, H. 1983b. Karyotypes and nuclear DNA content of species of the *Briza* complex and some other genera of Poaceae (Gramineae). Rev. Bras. Genet. (Brazil. J. Genetics) 4(2), 245-259.
- Soreng, R.J. 1990. Chloroplast-DNA phylogenetics and biogeography in a reticulating group: study in *Poa* (Poaceae). Amer. J. Bot. 77(11), 1383-1400.
- Soreng R.J., Davis, J.I. 2000. Phylogenetic structure in Poaceae subfamily Pooideae as inferred from molecular and morphological characters: misclassification versus reticulation. Grasses Systematics and Evolution. Eds. S.W.L.Jacobs and J.Everett. CSIRO: Melbourne.
- Souza-Chies, T.T., Essi, L., Rua, G. H., Valls, J.F.M., Miz, R.B. 2006. A preliminary approach to the phylogeny of the genus *Paspalum* (Poaceae). Genetica 126, 15-32.
- Stebbins, G.L. 1971. The role of hybridization in evolution. In: Processes of Organic Evolution. Prentice-Hall, Englewood Cliffs, New Jersey, pp. 116-135.
- Swofford, D.L. 2002. PAUP*. Phylogenetic Analysis Using Parsimony (*and other methods). Version 4. Sinauer Associates, Sunderland.
- Taberlet, P., Gielly, L., Pautou, G., Bouvet, J. 1991. Universal primers for amplification of three non-coding regions of chloroplast DNA. Plant Mol. Biol. 17, 1105-1109.
- Thompson, J.D., Gibson, T.J., Plewniak, F., Jeanmougin, F., Higgins, D.G. 1997. The Clustal X windows interface: flexible strategies for multiple sequence alignment aided by quality tools. Nucleic Acids Res. 24, 4876-4882.
- Torreçilla, P., Catalán, P. 2002. Phylogeny of broad-leaved and fine-leaved *Festuca* lineages (Poaceae) based on nuclear ITS sequences. Syst. Bot. 27(2), 241-251.
- Williams, C.A., Murray, B.G. 1972. Flavonoid variation in the genus *Briza*. Phytochemistry 11, 2507-2512.

Winge, H., Hickenbick, M.C.M, Longhi-Wagner, H.M., Torres de Lemos, C., Schifino, M.T., Fonseca-Born, I., Moure, J.S., Leyser da Rosa, V. 1984. Sistemática e evolução das espécies sul-americanas do Complexo *Briza* (Gramineae). In: Colóquio Sobre Citogenética e Evolução de Plantas, 1. Sociedade Brasileira de Genética, Piracicaba, pp. 185-203.

Capítulo II

Can ISSR markers help to solve taxonomic controversies? A first approach for species of the *Briza* Complex in South America (Poaceae)

Abstract

Inter-Simple Sequence Repeat (ISSR) markers have been widely used for several purposes, including systematics. In this work, four groups of closely related species of *Briza* from South America were evaluated based on ISSR markers.

Nine primers with an average of 89 fragments per group were used to build similarity dendrograms. The similarity indexes found were low (the average values per group ranged from 0.2771 to 0.5066), 93% of the bands being polymorphic. Some taxonomic problems could be clarified, although marker bands were not identified to species-level. ISSR were able to separate the two varieties of *Briza rufa*, supporting their acceptance. Two new species could be clearly separated from their morphologically related taxa, but *B. erecta*, *B. macrostachya* and *B. subaristata*, as well as *B. subaristata* var. *interrupta*, could not be separated, adding elements to the synonymization of these three taxa. The ISSR were able to group even accessions of disjunct regions (*B. monandra*), but grouped them with a lower than average index and detected a high genetic variability within the apparently homogeneous *B. bidentata*, not grouping one of the accessions. The ISSR contributed to solving some controversies on genus *Briza*, but its use as a species-marker is limited in the group, so that studies should be restricted to complexes of two or three species only.

Keywords: ISSR, *Briza*, Poaceae, systematics.

Introduction

Inter-Simple Sequence Repeat (ISSR) PCR is a fast, cheap approach to genetic characterization and is widely used for ecological (Wu et al., 2004; Poulin, Weller & Sakai, 2005), population (Esselman et al., 1999; Camacho & Liston, 2001; Alexander, Liston & Popovich, 2004; Wróblewska et al., 2003) or phylogenetic studies (Joshi et al., 2000; Bussell, Waycott & Chappill, 2005), as well as in crop cultivar or native plant fingerprinting (Mattioni et al., 2002; Souza et al., 2005).

It has been also applied to explore the hybrid origin of species (Ayres & Strong, 2001) and for systematic purposes (Blair et al., 1999; Vanderpoorten et al., 2003; Dinelli, 2004).

ISSRs have a few advantages over other markers. Their primers anneal to SSR (Simple Sequence Repeat) that are abundant throughout the eukariotic genome and evolve rapidly, and hence may reveal a high level of polymorphism (Zietkiewicz et al., 1994; Li & Ge, 2001). In addition, ISSR may produce more reliable and reproducible bands than RAPD (Random Amplified Polymorphic DNA), due to the higher annealing temperature and longer primer sequences (Qian et al., 2001).

It is often difficult to interpret morphological divergences among closely related taxa. Minor morphological differences can be a result of environmental variation, species polymorphism or even of evolutionary divergence processes, such as genetic isolation or speciation. The presence or not of trichomes on certain structures, for instance, may be taxonomically negligible for some groups, or be a stable and reliable morphologic marker for other taxa.

Decisions concerning accepting or not certain features such as taxon markers or like establishing a given taxonomic level for a morphologically distinct group, may give systematics a certain level of subjectivity. In such cases, the addition of extra approaches can bring light and objectivity to the studies.

Genus *Briza* L. *lato sensu* (Poaceae) is a morphologically diverse and taxonomically controversial genus from subtropical and temperate areas. It is represented by Eurasian and South American species, which can be separated into several genera. The circumscriptions of the genera change according to the author, and the group is under taxonomic and phylogenetic evaluation by our team (in preparation). The South American group is the most controversial, and besides the circumscription controversies at genus level, there are some doubts at species-level and sub-specific level. It is not clear, for instance, whether species like *B. erecta* Lam. and *B. macrostachya* (J. Presl) Steud deserve to be treated as separate taxa, as done by Longhi-Wagner (1987), or whether they represent only intra-specific variation, and should be synonymized, as proposed by Matthei (1975). The occurrence of several intermediates further complicates this case: morphological links can group these two species with the broadly distributed *B. subaristata* Lam. The extreme forms are quite easy to separate, but the intermediates are difficult to classify and instigate questions about the species circumscriptions.

There is also no consensus as to whether varieties based on measures or discrete characters, like *B. subaristata* var. *interrupta* (Hack. ex Stuck.) Roseng., Arrill. & Izag. or *B. rufa* var. *sparsipilosa* Roseng., Arrill. & Izag., respectively, should be accepted, or whether their variation is merely intra-specific polymorphism or environment plasticity.

Closely related species, like *B. juergensii* Hack., *B. calotheca* (Trin.) Hack., *B. ambigua* Hack. and *B. uniolae* (Nees) Nees ex Steud., also need genetic studies in order to

check whether characters like “lemma width” or “presence of trichomes on palea back” are the best [or are enough] to distinguish the species.

In order to verify whether ISSR can provide good species or infra-specific markers, four groups of related species were investigated, three with taxonomic controversies, and one with two completely distinct species.

The aim of this study was to test whether ISSR can provide markers to help at intra-specific or species-level decisions for taxa belonging to genus *Briza lato sensu*, whether the study of morphological characters alone is insufficient.

The results of this preliminary approach are compared to previous approaches on genus *Briza*, and suggestions for applications for ISSR in the group are provided.

Material and Methods

Taxon sampling

Taxon sampling included representatives of ten *Briza* species, which were analysed in four separate groups, according to their morphological similarities (Table 1). Each group was constituted by seven to 18 accessions, including representatives of morphological extremes, intermediates shapes or even inter and infra-specific categories. Characters such as spikelet colour and size, lemma width, presence of trichomes, or habitat, were most often considered for sampling. Geographical information was collected, although it was not considered for sampling purposes.

Molecular protocols

DNA was extracted from fresh, silica dried or herbarium leaves, following a CTAB method (Doyle & Doyle, 1987) adapted for micro-centrifuge tubes. Total DNA was quantified by agarose gel visualization. Thirteen primers described by Joshi et al. (2000),

Martins, Tenreiro & Oliveira, (2003) and Lin et al. (2005) were tested for ISSR amplification, and nine of them were selected (Table 2). PCR reactions were carried out in a total volume of 25 µl containing 12 µl sterile Milli-Q purified water, 0.2 µl Taq DNA Polymerase (5U/ul), 2.3 µl MgCl₂ (25mM), 2.5 µl 10× buffer, 1 µl primer 10 pmol, 1 µl of 40 mM dNTP mixture (10mM each dNTP), 1 µl DMSO (2%), and 5 µl DNA (total 30-50 ng). PCR amplifications included 40 cycles of 1 min at 94°C, 45 sec at 50°C and 2 min at 72°C, preceded by a period of 5 min at 92°C and completed by a final extension of 5 min at 72°C, for all the primers, except primer F11, whose annealing temperature was 48°C. The ISSR amplification products were stained by ethidium bromide, run until the complete separation of the ladder (100 and 50 bp, PB-L Produtos Bio-Lógicos, Universidad Nacional de Quilmes), approximately 2h30min, at 100V on 1.8% agarose gel, and visualized by UV.

Data analysis

Polymorph bands were scored for presence/absence. Analyses were performed using the NTSYS-pc version 2.10 software (Rohlf, 2000). For each of the four groups, the genetic similarity among individuals was calculated using Jaccard's Similarity Coefficient (J), which takes only shared presence into account. The similarity relationships were portrayed by dendrograms built using the clustering method Unweighted Pair Group Method of Arithmetic Average (UPGMA). Bootstrapping analyses, with 2,000 replicates each, were performed by the Winboot software (Yap & Nelson, 1996), to access the robustness of nodes in the dendrograms, as proposed by Felsenstein (1985).

Table 1: Accessions included in this study.

Species [morphological or habitat type]	Collector number	Voucher	Geographical origin (Country – State – Locality)
Group 1			
<i>B. erecta</i> Lam.	H15056	ICN	Uruguay, Carrasco
<i>B. erecta</i> [intermediate <i>subaristata-erecta</i>]	Li332	ICN 149.322	Brazil, Santa Catarina, Florianópolis
<i>B. subaristata</i> Lam.	Li7	ICN 132.506	Brazil, Rio Grande do Sul, Porto Alegre
<i>B. subaristata</i>	Li8	ICN 134.887	Brazil, Rio Grande do Sul, Porto Alegre
<i>B. subaristata</i>	Li10	ICN 132.509	Brazil, Rio Grande do Sul, Porto Alegre
<i>B. subaristata</i>	Li11	ICN 132.510	Brazil, Rio Grande do Sul, Porto Alegre
<i>B. subaristata</i>	Li12	ICN 132.511	Brazil, Rio Grande do Sul, Porto Alegre
<i>B. subaristata</i>	Li202	ICN 132.542	Brazil, Santa Catarina, Urubici
<i>B. subaristata</i>	H15029	ICN	Brazil
<i>B. subaristata</i>	H15040	ICN 131.398	Brazil, Rio Grande do Sul, Bagé - Minas de Camaquã

<i>B. subaristata</i> var. <i>interrupta</i> (Hack. ex Stuck.) Roseng., Arrill. & Izag.	Li19	ICN 132.518	Brazil, Rio Grande do Sul, São Pedro do Sul
<i>B. macrostachya</i> (J. Presl) Steud.	Li13	ICN 132.512	Brazil, Rio Grande do Sul, São Pedro do Sul
<i>B. macrostachya</i>	Li18	ICN 132.517	Brazil, Rio Grande do Sul, São Pedro do Sul
<i>B. macrostachya</i>	Li54	ICN 132.553	Brazil, Rio Grande do Sul, Piratini
<i>B. macrostachya</i>	Li57	ICN 132.556	Brazil, Rio Grande do Sul, Piratini
<i>B. macrostachya</i>	HI5038	ICN 131.396	Brazil, Rio Grande do Sul, Bagé - Minas de Camaquã
<i>Briza</i> sp. 1	Li201	ICN 132.700	Brazil, Santa Catarina, Urubici
<i>Briza</i> sp. 2	W10768	K, LPB	Bolivia, Chuquisaca, Oropeza
Group 2			
<i>B. rufa</i> var. <i>rufa</i> (J. Presl) Steud. [80 cm tall plant]	Li278	ICN 134.875	Brazil, Rio Grande do Sul, Canguçu
<i>B. rufa</i> var. <i>rufa</i>	Li279	ICN 134.876	Brazil, Rio Grande do Sul, Canguçu
<i>B. rufa</i> var. <i>rufa</i>	Li280	ICN 134.877	Brazil, Rio Grande do Sul,

			Canguçu
<i>B. rufa</i> var. <i>rufa</i> [broad leaves]	Li372	ICN 132.586	Brazil, Rio Grande do Sul, Porto Alegre
<i>B. rufa</i> var. <i>rufa</i> [broad leaves]	Li373	ICN 132.587	Brazil, Rio Grande do Sul, Porto Alegre
<i>B. rufa</i> var. <i>sparsipilosa</i> Roseng., Arrill. & Izag. [open panicle type]	Li281	ICN 134.878	Brazil, Rio Grande do Sul, Canguçu
<i>B. rufa</i> var. <i>sparsipilosa</i>	HI8061	ICN 135.198	Brazil, Rio Grande do Sul, São Francisco de Paula
Group 3			
<i>B. ambigua</i> Hack.	Li190	ICN 132.689	Brazil, Santa Catarina, Urubici
<i>B. ambigua</i>	Li192	ICN 132.691	Brazil, Santa Catarina, Urubici
<i>B. calotheca</i> (Trin.) Hack.	HI8062	ICN 135.199	Brazil, Rio Grande do Sul, São Francisco de Paula
<i>B. calotheca</i>	HI9662	ICN 136.997	Brazil, Minas Gerais, Serra da Caraça
<i>B. calotheca</i> [swamp habitat)	Li115	ICN 132.614	Brazil, Rio Grande do Sul, São Francisco de Paula
<i>B. calotheca</i>	Li308	ICN 134.848	Brazil, Santa Catarina, Bom Jardim da Serra
<i>B. calotheca</i> [shaded	HI5041	ICN	Brazil, Rio Grande do Sul, Bagé

habitat]			– Minas de Camaquã
<i>Briza</i> cf. <i>calothea</i> [large spikelets type]	H18749	ICN 136.600	Brazil, Rio Grande do Sul
<i>B. calothea</i> [small spikelets type]	Li222	ICN 134.816	Brazil, Paraná, Guaratuba
<i>B. juergensii</i> Hack.	Li132	ICN 132.631	Brazil, Rio Grande do Sul, Cambará do Sul
<i>B. juergensii</i> [narrow lemas type]	Li186	ICN 132.685	Brazil, Santa Catarina, Urubici
<i>B. juergensii</i> [small spikelets type]	H18713	ICN 136.595	Brazil, Santa Catarina, São Joaquim
<i>B. uniolae</i> (Nees) Nees ex Steud.	Li242	ICN 134.839	Brazil, Paraná, Piraí do Sul
<i>B. uniloe</i> [dwarf type]	LFL200	ICN 149.155	Brazil, Rio Grande do Sul, Osório
<i>B. uniolae</i>	H15019	ICN 131.378	Brazil, Rio Grande do Sul, São Lourenço - Pelotas
Group 4			
<i>B. monandra</i> (Hack.) Pilg.	Sb30397	LPB	Bolívia, La Paz, P.D. Murillo, Unduavi
<i>B. monandra</i> [small spikelets type]	Ib1356	ICN 132.576	Brazil, Santa Catarina, Urubici

<i>B. bidentata</i> Roseng., Arrill. & Izag.	Li55	ICN 132.554	Brazil, Rio Grande do Sul, Piratini
<i>B. bidentata</i>	Li106	ICN 132.605	Brazil, Rio Grande do Sul, Caçapava do Sul
<i>B. bidentata</i>	Li317	ICN 149.160	Brazil, Santa Catarina, Urubici
<i>B. bidentata</i>	Li175	ICN 132.674	Brazil, Santa Catarina, São Joaquim
<i>B. bidentata</i>	Soreng <i>s.n.</i> (BS)	MO	Brazil, Rio Grande do Sul, Caxias do Sul

Table 2: Primers included in the analyses.

Primer code:	Primer sequence:	Described by:
P1	(AC) ₈ T	Lin et al., 2005 (as 25)
P2	(GA) ₈ T	Joshi et al., 2000 (as 810)
P3	(CTC) ₄ RC	Poulin, Weller & Sakai, 2005 (as n. 15)
P4	(CT) ₈ G	Joshi et al., 2000 (as 815)
F3	(AG) ₈ YC	Joshi et al., 2000 (as 835)
F4	(GA) ₈ YC	Joshi et al., 2000 (as 841)
F7	(GT) ₈ A	Joshi et al., 2000 (as 819)
F11	(GACA) ₄	Lin et al., 2005 (as 73)
F12	(GTGC) ₄	Martins, Tenreiro & Oliveira, 2003 (as IS06)

Results and Discussion

The level of polymorphism

Considering the four groups, nine primers produced an average of 89 DNA bands, 93.5% of which were found to be polymorphic. Most primers produced only polymorphic bands, but some monomorphic bands were found in all groups. The group with the highest number of monomorphic bands was group 2, with 11 bands (14.1% of the matrix). Ten bands were produced per primer on average. The primer with the smallest number of bands was F12 (average of eight bands per group), and the highest number of bands was obtained with the primer P4, with an average of 23 bands per group.

The Jaccard similarity index ranged from 0.1, between *Briza bidentata* (BS) and *B. monandra* (Sb30397), to 0.7442, between *B. macrostachya* (Hw5038) and *B. erecta* (Hw5056). Low levels of similarity were found in all groups. The absence of a similarity index of 1.0 indicates that no clone or repeated germoplasm was included, and all accessions represent plants with distinct fingerprints. The limits for the acceptance of the clusters were established by the similarity average inside the group (see values of the averages for each group in Figures 1 to 4).

In Group 1, the two new species are very distinct from the remaining taxa, being grouped with a very low similarity index. The two are distinct taxa, and certainly grouped together due to the absence of other more closely related accessions. All other accessions appeared clustered, without good definition for species. Although the grouping between Cluster A and Cluster B is under the similarity average for the group, it is supported by bootstrap. The high similarity among accessions of *Briza macrostachya* and the accession of *B. erecta* is also supported by bootstrap.

In Group 2, the two varieties were perfectly separated, and variety *sparsipilosa* grouped with the highest similarity level (Cluster B), in spite of its different geographical origins. The main clusters (A and B) represent the two varieties, and the secondary clusters follow a geographical order (AI: Canguçu; AII: Porto Alegre).

In Group 3, it was not possible to separate any species in a single cluster. The similarity indexes were the lowest. This may be due in part to the high number of species included at the same group, and the low number of bands (76), proportionally to the number of taxa (four species). Only three clusters present bootstrap support, which indicates that the inclusion or exclusion of a few bands could change the clustering order dramatically.

Group 4, included to check the potential of ISSR as species-marker, presented low similarity indexes like other groups, and perfectly clustered the two accessions of *Briza monandra* (Cluster A) which were collected in disjunctive areas, so that a great genetic divergence was already expected. However, the accessions of *B. bidentata* were only partially grouped (Cluster B), and bootstrap values higher than 50 were obtained only for three clusters. A geographic separation was found within Cluster B (Cluster BI= Rio Grande do Sul State; Cluster BII= Santa Catarina State).

Low similarity levels were already expected, due to two main factors: 1) The ISSR are one of the most polymorphic molecular markers; 2) The South American *Briza* species are autogamous, mainly cleistogamous, so that a low intra-population variability and a high inter-population variability were expected. Although a high level of polymorphism was not a surprise, the use of ISSR as a species marker was disappointing: no bands were exclusive to a particular species, and the similarity indexes were lower than those usually published for species-complexes. Their use as phylogenetic markers for *Briza*

should be avoided, since a minimum monomorphism (20%) across all taxa is required to consider the markers potentially homologous to such studies (Bussell, Waycott & Chappill, 2005). Fortunately, the ISSR helped to solve some critical questions:

1) Should the two varieties of *Briza rufa* be accepted? Yes. The ISSR data obtained support the recognition of two genetically distinct groups, which correspond to the two varieties sampled. This is also in agreement with other approaches based on flavonoid variation, pollen and satellite chromosomes position (Winge et al., 1984).

2) Are the new species genetically distinguishable from their morphologically related species? Yes. The accessions sampled were absolutely distinct from the remaining accessions, thus supporting their acceptance as new species, in addition to the morphological findings.

3) Did the ISSR help to distinguish accessions of *Briza ambigua*, *B. calotheca*, *B. juergensii* and *B. uniolae*? No. Although there was a trend in grouping according to species, the separation was weak. The suggestion is to improve both plant and ISSR sampling, and to proceed with future analyses in pairs, instead of large species-complexes. For instance: *B. ambigua* X *B. calotheca*, *B. calotheca* X *B. juergensii* and *B. calotheca* X *B. uniolae*.

4) Did the ISSR help to find a good circumscription for the *Briza macrostachya* - *B. erecta* - *B. subaristata* complex? Yes. It is clear that *B. erecta* is extremely close to *B. macrostachya*, a relation supported by bootstrap. This is in agreement with the opinion of Matthei (1975), who considered these two species synonymous. But the different accessions of *B. subaristata* are widespread over the clusters, producing the same impression caused by morphological analysis – the impression that not only *B. macrostachya* and *B. erecta* should be synonymized, but all three, *B. erecta* and *B.*

macrostachya being considered simply an intra-specific polymorphism. This idea may appear extreme at first sight, especially when the Uruguayan material of *B. erecta* is analyzed, but when the Brazilian material is studied it is perfectly acceptable. Different spikelet sizes are commonly found in the same plant. And the material usually identified as *B. erecta* is collected mostly in sandy soils. In the case of this complex, a study observing the morphology under different soil conditions and in different plant generations would be useful to check the influence of the environment on the colour and size of the spikelets. If the three species are accepted as distinct taxonomic entities, it is necessary to consider hybridization among the species, due to the intermediates. The results obtained for this complex contradict the results of Winge et al. (1984), which allowed the differentiation of the three species, and influenced the circumscription published by Longhi-Wagner (1987). Although their sampling was broader (average of ten per species), it is important to emphasize that some of the markers utilized (morphologic, isoesterases) are more affected by environmental conditions – which could be the main cause of the distinct phenotypes – than the ISSR markers. Future works with ISSR should include a broader sampling of these three species.

Finally, are ISSR useful to solve taxonomic controversies in genus *Briza*? Yes, but important limits must be kept in mind:

a- The ISSR are more effective as markers for polymorphism than for conservative features. Therefore the population sampling must be huge and the questions to be answered have to be separately attacked. The study in pairs of closely related species appears to be appropriate.

b- Although the ISSRs were effective to group accessions of disjunct distribution, the sampling for broadly distributed species has to be proportionally broader, to avoid clustering of distinct entities due to a common high level of divergence.

Acknowledgments

We thank the researchers who kindly contributed plant samples to this molecular survey (Robert Soreng, Stephan Beck and Ilsi Boldrini), the financial support provided by CNPq and FAPERGS, the PhD fellowship provided by CNPq and the facilities provided by the Departamento de Genética at UFRGS-Federal University of Rio Grande do Sul. We also thank Gustavo Agostine and Fernanda Cidade, for their invaluable help with the technique.

Figures

Figure 1: UPGMA dendrogram of individuals from group 1. The cluster is based on the Jaccard similarity index. Vertical line marks the average of similarity indexes. Numbers below branches correspond to bootstrap values >50.

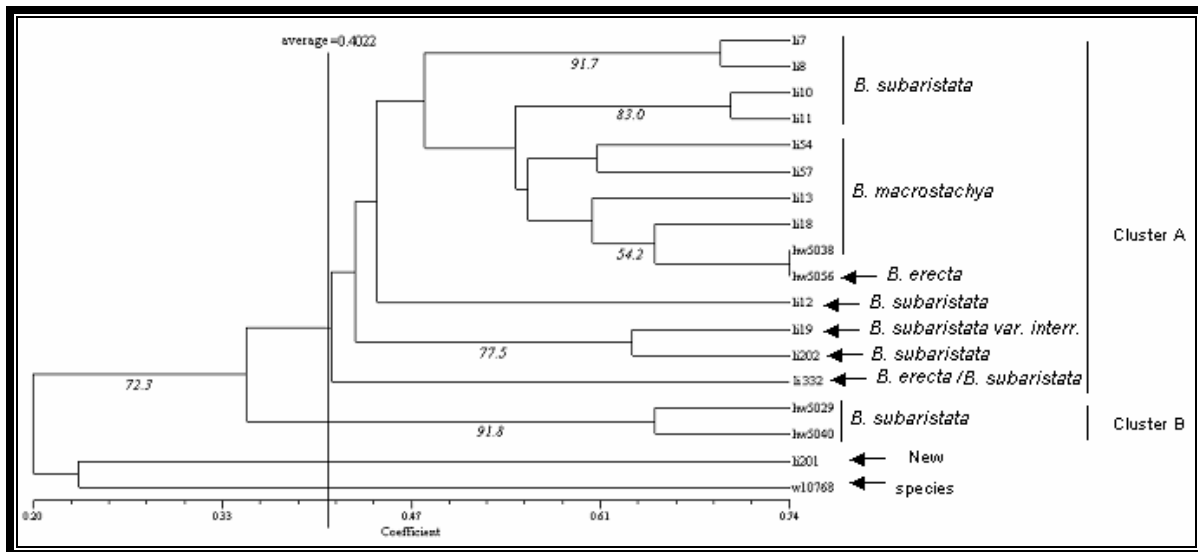


Figure 2: UPGMA dendrogram of individuals from group 2. The cluster is based on the Jaccard similarity index. Vertical line marks the average of similarity indexes. Numbers below branches correspond to bootstrap values >50.

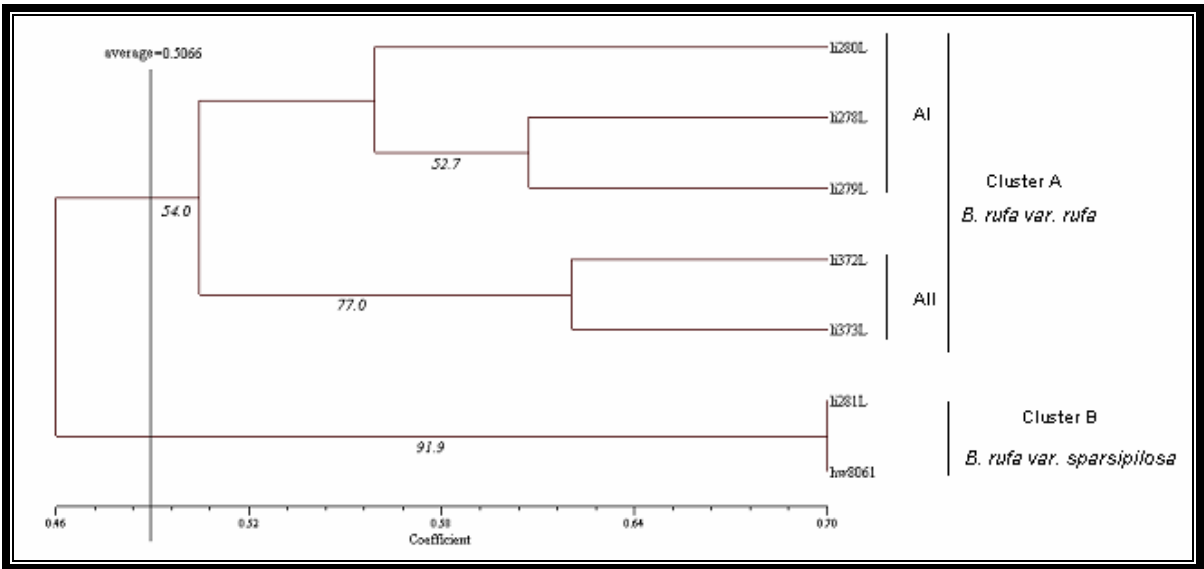


Figure 3: UPGMA dendrogram of individuals from group 3. The cluster is based on the Jaccard similarity index. Vertical line marks the average of similarity indexes. Numbers below branches correspond to bootstrap values >50.

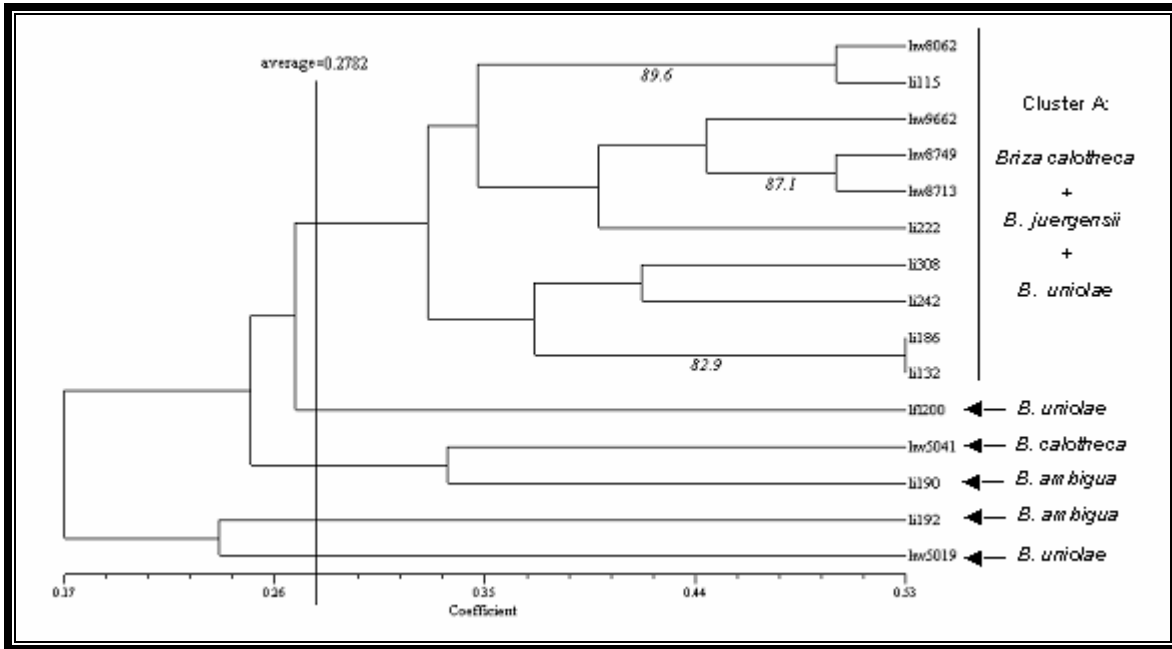
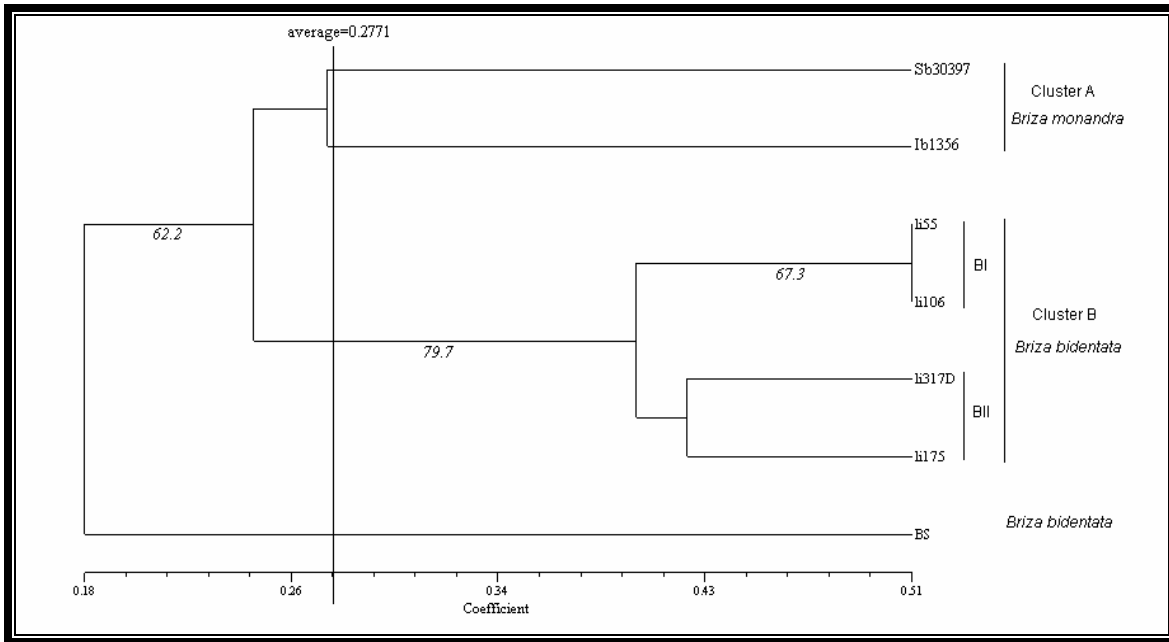


Figure 4: UPGMA dendrogram of individuals from group 4. The cluster is based on the Jaccard similarity index. Vertical line marks the average of similarity indexes. Numbers below branches correspond to bootstrap values >50.



References

- Alexander, J., A. Liston & S. Popovich, 2004. Genetic diversity of the narrow endemic *Astragalus oniciformis* (Fabaceae). *Am. J. Bot.* 91: 2004-2012.
- Ayres, D.R. & D.R. Strong, 2001. Origin and genetic diversity of *Spartina anglica* (Poaceae) using nuclear DNA markers. *Am. J. Bot.* 88(10): 1863-1867.
- Blair, M.W., O. Panaud & S.R. McCouch, 1999. Inter-simple sequence repeat (ISSR) amplification for analysis of microsatellites motif frequency and fingerprinting in rice (*Oryza sativa* L.). *Theor. Appl. Genet.* 98: 780-792.
- Bussell, J.D., M. Waycott & J.A. Chappill, 2005. Arbitrarily amplified DNA markers as characters for phylogenetic inference. *Perspectives in Plant Ecology, Evolution and Systematics* 7: 3-26.
- Camacho, F.J. & A. Liston, 2001. Population structure and genetic diversity of *Botrychium pumicola* (Ophioglossaceae) based on Inter-Simple Sequence Repeats (ISSR). *Am. J. Bot.* 88(6): 1065-1070.
- Dinelli, G., 2004. Characterization of Italian populations of *Lolium* spp. Resistant and susceptible to diclofop by inter simple sequence repeat. *Weed Sci.* 52:554-563.
- Doyle, J.D. & J.L. Doyle, 1987. A rapid DNA isolation procedure for small quantities of fresh leaf tissue. *Phytochem. Bull* 19: 11-15.
- Esselman, E.J., L. Jianqiang, D.J. Crawford, J.L. Windus & A.D. Wolfe, 1999. Clonal diversity in the rare *Calamagrostis porteri* ssp. *insperata* (Poaceae): comparative results for allozymes and random amplified polymorphic DNA (RAPD) and intersimple sequence repeat (ISSR) markers. *Molecular Ecology* 8: 443-451.
- Felsenstein, J., 1985. Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* 39: 783-791.

- Joshi, S.P., V.S. Gupta, R.K. Aggarwal, P.K. Ranjekar & D.S. Brar, 2000. Genetic diversity and phylogenetic relationship as revealed by Inter Simple Sequence Repeat (ISSR) polymorphism in the genus *Oryza*. *Theor. Appl. Genet.* 100: 1311-1320.
- Li, A. & S. Ge, 2001. Genetic Variation and Clonal Diversity of *Psammochloa villosa* (Poaceae) Detected by ISSR Markers. *Annals of Botany* 87: 585-590.
- Lin, W.X., H.Q. He, X.X. Chen, J. Xiona, B.Q. Sona, Y.Y. Liang & K.J. Liang, 2005. Use of ISSR molecular marker approach to estimate genetic diversity in rice and barley allelopathy. Proceedings of the 4th World Congress on Allelopathy, eds JDI Harper, M An, H Wu and JH Kent, Charles Sturt University, Wagga Wagga, NSW, Australia. August 2005. International Allelopathy Society.
- Longhi-Wagner, H.M., 1987. Flora Ilustrada do Rio Grande do Sul, Fasc. 17. Gramineae. Tribo Poeae. *B. Inst. Bioc.* 41, 1-191.
- Martins, M., R. Tenreiro & M.M. Oliveira, 2003. Genetic relatedness of Portuguese almond cultivars assessed by RAPD and ISSR markers. *Plant Cell Rep.* 22: 71-78.
- Matthei, O., 1975. Der Briza-Komplex in Südamerika: *Briza*, *Calotheca*, *Chascolytrum*, *Poidium* (Gramineae). *Willdenowia* 8: 7-168.
- Mattioni, C., M. Casasoli, M. Gonzalez, R. Ipinza & F. Villani, 2002. Comparison of ISSR and RAPD markers to characterize three Chilean *Nothofagus* species. *Theor. Appl. Genet.* 104: 1064-1070.
- Poulin, J., S.G. Weller & A.K. Sakai, 2005. Genetic diversity does not affect the invasiveness of fountain grass (*Pennisetum setaceum*) in Arizona, California and Hawaii. *Diversity Distrib.* 11: 241-247.

- Qian, W., S. Ge & D-Y Hong, 2001 Genetic variation within and among populations of a wild rice *Oryza granulata* from China detected by RAPD and ISSR markers. *Theor. Appl. Genet.* 102: 440–449.
- Souza, V. Q., A.S. Pereira, M.M. Kopp, J.L.M. Coimbra, F.I.F. Carvalho, V.K. Luz & A.C. Oliveira, 2005. Dissimilaridade genética em mutantes de aveia tolerantes e sensíveis a ácidos orgânicos. *Bragantia* 64(4): 569-575.
- Rohlf, F.J., 2000. NTSYS-pc: numerical taxonomy and multivariate analysis system, version 2.10 New York: Exeter Software.
- Vanderpoorten, A., L. Hedenäs & A-L Jacquemart, 2003. Differentiation in DNA fingerprinting and morphology among species of the pleurocarpous moss genus, *Rhytidiadelphus* (Hylocomiaceae). *Taxon* 52: 229-236.
- Winge, H., Hickenbick, M.C.M, Longhi-Wagner, H.M., Torres de Lemos, C., Schifino, M.T., Fonseca-Born, I., Moure, J.S. & V. Leyser da Rosa. 1984. Sistemática e evolução das espécies sul-americanas do Complexo *Briza* (Gramineae). In: Colóquio Sobre Citogenética e Evolução de Plantas, 1. Sociedade Brasileira de Genética, Piracicaba, pp. 185-203.
- Wróblewska, A., E. Brzosko, B. Czarnecka & J. Nowosielski, 2003. High levels of genetic diversity in populations of *Iris aphylla* L. (Iridaceae), an endangered species in Poland. *Botanical Journal of the Linnean Society* 142: 65-72.
- Wu, C., Z. Cheng, X. Huang, S. Yin, K. Cao & C. Sun, 2004. Genetic diversity among and within populations of *Oryza granulata* from Yunnan of China revealed by RAPD and ISSR markers: implications for conservation of endangered species. *Plant Science* 167: 35-42.

Yap, I.V. & R.J. Nelson, 1996. Winboot: a program for performing bootstrap analysis of binary data to determinate the confidence limits of UPGMA-based dendrograms.

Manila: IRRI. 22p.

Zietkiewicz, E., Rafalski, A. & D. Labuda. 1994. Genome fingerprinting by simple sequence repeat (SSR)-anchored polymerase chain reaction amplification. *Genomics* 20: 176-183.

Capítulo III

Three new taxa of *Chascolytrum* (Poaceae, Pooideae, Poeae) from South America

ABSTRACT. Two new species and one new variety of *Chascolytrum* (Poaceae) are described and illustrated. *Chascolytrum latifolium* and *C. juergensii* var. *angustilemma* occur in the Southern Brazilian highlands, while *C. altiplanense* occurs in the Bolivian highlands. *Chascolytrum latifolium* is related to *C. subaristatum* Lam. but differs mainly by the wider leaf blades and by some characteristics of the palea. *Chascolytrum altiplanense* is related to *C. paleapliliferum* and *C. subaristatum*, the main differences being the flattened trichomes at the lemma base of the former and the shape of the spikelets. Specimens of all these new taxa were included in a parallel molecular analysis, resulting genetically divergent from the closest morphologically related species.

Key words: Poaceae, Pooideae, Poeae, *Chascolytrum*, South America.

According to Matthei (1975), *Chascolytrum* Desv. (Poaceae-Pooideae-Poeae) includes six South American species related to the Eurasian genus *Briza* L. Before Matthei (1975), *Chascolytrum* was considered as a genus (Desvaux, 1810), as a section (e.g. Bentham & Hooker, 1883), or as a subgenus of *Briza* (e.g. Parodi, 1920), so that its circumscription was very controversial. A phylogenetic molecular analysis was performed (Essi, Souza-Chies and Longhi-Wagner, in preparation), resulting in a broader circumscription for the genus *Chascolytrum*, including, among others, the three new taxa described here.

Chascolytrum altiplanense Essi, Longhi-Wagner & Souza-Chies sp. nov. TYPE: Bolivia.

Chuquisaca: Yamparaez, on the ascent, c 3 km E. of La Cienaga towards, 16 March 1996, *Wood 10841* (holotype, LPB; isotype, K). Figures 1A-D and 5B.

Chascolytrum altiplanense affine *C. subaristato* Lam. et *C. paleopilifero* (Parodi)

Mathei lemmatibus gibbis et alis conspicuis et paleis elliptico-orbicularibus ad orbicularia, coriaceis. Etiam affine *C. subaristato* spiculis cylindricis et *C. paleopilifero* trichomatibus capitatis in dorso palearum, sed a duabus speciebus trichomatibus spatulatis, copiosis in parte basali lemmatum praecipue differt. *Chascolytrum paleopiliferum* etiam spiculis leviter rectangularibus differt.

Plants 45 -- 77 cm high. Basal entrenodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, smooth, margins non-overlapping. Leaf-blades linear, 7 -- 45 cm x 0.8 -- 2.2 mm, flat or convolute, glabrous. Ligule 1.8 -- 2.5 mm, acute.

Panicle contracted, erect, 3 -- 7 cm long. Pedicels smooth. Spikelets 5.5 -- 6 x 4 mm, 5 -- 8-flowered, cylindrical, oblong. Florets imbricate, obscuring the rhachilla. Glumes herbaceous, convex, glabrous, smooth, acute or obtuse, subequal; lower glume 2.3 -- 3 x 0.9 -- 1.5 mm, 5-nerved; upper glume 2.3 -- 3 x 1 -- 1.5 mm, 5-nerved. Lemmas chartaceous to coriaceous, dorsi-ventrally compressed, with a gibbous pale back, back strongly distinct from the broad margins, cordate or not at the base, 7-nerved, midvein non-salient on the back, glabrous or with sparse flattened trichomes on the back, truncate or acute at the apex, mucicous or mucronate, margins with dense flattened trichomes at the

base, inrolled or not at the base, without oil glands; lower lemma 2.5 -- 3.8 x 1.2 -- 2 mm. Paleas elliptic-orbicular to orbicular, coriaceous, with capitate trichomes between the keels, occasionally deciduous at maturity, keels ciliolate at the upper half; lower palea 1.5 -- 1.8 x 1 -- 1.2 mm. Lodicules flabelliform. Stamens 1 -- 2. Caryopsis sub-orbicular, plano-convex. Hilum elliptic to linear-elliptic.

Distribution. South America. Bolivia.

Notes. Many herbarium specimens of *C. altiplanense* have been identified either as *C. subaristatum* or *C. paleopiliferum*. Indeed, *C. altiplanense* is morphologically related to both species, the three of them sharing similar palea and lemma shapes, and coriaceous palea. Because it presents capitate hairs between the palea keels, some of its individuals were misidentified as *C. paleopiliferum*. However, the new species differs by the shape of the spikelets and by the presence of dense, flattened trichomes at the lemma base. The geographic distribution is also peculiar: *C. altiplanense* is restricted to the Bolivian highlands while *C. paleopiliferum* is mainly restricted to the Argentinian highlands. The presence of dense flattened trichomes at the lemma base is also a remarkable difference between *C. altiplanense* and *C. subaristatum*. The latter species is widely distributed in South America, extending to Mexico, in a broader range of habitats. A collection of this new species (*Wood 10768*, LPB) was included in a molecular phylogenetic analyses, which resulted in a genetic distance from the two morphologically closest species, *C. paleopiliferum* and *C. subaristatum* (Essi, Souza-Chies & Longhi-Wagner, in preparation).

Paratypes. **BOLIVIA. Chuquisaca:** on E side of Cerro Chataquila near Punilla (Sucre-Ravelo), 9 April 1995, *Wood 9665* (LPB); Tomina, Lampacillas, c. 30 km S. of Padilla towards Monteagudo, 31 Dec. 1994, *Wood 9068* (LPB, K); Oropeza, c 2 km beyond

sucre airport towards Ravelo, 25 Feb. 1996, *Wood 10768* (LPB, K); Vamporz, Sucre, on a grassy bank on sandstone rocks by a seasonal Watercourse, SE of the town, 17 April 1994, *Wood 8314* (K). **Cochabamba:** Mizgue a 10 km S of Totora towards Aiquile, 19 Dec. 1995, *Wood 9461* (K); Tapacarí: entre Parotani y Challa, 21 Feb. 1979, *Ceballos, Charpin, Casas & Bermejo BO-445* (G); Tiraque, on pass just W of Sacabambilla on old Cochabamba-Santa Cruz road, 2 Feb. 1996, *Wood 10486* (LPB); Prov. Chapare, Parque Tunari, bosque de Polylepsis, 23 Feb. 1991, *Hensen 998* (LPB); 51 km W of Cochabamba, 3100 m Schub Slender, loosely tufted perennial, 25 March 1981, *Renvoize & Cope 4082* (K). **Santa Cruz:** Caballero, at summit of road on Loma Grande, c. 6-8 km W of Comarapa on road to Siberia, 13 March 2002, *Wood 17802* (K); Vallegrande, c. 16 km from Vallegrande on road to Masucuri, 11 Feb. 1996, *Wood 10601* (LPB). Voladerogebirge, 29 Jan. 1928, *Troll 1101* (M, B).

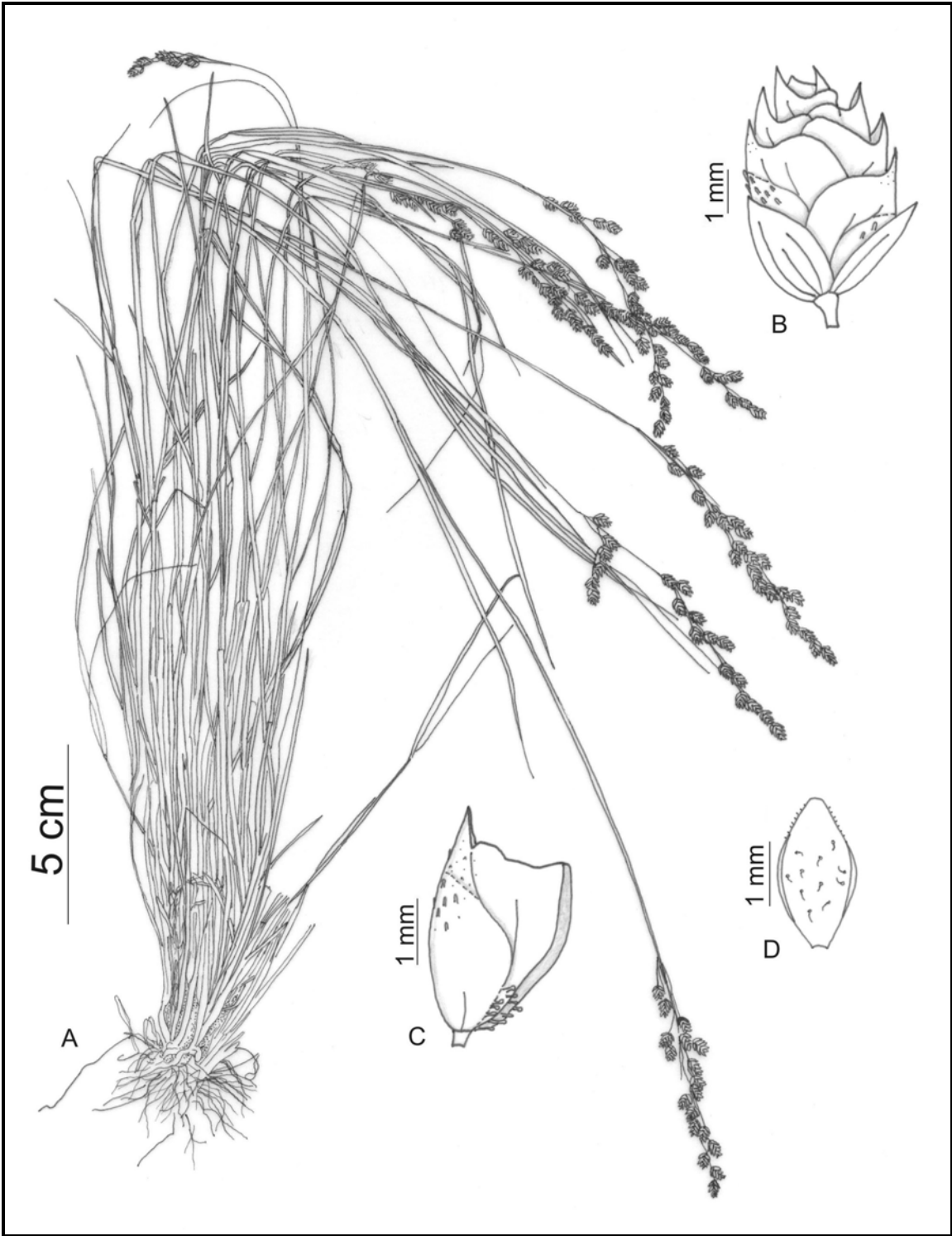


Figure 1. *Chascolytrum altiplanense*. A. Habit. B. Spikelet. C. Lemma (lateral view). D. Palea (dorsal view). (Wood 10841).

Chascolytrum juergensii (Hack.) Essi, Longhi-Wagner & Souza-Chies comb. nov.

Basionym: *Briza juergensii* Hack., Verh. K.K. Zool.-Bot. Ges. Wien 65: 76. 1915.

Synonym: *Poidium juergensii* (Hack.) Matthei (1975). TYPE: Rio Grande do Sul, Pinheiral, Município Rio Pardo 70 m., 1909, C. Jürgens (holotype, W). Figures 2A-B.

Chascolytrum juergensii (Hack.) Essi, Longhi-Wagner & Souza-Chies var.

angustilemma Essi, Longhi-Wagner & Souza-Chies, var. nov. TYPE: Brasil, Rio Grande do Sul, Cambará do Sul, perto do Fortaleza, S29°04'54,6" W050°00'14,4", 1 Dec. 2003, Essi, Guglieri & Hefler, 122 (holotype, ICN; isotypes, MO, K). Figure 3A-B.

A varietate typica lemmatibus angustioribus, sine gibbis et alis lateralibus distinctis differt.

Plants 28 -- 70 cm high. Basal entrenodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, margins non-overlapping. Leaf-blades linear-lanceolate, 7 -- 23 cm x 2 -- 7 mm, flat, glabrous. Ligule 1 -- 2.5 mm, obtuse to truncate.

Panicle open, erect or pendulous, 4.5 -- 15 cm long. Pedicels smooth. Spikelets 5.2 -- 6 x 2 -- 3.2 mm, 4 -- 6-flowered, laterally compressed, elliptic-lanceolate. Florets imbricate, obscuring the rhachilla, or loosely imbricate, rhachilla apert. Glumes herbaceous, naviculate, glabrous, smooth, subequal; lower glume 2.8 -- 3 x 0.5 -- 0.9 mm, 3-nerved; upper glume 3 -- 3.3 x 0.7 -- 1 mm, 3 -- 5-nerved. Lemmas chartaceous or coriaceous, laterally compressed, pale, without a gibbous back, margins narrow, non-distinct from the back, non-chordate at the base, 5-nerved, midvein non-salient on the back, pilose, acute at the apex, muticous, margins glabrous or sparsely pilose, inrolled or not at

the base, without oil glands; lower lemma 3.1 -- 3.9 x 1 -- 1.2 mm. Paleas lanceolate, membranaceous, pilose between the keels, keels ciliate; lower palea 2 -- 2.5 x 0.7 -- 1 mm. Lodicules linear-lanceolate. Stamens 1 -- 3. Caryopsis elliptic, concavo-convex. Hilum elliptic.

Distribution. South America. Brazil, mainly Southern Brazil, in Rio Grande do Sul and Santa Catarina States, with only one record from Minas Gerais State and another from São Paulo State.

Habitat. *Chascolytrum juergensii* var. *angustilemma* is found in high altitude wet grasslands surrounded by *Araucaria angustifolia* Kuntze (Araucariaceae) forests, in Rio Grande do Sul and Santa Catarina, the southernmost States in Brazil. Only two records for other states, in highlands.

Notes. Matthei (1975) mentioned that the Colombian specimens of *Chascolytrum juergensii* present slightly narrower lemmas than the Brazilian material. However, *Chascolytrum juergensii* var. *angustilemma* lemmas are even narrower than the Colombian material. Longhi-Wagner (1987) already emphasized this difference, provisionally naming this new taxon as "*Briza* aff. *juergensii*". The new variety presents a more restricted distribution and its plants are shorter than those of *B. juergensii* var. *juergensii*, with more delicate culms.

A collection of this new variety (Longhi-Wagner 8759, ICN) was included in a molecular phylogenetic analysis of the *Briza* complex (Essi, Souza-Chies & Longhi-Wagner, in preparation), and was found to be separate from the typical variety.

Paratypes: **BRAZIL. Minas Gerais:** Camanducaia, Monte Verde, 22 Jan. 2002, Longhi-Wagner 8009 (ICN). **Rio Grande do Sul:** Bom Jesus, sede, 300 m após entroncamento p/ Vacaria, 29 Nov. 1975, Sampaio, Longhi & Winge 102 (ICN); Bom Jesus, Aparados da

Serra, Dec. 1954, Barreto (BLA); Cambará do Sul - Itaimbezinho, Bela Vista, 1 Dec. 1981, *Sacchet* 28 (ICN); Cambará do Sul, Fortaleza dos Aparados, 28 Nov. 1975, *Longhi, Sampaio & Winge* 315 (ICN); Cambará do Sul, Fortaleza dos Aparados, 28 Nov. 1975, *Longhi, Sampaio & Winge* 325 (ICN); Cambará do Sul, Fortaleza dos Aparados, 28 Nov. 1975, *Longhi, Sampaio & Winge* 328 (ICN); Cambará do Sul, Itaimbezinho, 1 Dec. 1981, *Longhi-Wagner et al.* 957, 959, 960 (ICN); Cambará do Sul, Itaimbezinho, 15 Jan. 1979, *Lemos & Sampaio* 33, 36 (ICN); Cambará do Sul, Itaimbezinho, 28 Nov. 1975. *Sampaio, Longhi & Winge* 58, 59, 61, 62 (ICN); Cambará do Sul, Itaimbezinho, 4 Jan. 1974, *Hickenbick* 92 (ICN); Cambará do Sul, Itaimbezinho, 7 Jan. 1977, *Sampaio, Winge & Arzivenco* 406, 411 (ICN); Cambará do Sul, Itaimbezinho, Bela Vista, 1 Dec. 1981, *Longhi-Wagner et al.* 949 (ICN); Cambará do Sul, Parque Nacional da Serra Geral, S29°03'43.9" W049°57'23.7", 1 Dec. 2003, *Essi, Guglieri & Hefler* 135, 136, 139 (ICN); Cambará do Sul, a caminho do Parque, em basalto com *Sphagnum* sp., na beira da estrada, 17 Nov. 2004, *Essi* 295 (ICN); Itaimbezinho, 100 m p. S. Francisco de Paula, in campestribus subumidis, 20 Feb. 1953, *Rambo* 54022 (B); Itaimbezinho - São Francisco de Paula, 20 Feb. 1953, *Rambo* (PACA); São José dos Ausentes, Monte Negro, S28°37'00.6" W049°47'42.1", 2 Dec. 2003, *Essi, Guglieri & Hefler* 152 (ICN). **Santa Catarina:** Água Doce, 6 Nov. 1971, *Smith, Klein & Hatschbach* 15700 (HBR); Bom Jardim, Curral Falso, 19 Nov. 1959, *Reitz & Klein* 8405 (HBR); Bom Jardim, Serra do Oratório, 9 Dec. 1958, *Reitz & Klein* 7642 (HBR, HB); Caçador, 2 Dec. 1964, *Smith & Klein* 13367 (HBR, MO, S); Campo dos Padres, 1700 m. In subhumidis graminosis, 22 Jan. 1957, *Rambo* (B 1001176910); Lauro Müller, a 25 km pasando la Serra do Rio do Rastro, 7 Dec. 1992, *Rúgolo, Longhi-Wagner, Boechat & Molina* 1478 (MO); Porto União, 27 Oct. 1962, *Reitz & Klein* 13670 (HBR); Porto União, S. Miguel, 29 Dec. 1936, *Rambo* (HBR 2643); Serra

do Oratório, Bom Jardim campo alt. 1400 m, 9 Dec. 1958, *Reitz & Klein 7642 (L)*; **São Paulo**: Campos do Jordão, 9 Dec. 1955, *Longhi-Wagner & Witten 2793 (ICN)*.

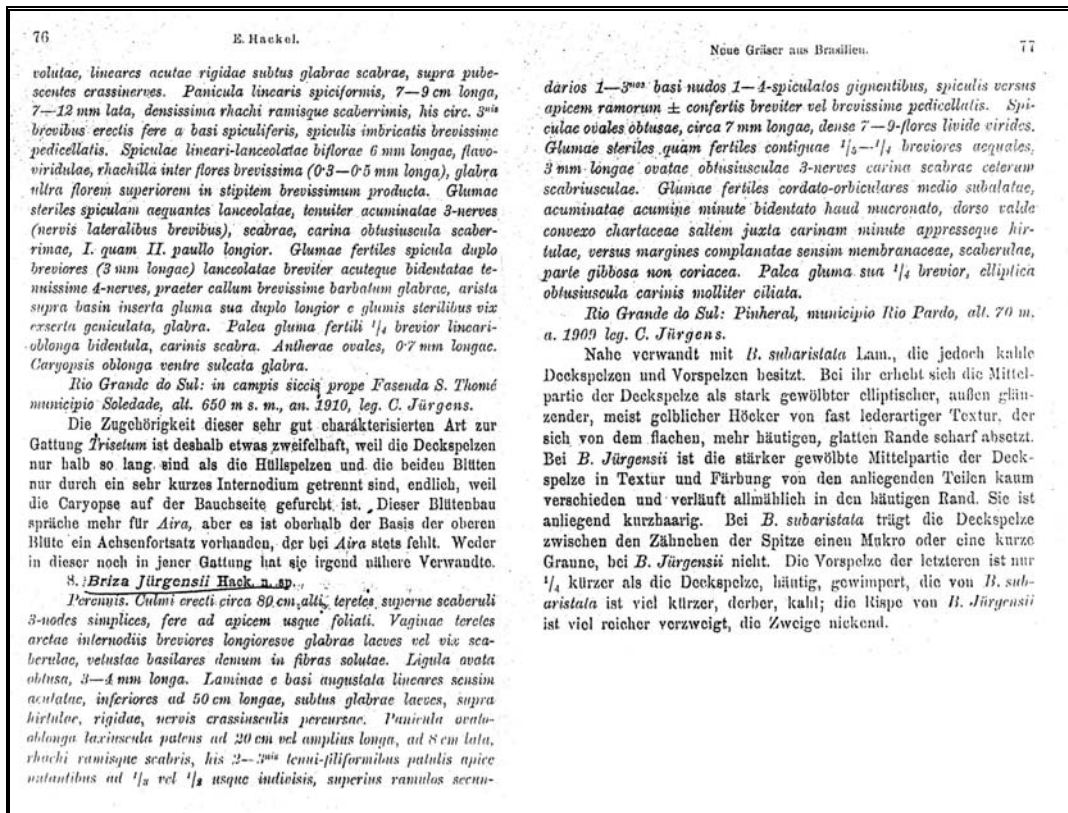


Figure 2. *Briza juergensii*: *Opus princeps*, pages 76 and 77.

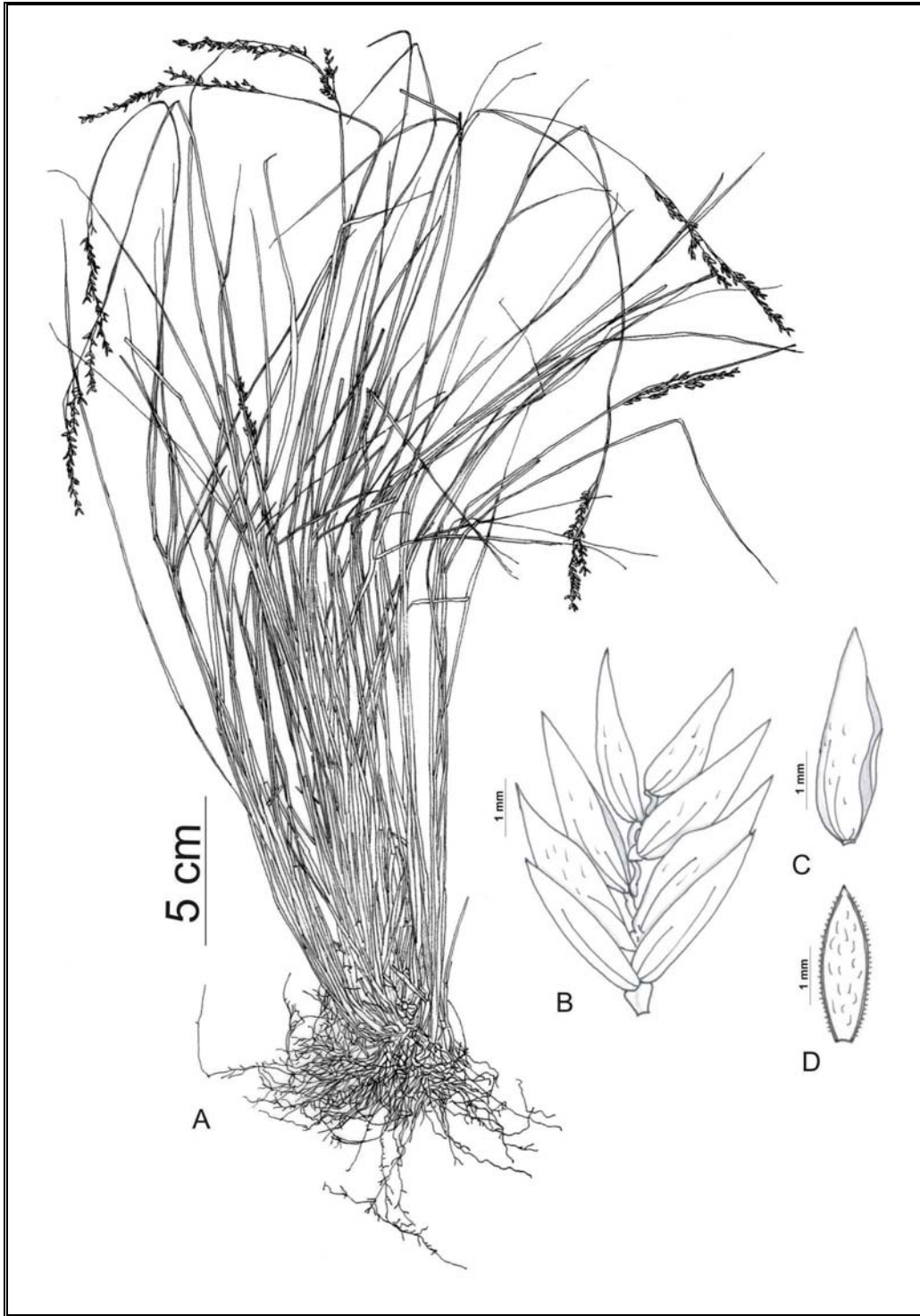


Figure 3. *Chascolytrum juergensii* var. *angustilemma*. A. Habit. B. Spikelet. C. Lemma (lateral view). D. Palea (dorsal view). (Essi, Guglieri & Hefler 122).

Chascolytrum latifolium Essi, Longhi-Wagner & Souza-Chies, sp. nov. TYPE: Brazil.

Santa Catarina: Urubici, Fazenda Arno Philippi. 7 Dec. 2006, *Longhi-Wagner, Zanin & Souza 10228* (holotype, ICN; isotypes, K, FLOR, MO). Figures 4A-D and 5A.

Chascolytrum latifolium affine *C. subaristato* spiculis cylindricis et lemmatibus aristulatis, alis latis, sed paleis elliptico-lanceolatis, membranaceis, trichomatibus longis supra costas ornatis et laminis generaliter latioribus (paleae elliptico-orbiculares ad orbiculares, coriaceae in *C. subaristatum*) praecipue differt.

Plants 27 -- 87 cm high. Basal entrenodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, margins non-overlapping. Leaf-blades linear-lanceolate, 7.5 -- 40 cm x (4) 7 -- 12 mm, flat, glabrous. Ligule 1.5 -- 4 mm, truncate.

Panicle open, pendulous, 9 -- 15 cm long. Pedicels scabrous. Spikelets 5 -- 6 x (1.5) 4 -- 5.1 mm, 4 -- 8-flowered, subcylindrical, oblong. Florets imbricate, obscuring the rachilla. Glumes herbaceous, convex to naviculate, non-cordate at the base, glabrous, smooth, subequal; lower glume 3.2 -- 3.9 x 0.6 -- 1.5 mm, 3-nerved; upper glume 3.2 -- 4 x 1.1 -- 1.5 mm, 3 -- 5-nerved. Lemmas herbaceous, slightly laterally compressed, without a gibbous back, with broad margins non-distinct from the back, non-cordate at the base, 7-nerved, midvein non-salient on the back, glabrous, acute, truncate or bidentate at the apex, mucronate, margins glabrous, smooth or scaberulous at the apex, non-inrolled at the base, without oil glands; lower lemma 3.9 -- 4 x 1.4 -- 2.5 mm. Paleas elliptic-lanceolate, membranaceous, glabrous and smooth between the keels, keels ciliate; lower palea 2.8 -- 3.2 x 1.1 -- 1.7 mm. Lodicules linear. Stamen 1. Caryopsis sub-orbicular, plano-convex. Hilum elliptic.

Habitat. Wet soils with *Sphagnum* spp. in high altitude grasslands, mainly in the Southeast of the State of Santa Catarina, alt 1400-1580 m above MSL, and also in the Northeast region of the state of Rio Grande do Sul, Southern Brazil, at c. 800 m above MSL.

Notes. This new species seems to be a morphological link between the *Chascolytrum* Desv. and the *Poidium* Nees genera, as accepted by Matthei (1975). It presents wide lemmas, like the typical *Chascolytrum* species, but the paleas are elliptic-lanceolate and membranaceous, like the *Poidium* species. The delicate consistency of the lemma is also remarkable. The wide, flat, shiny green leaf blades, and the pendulous, dense inflorescence of this new species, are easy to recognize in the field. A collection of this new species (*Essi 201*, ICN) was included in a phylogenetic molecular analysis (Essi, Longhi-Wagner & Souza-Chies, in preparation), and was found to be closer to *C. juergensii* (genus *Poidium*, according to Matthei, 1975) than to *C. subaristatum* (genus *Chascolytrum*, according to Matthei, 1975).

Paratypes. **BRAZIL. Rio Grande do Sul:** São José dos Ausentes, Monte Negro: na beira de lageado, próximo a mata de Araucária e de ponte de madeira, na descida do Monte Negro, 2 Dec. 2003, *Essi, Guglieri & Hefler 159* (ICN). **Santa Catarina:** Bom Retiro, Campo dos Padres, 16 Dec. 1948, *Reitz 2389* (HBR); Bom Retiro, Campo dos Padres, 18 Dec. 1948, *Reitz 2553* (HBR, S); Bom Retiro, Campo dos Padres, 23 Jan. 1957, *Smith & Reitz 10311* (HBR, RB); Bom Retiro, Campo dos Padres, between Fazenda Campo dos Padres and Fazenda Santo Antônio, 21 Nov. 1956, *Smith & Klein 7804* (HBR, NY); São Joaquim [Urubici] Morro da Igreja, Fazenda Morrinhos, Campestre do Malacara, 22 Jan. 1960, *Mattos 7448* (BLA); São Joaquim, Campestre do Malacara, Faz. de Morrinhos, 22 Jan. 1960, *Mattos 8462* (HAS); Parque Nacional São Joaquim, 2001, *Longhi-Wagner &*

Garcia 7377 (ICN); Urubici, Fazenda Arno Philippi, 7 Dec. 2006, *Longhi-Wagner, Zanin & Souza 10229* (ICN); Urubici, Fazenda Arno Philippi, 7 Dec. 2006, *Longhi-Wagner, Zanin & Souza 10230* (ICN); Urubici, Morro da Igreja: em frente à Cavalgada, próximo do quartel, 4 Dec. 2003, *Essi, Guglieri & Hefler 201* (ICN).

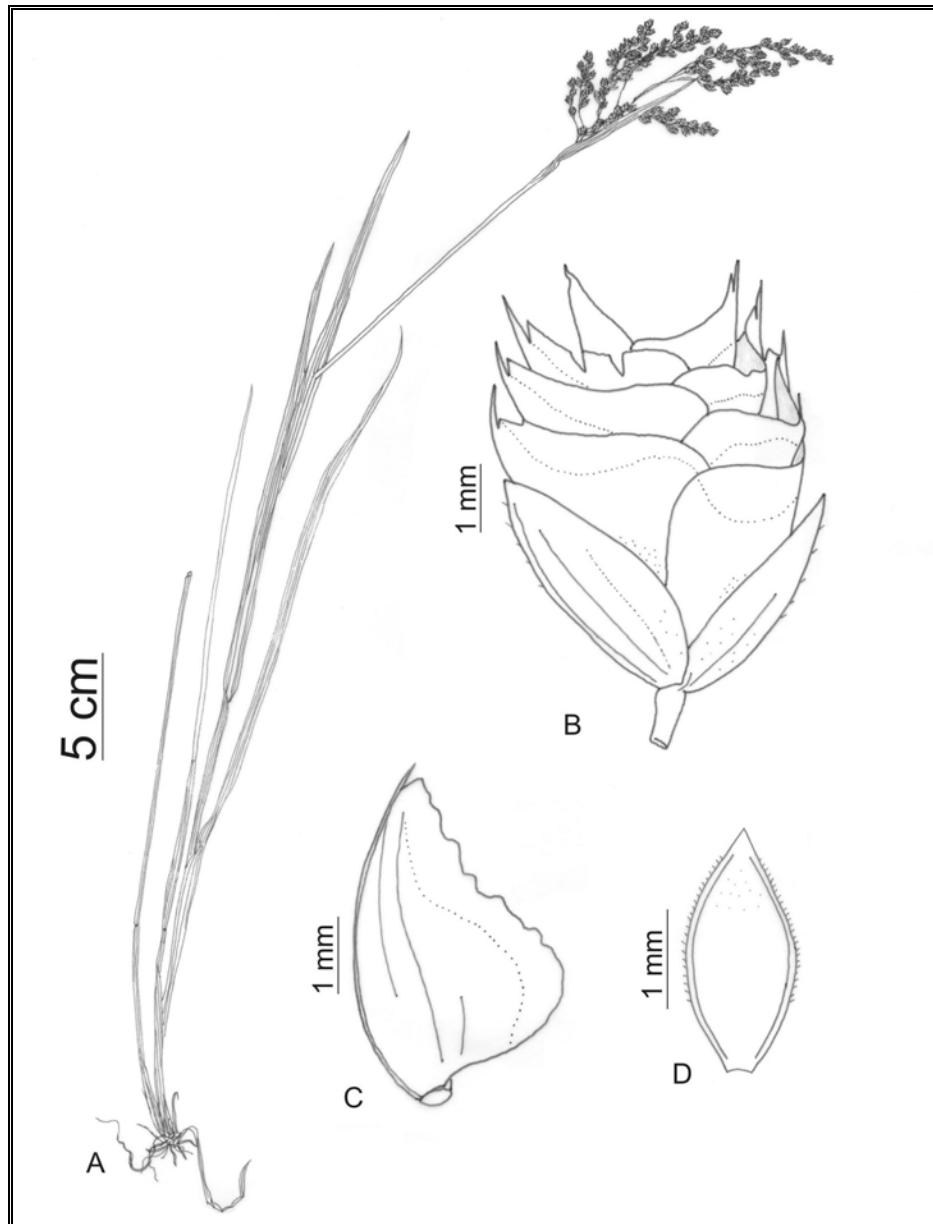


Figure 4: *Chascolytrum latifolium*. A. Habit. B. Spikelet. C. Lemma (lateral view). D. Palea. (*Longhi-Wagner, Zanin & Souza 10228*).

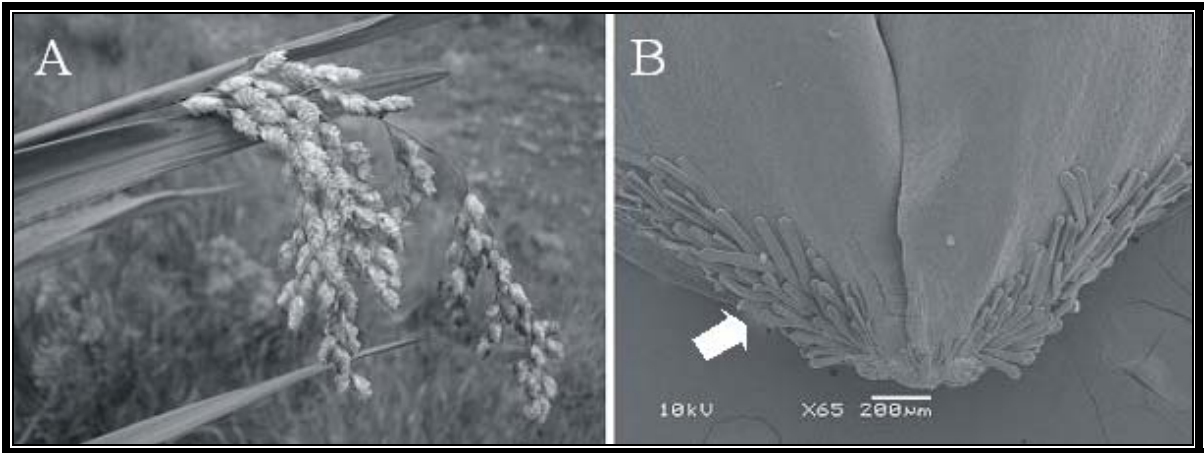


Figure 5: A. *Chascolytrum latifolium*: Panicle. B. *Chascolytrum altiplanense*: Scanning electron micrograph of the lemma base, dorsal view. The arrow indicates the flattened trichomes.

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Literature Cited

Bentham, G., & Hooker, J. D., 1883: *Genera Plantarum*. Bd. 3. – London.

Hackel, E. 1915. Neue Gräser aus Brasilien. *Verh. K.K. Zool.-Bot. Ges. Wien* 65: 76.

Matthei, O. 1975. Der Briza-Komplex in Südamerika: *Briza*, *Calothea*, *Chascolytrum*,
Poidium (Gramineae). *Willdenowia* 8, 7-168.

Parodi, L.R. 1920. Notas sobre las especies de *Briza* de la Flora Argentina. *Rev. Fac. Arg.
Agr. y Vet. B. Aires* 3, 113-137.

Capítulo IV

Taxonomic Revision of the *Briza* Complex: *Briza* and *Chascolytrum* (Poaceae)

Summary. A taxonomic revision of the *Briza* Complex was carried out, including the genera accepted by past authors as subgenera or sections of *Briza sensu lato* (*Briza* L., *Calothea* Desv., *Chascolytrum* Desv., *Lombardochloa* Roseng. & Arrill., *Microbriza* Parodi ex Nicora & Rúgolo and *Poidium* Nees) as well as other related genera (*Erianthecium* Parodi, *Gymnachne* Parodi and *Rhombolytrum* Link). As a result of the morphological analysis presented here, confirmed by molecular data (Essi, Longhi-Wagner & Souza-Chies, in preparation), *Briza* is accepted with four Eurasian species, while a new circumscription for the genus *Chascolytrum* is proposed. This new circumscription includes all the accepted South American species that were previously assigned to the other genera cited above. Descriptions and illustrations of four species of *Briza* and twenty-two species and two varieties of *Chascolytrum* are provided. Keys for genera and species, and geographic distribution maps are also provided.

Key Words. *Briza* Complex, *Briza*, *Chascolytrum*, Poaceae, Poaeae.

Introduction

The genus *Briza* L. (Poaceae: Poeae) is relatively easy to circumscribe. The three Eurasian species described by Linnaeus (1753), *B. maxima* L., *B. media* L. and *B. minor* L. are accepted, but all other species more recently described for the Old World, except *B. humilis* M. Bieb., are considered synonymous. However, the placement of the group of South American species described under *Briza* or other morphologically related genera is controversial.

The first genera described to place South American species related to *Briza* were *Chascolytrum* Desv. and *Calotheca* Desv. (1810). In 1836, Nees described the genus *Poidium*, based on the new species *P. brasiliensis* Nees. Further treatments (Bentham & Hooker, 1883; Ekman, 1913; Parodi, 1920; Rosengurt et al., 1968 - 1970) considered the genus *Briza* in a broader circumscription, describing sections or subgenera to accommodate the known morphological diversity and the new taxa.

In 1975, Matthei published one of the most complete revisions for this group of morphologically related species, which he named *Briza* Complex. In that revision, he recognized the affinities between all the species already treated as *Briza lato sensu*, although he preferred to split the group into four genera: *Briza stricto sensu* (Eurasian species), *Poidium*, *Chascolytrum* and *Calotheca*. After Matthei (1975), new combinations of splitting circumscriptions were proposed. Rosengurt & Arrillaga de Maffei (1979) described the new monoespecific genus *Lombardochloa*, to accommodate *L. rufa* (= *Chascolytrum rufum* J. Presl), a species presenting unusual oil glands on its lemmas, placed by Matthei (1975) in *Poidium*.

Nicora & Rúgolo de Agrasar (1981) proposed four genera for the *Briza* Complex, the genus *Microbriza* being described to place two species of the *Briza* Complex with small spikelets:

M. poaemorpha and *M. brachychaete*. In 1987, Longhi-Wagner considered a *lato sensu* circumscription for the genus *Briza* in the state of Rio Grande do Sul (Brazil), a center of diversity for the *Briza* Complex. In her treatment, some species previously excluded from the complex were included again.

The various circumscription options for the group make it possible to consider a single *lato sensu* genus or even eight genera: *Briza*, *Chascolytrum*, *Calotheca*, *Poidium*, *Rhombolytrum* Link, *Gymnachne* Parodi, *Microbriza* and *Lombardochloa*. The genera *Rhombolytrum* and *Gymnachne* were involved in the complex due to the controversial placement of *B. bidentata*. The latter was excluded from the complex by Matthei (1975), with the suggestion of placement in *Rhombolytrum* or *Gymnachne*, and was placed by Nicora & Rúgolo de Agrasar (1981) in the genus *Rhombolytrum*. Another species excluded from the complex by Nicora & Rúgolo de Agrasar (1981) was *B. ambigua* Hack., which they considered as belonging to the genus *Poa* L.

The first cladistic analysis published about the group was performed by Bayón (1998), based on 32 morphological characters, and resulted in a suggestion of a circumscription very similar to that proposed by Nicora & Rúgolo de Agrasar (1981). Recently, a new phylogenetic analysis was performed, based on DNA sequences (Essi, Longhi-Wagner & Souza-Chies, in preparation), resulting in a totally different phylogeny, which was utilized to support the circumscription presented here. In this work the genus *Briza* appears monophyletic, when circumscribed as proposed by Matthei (1975), with only four species. All the South American species of the complex are placed in a single monophyletic genus, here named *Chascolytrum*. The genus *Calotheca* Desv. was described simultaneously with *Chascolytrum*, published at the same work and at the same page, and is another possible

choice. However, the latter name was preferred in order to minimize the number of new combinations.

In the present work, the Eurasian genus *Briza* and the South American species cited for the *Briza* Complex, treated here as *Chascolytrum lato sensu*, are revised. Twenty-six species (four *Briza*, and twenty-two *Chascolytrum* species) and two varieties are described and illustrated. Geographic distribution maps and keys for genera, species and varieties are also provided. The endemic monotypic genus *Erianthecium* Parodi is included in the newly circumscribed *Chascolytrum*, and several new combinations are proposed.

Materials and Methods

This work was based on field collections, traditional taxonomic revision of herbaria and review of literature. Field collections covered the southernmost Brazilian states. The herbaria revised were: B, BLA, BM, BR, CNPO, CONC, CORD, FLOR, G, GUA, HAS, HB, HBR, ICN, K, L, LINN, LPB, M, MBM, MO, MSB, NY, P, PACA, RB, S, SGO, SMDB, UEC, VIC and W. Electronic pictures provided by BAA, GOET and PR were also examined.

Morphological characters of the vegetative and reproductive structures with taxonomic value were examined. Five exsiccates were selected for the measures, including specimens with extremes of variation. For species with higher morphological variation, 10 specimens were included in the measures.

For the species with a few specimens available, all the examined material is listed. In the case of species with a high number of revised specimens, only selected material is cited.

The complete list is available on request.

The terminology of shape, consistency and indument of the structures followed mainly Stearn (1992). The abbreviation of the author names followed Brummitt & Powell (1992). The types examined were indicated by a “!”, including those analysed through digital images. The types not examined were indicated by an “n.v.” (*non vide*). Types are indicated for all valid names.

Vernacular names were based on literature and on the information on the labels. When available, vernacular names in multiple languages are indicated. Flowering data were based on the information on the labels, on literature, and on field observation.

Data on geographical distribution are based on the material examined. Coordinate-based distribution maps were prepared with the DIVA-GIS ver. 5.2.0.2 software (<http://www.diva-gis.org>). Coordinates obtained during fieldwork or recorded on the labels were used. In most cases, however, the coordinates were not available, and the inclusion on the maps was made based on the coordinates for the headquarters of the town. Coordinates for Brazilian cities were obtained on the IBGE home page (Instituto Brasileiro de Geografia e Estatística – database for cities and villages 1998). Coordinates for cities from other countries were obtained through Google Earth (<http://earth.google.com>) and through the site <http://www.fallingrain.com/world>.

Environmental data were based on information contained on the labels, or observations in field.

The illustrations of the reproductive structures were done using a drawing tube attached to a WILLD M32 stereoscopic microscope, and the habit and inflorescences were obtained by free-hand drawing based on scanned images of the plants or of the exsiccates. The illustrations were covered with Indian ink, digitized in high resolution, and the plates were

built digitally. Reproductive characters were drawn by Liliana Essi, and the habits were drawn by Anelise S. de S. Nunes.

Results and Discussion

Key to genera:

1. Paleas membranaceous, elliptic, ovate to obovate, with glabrous keels, less frequently ciliolate keels (*Briza maxima*). Spikelets laterally compressed. Pedicels usually recurved. Lemmas wide and generally cordate at the base. Plants annual, with intravaginal basal innovations, less frequently perennial with extravaginal innovations (*B. media*).

.....*Briza*

1. Paleas chartaceous to coriaceous, elliptic, elliptic-orbicular to orbicular, glabrous or ciliate on the keels, or membranaceous, than linear-lanceolate, elliptic-lanceolate to lanceolate, ciliate on the keels. Spikelets laterally compressed, cylindrical to globose. Pedicels straight. Lemmas narrow or wide, cordate or not at the base. Plants perennial, with extravaginal basal innovations, rarely intravaginal (*C. brizoides*).

.....*Chascolytrum*

Briza L. (1753: 70); Typus generis: *Briza minor* L.

Tremularia Heister ex Fabricius (1759: 207), nom. illeg. superfl. based on *Briza* L.

Brizochloa Jirásek & Chrték (1966: 39)

Briza sect. *Macrobriza* Tzvelev (1970: 21)

Macrobriza (Tzvelev) Tzvelev (1987: 32)

Annual or perennial, caespitose, rhizomatous or not. Basal internodes of the culms non-thickened. Basal innovations intravaginal, less frequently extravaginal. Leaf-sheaths glabrous, rarely pilose, margins overlapping or not. Leaf-blades linear, flat or conduplicate, glabrous or pilose, smooth or scabrous. Ligule membranaceous, acute to truncate.

Panicle open to contracted, erect or pendulous. Pedicels smooth or scaberulous, recurved, occasionally straight. Spikelets many-flowered, laterally compressed, disarticulating above the glumes, triangular, ovate to ovate-elliptic. Florets imbricate, obscuring the rhachilla. Glumes persistent, herbaceous or chartaceous, convex, cordate at the base, occasionally non-cordate and tapering towards the base, glabrous, smooth or scaberulous, equal to subequal, 3 – 7 (-11)-nerved. Lemmas herbaceous or chartaceous, laterally or dorsiventrally compressed, with or without a gibbous back, back strongly to slightly distinct from the narrow or broad margins, cordate at the base, occasionally non-cordate and tapering towards the base, 5 - 11-nerved, glabrous or pilose on the back, obtuse to acute at the apex, muticous, margins glabrous, smooth or scaberulous, non-inrolled at the base, inrolled or not at the apex, without oil glands. Paleas 2-keeled, elliptic, ovate to obovate, membranaceous, smooth, glabrous or with thick translucent trichomes between the keels, keels glabrous, less frequently ciliolate. Lodicules 2, linear, linear-lanceolate or flabelliform. Stamens 3. Ovary glabrous, styles terminal. Caryopsis sub-orbicular, transverse section plano-convex. Hilum punctiform or elliptic.

Flowering: Spring and Summer.

Chromosome counts: $2n=10$ ($x=5$), $2n=14$ ($x=7$) and $2n=28$ ($x=7$). (Matthei, 1975)

Briza, as understood here, includes four Eurasian species. *Briza maxima*, *B. media* and *B. minor* also occur in other continents as adventitious.

NOTES. 1. The genus *Briza* is easy to identify by the laterally compressed and trembling spikelets, due to the usually curved pedicels. However, *B. humilis*, which is restricted to the Caucasus and Balkan Peninsula, presents erect spikelets, but is easily placed in *Briza* by its membranaceous and totally glabrous paleas. Some species of *Chascolytrum* can also present membranaceous paleas, but they are always pilose, ciliate or ciliolate on the keels.

2. *Briza minor* L. was the earlier choice for lectotype of the genus (Nash in Britton & Brown, 1913), which was followed by Matthei (1975), Nicora & Rúgolo de Agrasar (1981) and Clayton & Renvoize (1986). *Briza media* L. was later chosen as the type by Hitchcock (1923), which was adopted by Rosengurtt et al. (1968), Kerguëlen (1975) and Longhi-Wagner (1987). We adopted the suggestion of Nicolson (1992), accepting the older lectotype, *Briza minor*.

Key for *Briza* species

- 1. Spikelets 10 - 20 mm long. Glumes 7 - 11-nerved. Hilum linear.....*B. maxima*
- 1'. Spikelets 2.2 – 7 mm long. Glumes 3 - 5-nerved. Hilum elliptic or punctiform.....2
- 2. Lemmas non-cordate, tapering towards the base, pilose on the back.....*B. humilis*
- 2'. Lemmas cordate at the base, glabrous or with translucent thick trichomes on the back
- 3. Ligule 3 – 7 mm long, acute. Spikelets triangular. Lemma with broad margins clearly distinct from the gibbous back, apex usually strongly recurved. Palea with translucent thick trichomes between the keels, rarely glabrous. Lemma with translucent thick trichomes on the back, rarely glabrous. Basal innovations intravaginal.....*B. minor*

3'. Ligule 0.5 – 2.5 mm long, truncate. Spikelets ovate. Lemma with broad margins distinct from the slightly gibbous back, apex not recurved. Palea glabrous, rarely with translucent thick trichomes between the keels. Lemma usually glabrous on the back, rarely with translucent thick trichomes. Basal innovations extravaginal.....*B. media*

1. *Briza humilis* M.Bieb. (1808: 66). Type: Tauria (lectotype LE n.v. (designated by Tzvelev, 1983); isolectotypes BM! MO!)

Brizochloa humilis (M.Bieb.) Chrtek & Hadač (1969: 170)

Briza spicata Sibth. & Sm. (1806: 60), nom. illeg. hom., non *Briza spicata* Burm f. (1768) nec *B. spicata* Lam. (1783)

Brizochloa spicata (Sibth. & Sm.) V. Jirásek & Chrtek (1966: 40), nom. illeg.

Annual, without rhizomes, 21 - 40 cm high. Basal innovations intravaginal. Leaf-sheaths glabrous, margins overlapping or not. Leaf-blades 2.4 - 5.5 cm x 0.2 - 0.8 mm, flat or conduplicate, glabrous or pilose, scabrous. Ligule 2.2 - 4 mm long, acute or obtuse. Panicle contracted, erect, 3 - 21 cm long. Pedicels scaberulous, straight. Spikelets 4.2 - 6.5 x 3 - 5 mm, 4 – 9 (-12)-flowered, ovate. Glumes herbaceous, non-cordate and tapering towards the base, scaberulous, equal to subequal; lower glume 2.9 - 3.2 x 1 - 1.8 mm, 3 - 5-nerved; upper glume 2.9 - 3.2 x 1 - 1.8 mm, 3 - 5-nerved. Lemmas herbaceous, dorsiventrally compressed, without a gibbous back, margins non-distinct from the back, non-cordate and tapering towards the base, 5 - 7-nerved, scaberulous, pilose on the back, acute to obtuse at the apex, muticous, margins scaberulous, non-inrolled at the apex; lower lemma 3 - 3.5 x 1.2 - 1.5 mm. Paleas elliptic to obovate, glabrous between the keels, keels glabrous; lower palea 2.8 - 3 x 1.5 - 2 mm. Lodicules n. v. Stamens 3. Hilum punctiform.

Fig. 1.A–D.

DISTRIBUTION. Restricted to the Balkan Peninsula and Caucasus highlands. Map 1.

SELECTED COLLECTIONS. ALBANIA. Janina. Mt. Mikéirali, 16 June 1895, *Baldacci* (BM, K). **Gjinokastrë.** above Zhej, 14 June 1933, *Alston & Sandwith 1678* (BM, K). **BULGARIA.** Tatar Pazardzik, June 1906, *Stribrny* (K, M). Tekir, 21 July 1899, *Stribrny* (BM, K). **CYPRUS.** "Pass" in mountains above Chakistra, 28 April 1962, *Meikle 2742* (K). Perapedhi, by side of Kryos Potamos, 2 May 1962, *Meikle 2806* (K). **GREECE. Nomos Achaias.** M. Panachaikon, Peloponnes, 5 Oct. 1975, *Damboldt* (M). **Nomos Attikis.** Mt. Parnes, 23 Sept. 1971, *Merxmüller 27311* (M). **Nomos Kefallinias.** Ion, Ep. Kraneas, Aenos, 30 July 1971, *Damboldt 204/71* (M). **Nomos Kefallinias.** Ionische Inseln, Insel Kefallinia Omalá: M. Ag. Gerasimou Kulturland um das Ruinendorf Miháta e des Klosters, 24 April 1995, *Hörandl et al. 6487* (W). **IRAQ. Muhafazat Arbil.** Galli Ali Beg, 22 May 1973, *Karim et al. 40905* (K). **Muhafazat as Sulaymaniyah.** Chewa Rash, NE of Rania, 9 May 1959, *Rawi et al. 28505* (K). **Muhafazat Dahuk.** Dohuk, 13 May 1947, *Raur 8966* (K). **LEBANON.** Kamakem, 24 May 1931, *Gombault 942* (P). **PALESTINE.** Hermon, 1 June 1987, *Danin* (K). **SYRIA.** Aintab, May 1884, *Postian* (K). Ouadi-el-Karn, 26 May 1932, *Samuelsson 1739* (BM, MO, K). **TURKEY. Ankara.** Hacikadun valley nr. Kecioren, 11 June 1952, *Davis & Dodds D. 18807* (K). Denizli Region 6 km NE of Tavas on KIZIlhisar road, 15 June 1994, *Nesbitt & Samuel RMN 4568* (K). **Elazig.** Maden, 2 June 1957, *Davis & Hedge 28893* (K). **Isparta.** 52 km NE of Eğridir, on Akşehir road, 17 June 1994, *Nesbitt & Samuel RMN 4603* (K). **Malatya.** Doganschir, 9 June 1960, *Stainton & Henderson 5473* (K). **Manisa.** S slope of Karadağ, 4

km SE of Manisa, 11 June 1994, *Nesbitt & Samuel RMN 4502* (K). **UKRAINE.**

Simferopol, Neusatz, 14 June 1896, *Callier 228* (K, W, M).

HABITAT. Rocky places on mountain slopes and steppes, often in forest.

VERNACULAR NAMES. Giya Rischalot (Kurdistan, Iraq), Spiked Quaking Grass (English, Townsend & Guest, 1968).

USES. Tzvelev (1983) referred *Briza humilis* as a pasture plant.

2. *Briza maxima* L. (1753: 70); Type: hab. in Italia, Lusitania. (lectotype LINN-88.6! (designated by Sherif & Siddiqi, 1988))

Briza grandis Salisb. (1796: 21), nom. illeg. superfl., based on *Briza maxima* L.

Macrobriza maxima (L.) Tzvelev (1993: 91)

Annual, without rhizomes, 38 - 70 cm high. Basal innovations intravaginal. Leaf-sheaths glabrous, margins non-overlapping. Leaf-blades 6.6 – 17 cm x 3 - 4.5 (7) mm, flat, glabrous. Ligule 3.4 - 5.6 mm, acute.

Panicle open, erect or pendulous, 5 - 9 cm long. Pedicels smooth and recurved. Spikelets 10 – 20 (31) x 7 – 14 mm, 9 – 17 (21)-flowered, laterally compressed, ovate-elliptic. Glumes chartaceous, cordate at the base, glabrous, smooth, equal to subequal; lower glume 4.6 - 6.2 x 2.1 - 3.2 mm, 7 (11)-nerved; upper glume 5 - 7 x 2.8 - 3.8 mm, 9 - 11-nerved. Lemmas chartaceous, laterally compressed, with a slightly distinct gibbous back, distinct from the broad margins, cordate at the base, 7 - 11-nerved, glabrous or pilose, obtuse at the apex,

muticous, margins glabrous or pilose, non-inrolled at the apex; lower lemma 6.2 - 7 x 3.4 - 4.2 mm. Paleas ovate to obovate, glabrous between the keels, keels glabrous to ciliate; lower palea 3.4 - 4 x 2 - 2.7 mm. Lodicules linear. Stamens 3. Hilum linear.

Fig. 2.A-D.

DISTRIBUTION. Temperate regions. Native in Eurasia, adventitious in America, Africa and Australasia. Map 2.

SELECTED COLLECTIONS. ALGERIA. Moissons de Ehéragas a 12 Km d' Alger, 23 May 1859, *Laltemant* (W). **ARGENTINA.** Buenos Aires. Buenos Aires, Partido Tandil, 8 Jan. 1908, *Stuckert 18433* (B). **AUSTRALIA.** South Australia, Adelaide, Region 11, southern Lofty, 9 Nov. 1983, *Whibley 8770* (RB). **AUSTRIA.** An grasigin Plätzen bei Abbazia im Österr., Küstenland; Sandstein, May 1899, *Richter 41* (L). **AZORES.** Ilha de São Miguel, 27 May 1999, *Vitek 99-103* (W). **BRAZIL.** Rio Grande do Sul, Pelotas, I.A.S., 22 Nov. 1954, *Sacco 245* (HBR, ICN, FLOR). **BULGARIA.** Southern coastal area of the Black Sea, ca. 6.5 km SSW of Sinemorets, ca. 3.5 km. NNW of Rejovo, 9 June 1998, *Uzunow & Vitek 98 -641* (W). **CANARY ISLANDS.** Tenerife Island, Bajamar, 25 Jan. 1973, *Aldridge 432A* (BM). **CEYLON.** Haputale, 18 April 1969, *Kostermans* (K). **CHILE.** IX Region, Prov. Malleco, Camino desde Cruce Puente Miraflores a Nahuelve, sector Santa Julia, 7 Dec. 1997, *Baeza & Kottirsch 886* (CONC). **COLOMBIA.** Dep. Cauca, Between Paspamba and Chapa, 10 July 1944, *Core 77* (S). **ECUADOR.** Prov. Azuay, market in Cuenca, 1 July 1943, *Steyermark* (S). **ENGLAND.** London, Borough of Wandsworth, 29 May 1995, *Vickery 861* (BM). **FRANCE.** Gard Audas near Le Vigan, 22 Aug. 1988, *Feltwell 12* (K). **GERMANY.** Bayern Schwaben, Bahnhof Lindau – Reutin, 9 June 1965, *Dörr* (M). **GREECE.** Kriti, 5 km E Ziros, near Kalo-Morion, 24 May 1974, *Ebbels et al.*

19/3 (K). **HONDURAS.** Bosque de nubes, floresta de Montaña Grande, entre Santa Lucía y Valle de Angeles, Dep. Morazán, 26 Sept. 1950, *Molina 3319* (BM). **HUNGARY.** In declivibus Graminosis ariolis littoralibus ad Cantridam, 5 April 1902, *Degent 69* (W).

HAWAII. Kawai, Waimea Distr., Waimea Canyon State Park, 28 April 1997, *Flynn & Lorence 6144* (K). **INDIA.** Madras Palni (Pulney) Hills, Loc. Dingul, Kodai-Berijam road, Distr. Kodaikanal, 29 July 1986, *Matthew & Charles* (K). **ISRAEL.** Jerusalem, Avizohas Steet, c. 500 m west of Giv'at Ram campus, 5 May 1994, *Nesbitt 4207* (K). **ITALY.** Reg. Campania, Prov. Napoli E slopes of Vesuvio, 7 July 1983, *Akeroyd et al. 3295* (BM).

JAMAICA. Parish of St. Andrew, headwater slopes of Clyde River, 4 June 1991, *Bellingham 1482* (BM). **JAPAN.** Yokohama city, Kanagawa Pref., 5 June 1915, *Makino* (M). **KENYA.** Nairobi, Timboroa, May 1958, *Ivans* (K). **LEBANON.** Near Trifoli, 7 May 1945, *Norris* (BM). **LYBIA.** Corradini, 6 May 1953, *Guichard KG/Lib/544* (BM).

MOROCCO. Chefchaouene, Montes de Jebala, entre Derdara y Ksar-el-Kebir, Akarate, 31 May 1995, *Ferguson et al. 6322/95* (K). **NEW ZEALAND.** North Island, Piha, West of Auckland, Nov. 1955, *Lovis 535* (BM). **PORTUGAL.** Beira Baixa, Serra da Estrela, NW Covilha, 10 July 1974, *Leadlay & Petty 333* (MO). **SOUTH AFRICA.** Eastern Cape, 9 Nov. 1986, *Phillipson 1525* (K). **SPAIN.** Prov. Málaga Alóka, sierra above the town, 14 April 1969, *Gibbs et al.* (BM). **SYRIA.** Antioch Lake, 17 May 1933, *Meinertzhagen* (BM).

TURKEY. Prov. Antalya, Kumköy, between Antalya & Serik, 6 April 1956, *Davis & Polunin 25703* (BM). **URUGUAY.** Montevideo, Nov. 1875, *Arechavaleta 5585* (L).

UNITED STATES OF AMERICA. California, Siskiyou County, 20 June 1905, *Heller 8075* (P).

HABITAT. Open places, adventitious near roads, inhabited areas and dry meadows.

VERNACULAR NAMES. Quaking-grass (English), Shivery Grass (English, Australia), Guizo-de-cascavel (Portuguese, Brazil), Guizo-de-homem (Portuguese, Portugal).

USES. Ornamental.

3. *Briza media* L. (1753: 70); Type: Hab. in Europae partis siciccoribus. (lectotype LINN-88.5! (designated by Meikle, 1985)).

Poa media (L.) Cav. (1803: 28)

Briza tremula Lam. (1778: 587). nom. illeg. superfl., based on *Briza media* L.

Briza elatior Sibth. & Sm. (1806: 57) Type: In monte Athone, *D. Hawkins 4* n.v.

Briza media f. *caucasica* Marcow. (1902: 97). Type: Auf Alpenwiesen in der Landschaft Ossetien in Kaukasus (Russland). 24. July 1899, *B. Marcowicz* (holotype LE n.v; isotypes BM!, MO!, K!)

Briza marcowiczii Woron. ex Fedtsch. (1915: 74) Lectotype: Auf Alpenwiesen in der Landschaft Ossetien in Kaukasus (Russland). 24. July 1899, *B. Marcowicz* (lectotype LE n.v (designated by Tsvelev, 1983); isolectotypes BM!, MO!, K!)

Briza australis Prok. (1954: 42) Type: "Krymskij gos. zapovednik bliz Alyshty, u Chuchelskogo perevala [Ukraine: [Crimea]: near Alushta: Krymskij reservation], 6 Aug. 1926, *G. Poplavskaja* (holotype LE n.v.).

Perennial, rhizomatous, 19 - 80 cm high. Basal innovations extravaginal. Leaf-sheaths glabrous, rarely pilose, margins non-overlapping. Leaf-blades 2.5 - 7 (42) cm x 3 - 5 (-11.5) mm, flat, glabrous. Ligule 0.5 - 2.5 (-4) mm, truncate.

Panicle open, erect, 5 - 16 cm long. Pedicels smooth and recurved. Spikelets 4 - 7 x 4 - 6 mm, 4 - 10-flowered, laterally compressed, ovate. Glumes herbaceous, cordate at the base, glabrous, smooth, equal to subequal; lower glume 2.8 x 1.3 - 1.9 mm, 3 (5)-nerved; upper glume 3 - 4 x 1.3 - 1.9 mm, 3-nerved. Lemmas herbaceous, laterally compressed, with a slightly distinct gibbous back, distinct from the broad margins, cordate at the base, 7 - 9-nerved, glabrous, rarely with translucent thick trichomes on the back, obtuse at the apex, muticous, margins glabrous, non-inrolled at the apex; lower lemma 3.2 - 4.2 x 2 - 2.5 mm. Paleas elliptic, glabrous between the keels, rarely with translucent thick trichomes, keels glabrous; lower palea 2.1 - 3.5 x 1.1 - 3.4 mm. Lodicules linear-lanceolate or flabelliform. Stamens 3. Hilum elliptic.

Fig. 3.A-D.

DISTRIBUTION. Temperate regions. Native in Eurasia, adventitious in South America.

Map 3.

SELECTED COLLECTIONS. AUSTRIA. Niederösterreich, St. Pölten, Lössterasse beim Prater, 7 June 1875, *Hackel* (W 1916-17228). **BELGIUM.** Bouffouesc, 11 June 1864, *Eosoprinet* (W 1956-1732). **BOSNIA.** Sarajevo Dist., Miljaska Valley, 19 June 1931, *Gilliat-Smith* 2926 (K). **BULGARIA.** M. Vitosa, in pratis silvaticis, 3 July 1952, *Vikodzevssky* 219 (L). **CZECH REPUBLIC.** Stredocesky Nymburk, 15 June 1966, *Dolezabová & Cistulkavá*, 59 (L). **ENGLAND.** Surrey, 27 June 1947, *Van Rossen* (L). **FINLAND.** Alandia, 25 June 1934, *Georg-Rydman* (L0532695). **FRANCE.** Dep.Dordogne, Les-Eyzis-de-Tayac, 23 May 1970, *Leenhouts* 2278 (L). **GEORGIA.** Caucasus occidentalia, dist. Sukhumi, vicinitas latior pagi Pskhu, in pascuo Bashta dicto, 7

Aug. 1983, *Vasák* (W 18599). **GERMANY.** Salzburg, Osterhorngruppe, Hinterwinkel am Gaisberg, 11 July 1990, *Geisler* (W). **GREECE.** Shady streamside bank e below Papignon, N. E. of Joannelna, 18 June 1974, *Lancaster* 4 (BM). **HUNGARY.** Comet, Pest, im aperti sylvarum inter Pils Szent Kerenzt et Sikaros, 16 June 1904, *Degen* 376 (L). **ITALY.** Piemonte, Valle Maiara, next San Damiano Macro, 27 April 1977, *Newman et al.* 134/1 (K). **LITHUANIA.** 1893, *Rudominorona* (W). **MOROCCO.** El Rif, 1903, *Gandoger* (BM). **NEPAL.** Bhurchula Lekh. near Jumla, 14 July 1952, *Polunin et al.* 4664 (K). **POLAND.** Carpati Occidentales, montes Pieniny (districtus Norwy Targ), supra oppidulum Krócienko, locus vocatus Stolarzówka, 14 July 1972, *Neck* (W). **RUSSIA.** Auf Alpenwiesen in der Landschaft Ossetien im Kaukasus, 24 July 1899, *Marcowicz* 263 (W). **SCANDINAVIA.** Isthmus Karekicus, par. Kivinebb, Polviselki, in pratis, 3 July 1913, *Sillman* 472 (K). **SPAIN.** Madrid, Embalse de Santillana, 10 June 1980, *Sánchez-Mata* (K). **SWISS.** Leysin Waadt, 5 June 1921, *Smit & Smit-Everts* 2033 (L). **TURKEY.** Prov. Bursa, along the road Ulu-Dag-Bursa, 21 June 1959, *Henmipman et al.* 2081 (L). **UNITED STATES OF AMERICA.** Danvers, McLean, Illinois, s.d., *Oakes* (L).

HABITAT. On meadows, forests glades, among shrubs up to upper mountain belt.

VERNACULAR NAMES. Common Quaking-grass (English, Massachussets, Danvers), Gewöhnlicher Zittergrass (German, Conert, 2000).

USES. Ornamental. Tzvelev (1983) also refered the use as fodder plants.

4. *Briza minor* L. (1753: 70) Type: Hab. in Helvetia. Italia. (lectotype LINN 88.1!

(lectotype designated by Hubbard, 1970))

Briza aspera Knapp (1804: 61) nom. ill. superfl. based on *B. minor* L.

Briza virens L. (1762: 103) Type: Hab. In Oriente, Hispania. Alfrömer (lectotype LINN-88.3!)

Briza trichotoma Steud. (1846: 20). Type: Japan, *Göring 4* (holotype P! isotype MO!)

Annual, without rhizomes, 10 - 60 cm high. Basal innovations intra-vaginal. Leaf-sheaths glabrous, margins non-overlapping. Leaf-blades (4) 9 - 10 cm x 2.3 - 7 mm, flat, glabrous. Ligule 3 - 7 mm, acute.

Panicle open, erect, 4 - 15 cm long. Pedicels smooth and recurved. Spikelets 2.2 - 4.8 x 3.2 - 4.7 mm, 4 - 8-flowered, laterally compressed, triangular. Glumes herbaceous, cordate at the base, glabrous, smooth or scaberulous, equal to subequal; lower glume 1.7 - 3 x 0.9 - 1.5 mm, 3 (5)-nerved; upper glume 2.1 - 2.8 x 1 - 1.5 mm, 3 (5)-nerved. Lemmas herbaceous, slightly laterally compressed, with a gibbous back, strongly distinct from the broad margins, cordate at the base, 7-nerved, with translucent thick trichomes on the back, rarely glabrous, obtuse at the apex, mucicous, margins glabrous, inrolled at the apex; lower lemma 1.8 - 2.8 x 1.4 - 2.2 mm. Paleas elliptic to obovate, with translucent thick trichomes between the keels, rarely glabrous, keels glabrous; lower palea 1.3 - 1.8 x 0.7 - 1 (1.4) mm. Lodicules linear. Stamens 3. Hilum elliptic.

Fig. 4.A-E.

DISTRIBUTION. Temperate and subtropical regions. Native in Eurasia, adventitious in America, Africa and Australasia. Map 4.

SELECTED COLLECTIONS. ALGERIA. Wilaya Tizi Duzou: norliche Djur djura, 8 km E Azazga nahe der strasse nach El Kseur, 14 June 1984, *Podlech 39280* (M).

ARGENTINA. Misiones, Apóstoles, Azara, Arroio Chimiray, 19 Feb. 1989, Campo & Uheita H. 3775 (MPUC). **AUSTRALIA.** Western Australia, Near Yangebup Lake, 23 Nov. 1982, *Rechinger 59995* (W). **AZORES.** Ilha do Pico, E of Madalena, from Furna de Frei to Madalena, through Dois caminhos, 2 June 1999, *Vitek 99-273* (W). **BOLIVIA.** Dep. La Paz, Prov. Bautista, Saavedra, Charazani, weg von Charazani nach Jatichulaya, 17 May 1985, *Feuerer 23326* (K). **BRAZIL.** Paraná, Parque Barigui, Curitiba, 8 Nov. 1996, *Kozera & Dittrich 347* (MBM). **CANARY ISLANDS.** Tenerife, Div. Adm. Santa Cruz, Anagagebirge, E of Roque de los Pasos, 20 June 1985, *Vitek 21/03* (W). **CHILE.** Región de Valparaíso, Prov. Quillota, Cuesta El Melon, 2 Nov. 1987, *Rechinger & Rechinger 63219* (W). **CHINA.** Shanghai, May 1908, *Schindler 275* (W). **COLOMBIA.** Dep. Antioquia, Planicie de selva abierta y esparcida, poco húmeda, 17 April 1949, *Silva et al. 402* (BM). **COSTA RICA.** Prov. Cartago, south of Volcan Irazu, along Carretera Nacional, 18-19 Feb. 1984, *Khan et al. 993* (BM). **ECUADOR.** 1860, *Fraser* (BM). **FRANCE.** Corsica; Calvi, 9 May 1956, *Wyk* (L0532739). **GREECE.** Peloponnesus, Laconia, Mt. Malevo, 17 June 1958, *Pinatzi* (W). **GUATEMALA.** Sololá, on moist rocks of Panajachel water falls, road to Sololá, 12-23 Jan. 1966, *Molina et al. 16253* (W). **HAWAII.** Hamahua, Waimea, Paauhau Paddock, 9 June 1938, *Hosaka 2266* (L). **HOLLAND.** Côte ordantali, Jan. 1845, *Verreaux 226* (P). **INDIA.** Medras State, Nilgizis Ootacamund, 10 Dec. 1969, *Werff 233* (L). **INDONESIA.** Java, Pasuruan Wonosari, Kletak Forest, 14 Sept. 1992, *Veldkamp &*

Roos 8721 (L). **IRAN.** Prov. Gilan, Mordab Lake, 7 km W of Bandar-e Pahlavi, 14 May 1971, *Rechinger 39667* (W). **ITALY.** Isola di Capri, 4 June 1968, *Lippert & Zollitsch 8150* (M). **JAMAICA.** Abbey Green, in the Blue Mountains, 4 Oct. 1913, *Hitchcock* (W, L). **JAPAN.** Kyushu Island, Fukuokaken Hakozaki, near Fukuoka city, 20 May 1928, *Ichikawa 48* (W). **MALAYSIA.** Pahang, Dist. of Cameron, Cameron Highlands, Gumong Brinchang, 22 March 1987, *Worthington 12642* (L). **MEXICO.** Chiapas, Motozintla, 7 Feb. 1990, *Stafford et al. 254* (BM). **MOROCCO.** Chefchaouene, Montes de Jebala, entre Derdara y Ksar-el-Kebir, Akarate, 31 May 1995, *Fergusson et al. 6321/95* (K). **NEW GUINEA.** Edie Creek, Wau, Morobe District, 13 Aug. 1968, *Millar NGF.12147* (K). **NEW ZEALAND.** North Island, Waitemata County, Forest Hill, roadside bordering Reserve, 12 Nov. 1979, *Bangerter 5433* (L). **PERU.** Dep. Cuzco, Prov. Urubamba, Loc. Winaiwalina, Inca land, 10 March 1944, *Vargas 4134* (B). **PORTUGAL.** Ilha da Madeira, Levada da Caniçal, in the E side of the Island, ca. 1 km W of the Tunel Caniçal, 13 April 1997, *Vitek 97-81* (W). **SOUTH AFRICA.** Saron, Oct. 1896, *Schlechter 10611* (W). **SPAIN.** Prov. Coruña, Meline, Meire, 15 July 1988, *Rechinger & Rechinger* (W). **SRI LANKA.** Loc. Horton Plains, near Farr Inn, s.d., *Van Beusekom & Van Beusekom 1462* (L). **TURKEY.** Mugla Fethiye-Köycegiz, at Küçük Kargı, at the Sea, 28 April 1970, *Bozakman & Fitz. 345* (W). **URUGUAY.** Montevideo, Sayago, Nov. 1968, *Rosengurt 10957* (B). **UNITED STATES OF AMERICA.** Amador County/ New York falls, 12 June 1894, *Hansen 608* (L).

HABITAT. Open grasslands and road sides.

VERNACULAR NAMES. Small Quaking-grass (English, Townsend & Guest, 1968), Green Quaking-grass (English, Knapp, 1804), Lesser Quaking-grass (English, Isles of

Sally), Small Shivering Grass (English, Lord Howe Island), Shake-shake-grass (English, Hawaiian Islands), Ilusión (Spanish, Colombia), Capim-treme-treme (Portuguese, Brazil).

USES. Ornamental.

Chascolytrum Desv. (1810: 190). Typus generis: *Chascolytrum subaristatum* (Lam.) Desv.

Briza L. sect. *Chascolytrum* (Desv.) Benth. & Hook. (1883: 72)

Briza L. subgen. *Chascolytrum* (Desv.) Parodi (1920: 120)

Calothea (“Calosteca”) Desv. (1810: 190)

Briza L. sect. *Calothea* (Desv.) Benth. & Hook. (1883: 1194)

Briza L. subg. *Calothea* (Desv.) Parodi (1920: 130)

Poidium Nees (1836: 450)

Briza L. sect. *Poidium* (Nees) Ekman (1913: 56)

Briza L. subg. *Poidium* (Nees) Parodi (1920: 133)

Chondrachyrum Nees (1836: 449)

Lombardochloa Roseng. & Arrill. (1979: 260), **synon. nov.**

Briza L. sect. *Lombardochloa* (Roseng. & Arrill.) Longhi-Wagner (1987: 22)

Microbriza Parodi ex Nicora & Rùgolo (1981: 292), **synon. nov.**

Rhombolytrum Link (1833: 296), **synon. nov.**

Gymnachne Parodi (1938: 29), **synon. nov.**

Erianthecium Parodi (1943: 75), **synon. nov.**

Perennial, caespitose, rhizomatous. Basal internodes of the culms non-thickened, occasionally thickened and bulbous. Basal innovations extravaginal, rarely intravaginal. Leaf-sheaths glabrous or pilose, smooth or scabrous, margins overlapping or not. Leaf-blades linear to linear-lanceolate, flat, convolute or conduplicate, glabrous or pilose, smooth or scabrous. Ligule membranaceous, acute to truncate.

Panicle open to contracted, erect or pendulous. Pedicels smooth or scabrous, straight. Spikelets many-flowered, laterally compressed or cylindrical to globose, disarticulating above the glumes, orbicular, oblong, elliptic, elliptic-lanceolate, rhomboid, lanceolate to ovate. Florets imbricate, obscuring the rhachilla, occasionally loosely imbricate, rhachilla apparent. Glumes persistent, herbaceous to chartaceous, convex to naviculate, non-cordate and tapering towards the base, glabrous or pilose, smooth or scabrous, equal, subequal or unequal, 3 – 7 (-11)-nerved. Lemmas herbaceous, chartaceous to coriaceous, laterally to dorsi-ventrally compressed, with or without a gibbous back, back strongly or slightly distinct from the narrow or broad margins, cordate at the base, or non-cordate and tapering towards the base, 5 - 11-nerved, glabrous or pilose on the back, obtuse, truncate to acute, or occasionally bidentate at the apex, muticous, mucronate or awned, margins glabrous, ciliate or pilose, smooth or scabrous, inrolled or not at the base, non-inrolled at the apex, rarely with one oil gland (or its scar) in each side at the base (*C. rufum*). Paleas 2-keeled, linear-lanceolate, lanceolate, elliptic-lanceolate, elliptic, obovate, elliptic-orbicular to orbicular, membranaceous, chartaceous to coriaceous, smooth, echinulose, tuberculate to punctuate between the keels, glabrous or pilose between the keels, keels glabrous, ciliate or ciliate.

Lodicules 2, linear, linear-lanceolate, elliptic-lanceolate to flabelliform, occasionally bilobate. Stamens 1 - 3. Ovary glabrous, styles terminal. Caryopsis sub-orbicular, than

transverse section plano-convex, or oblong to elliptic, transverse section concavo-convex.

Hilum linear, linear-elliptic, elliptic or punctiform.

Flowering: Spring and early Summer.

Chromosome counts: $2n=28$ ($x=7$). (Matthei, 1975)

Chascolytrum, as accepted here, includes 22 American species, with greater species richness in Southern Brazil.

NOTE.1. The genus *Chascolytrum* is morphologically diverse and needs to be identified combining many characters. It is not possible, however, to accept sections or subgenera, due to the existence of species with intermediate shapes, which link the various morphologic groups. Besides, all these species appear linked in a monophyletic group at the molecular level, and splitting the genus would only generate several small paraphyletic groups (Essi, Longhi-Wagner & Souza-Chies, in preparation).

Key to *Chascolytrum* species

- 1. Lemmas awned, awn 1 – 4.4 mm.....2
- 1'. Lemmas muticous, occasionally mucronate, mucron up to 0.5 mm.....3
- 2. Basal internodes of the culms thickened, bulbous. Leaf-blades flat, pilose on both surfaces.....*C. bulbosum*
- 2'. Basal internodes of the culms non-thickened. Leaf-blades conduplicate, glabrous.....*C. brizoides*
- 3. Lemmas with a gibbous back strongly distinct from the margins, rufous or yellow-gold on the back, occasionally pale yellow.....4

- 3'. Lemmas without or with a gibbous back, back distinct, slightly distinct or non-distinct from the margins, cream, pale, whitish or purplish on the back.....6
4. Paleas membranaceous, lanceolate. Lemmas rufous on the back with broad margins, one oil gland (or its scar) on each side, at the base*C. rufum*
- 4'. Paleas coriaceous, elliptic to orbicular. Lemmas rufous, pale yellow or yellow-gold on the back, with broad or narrow margins, without oil glands.....5
5. Panicle contracted, erect. Spikelets laterally compressed, 2 - 3-flowered. Lemmas with pale yellow or yellow-gold gibbous back, and narrow margins.....*C. scabrum*
- 5'. Panicle open, pendulous. Spikelets cylindrical, 3 - 6-flowered. Lemmas with rufous or less frequently yellow-gold gibbous back, and broad margins.....*C. lamarkianum*
6. Paleas coriaceous or chartaceous, glabrous or ciliolate on the keels.....7
- 6'. Paleas membranaceous, ciliate on the keels.....11
7. Paleas chartaceous, obovate.....*C. parodianum*
- 7'. Paleas coriaceous, elliptic, elliptic-orbicular to orbicular.....8
8. Spikelets 1.2 - 1.8 mm long, 2 - 3-flowered. Paleas elliptic. Lemmas and paleas echinulose.....*C. poaemorphum*
- 8'. Spikelets 2.4 - 9 mm long, 4 - 14-flowered. Paleas elliptic-orbicular to orbicular. Lemmas and paleas non-echinulose.....9
9. Paleas glabrous between the keels, rarely pilose, trichomes non-capitate. Lemmas' margins glabrous.....*C. subaristatum*
- 9'. Paleas with capitate trichomes between the keels, occasionally deciduous at maturity (*C. altiplanense*). Lemmas with or without flattened trichomes on the margins, at the base.....10

10. Margins of the lemma with dense flattened trichomes at the base. Lower lemma 2.5 – 3.8 mm long. Spikelets cylindrical.....*C. altiplanense*
- 10'. Margins of the lemma glabrous (rarely 2 - 4 capitate trichomes around the callus). Lower lemma 4.5 – 5 mm long. Spikelets subcylindrical to laterally compressed.*C. paleapiliferum*
11. Margins of the lemmas ciliate at the lower third, trichomes longer than 0.3 mm.....12
- 11'. Margins of the lemmas glabrous.....14
12. Paleas pilose between the keels. Lemma surface pilose.....*C. bidentatum*
- 12'. Paleas glabrous between the keels. Lemma surface glabrous, scaberulous.....13
13. Panicle open. Spikelets elliptic-lanceolate, 3 - 6 mm long. Glumes herbaceous, lower glume 1.5 – 3 mm long. Lower palea 1.5 – 2 mm long.....*C. monandrum*
- 13'. Panicle contracted. Spikelets rhomboid, 5 - 8 mm long. Glumes chartaceous, lower glume 4.5 - 5 mm long. Lower palea 3 – 3.2 mm long.....*C. rhomboideum*
14. Lemmas with a gibbous back strongly distinct from the broad margins, glabrous, whitish, papillose and with a salient midvein on the back, rarely without papillae.....*C. uniolae*
- 14'. Lemmas without a gibbous back distinct from the narrow or broad margins, or only slightly distinct, glabrous or pilose, pale, pale green or purplish, without papillae, smooth, scaberulous or tuberculate, midvein non-salient on the back.....15
15. Panicle contracted, erect. Spikelets rhomboid. Glumes chartaceous, surface scabrous.....*C. koelerioides*
- 15'. Panicle open, pendulous or erect. Spikelets elliptic, lanceolate, oblong or ovate. Glumes herbaceous, surface smooth, occasionally scaberulous or tuberculate towards the apex.....16

16. Lemmas glabrous, tuberculate. Leaf-sheaths pilose.....*C. brachychaete*
- 16'. Lemmas glabrous or pilose, smooth or scaberulous. Leaf-sheaths glabrous.....17
17. Lemmas without a gibbous back, without distinction between back and margins, lower lemma 0.4 – 1 mm wide. Spikelets ovate.....18
- 17'. Lemmas without or with or a slightly gibbous back, distinct or not from the margins, lower lemma (1-) 1.2 – 2.5 mm wide. Spikelets oblong, elliptic or lanceolate.....19
18. Leaf-blades convolute, 1.3 – 2 mm wide. Lower lemma 0.4 – 0.6 mm wide. Florets loosely imbricate, rhachilla aparent.....*C. brasiliense*
- 18'. Leaf-blades flat, 3 - 4 mm wide. Lower lemma 0.7 – 1 mm wide. Florets imbricate, obscuring the rhachilla.....*C. itatiaiae*
19. Paleas pilose between the keels. Lemmas surface usually pilose, occasionally glabrous.....*C. juergensii*
- 19'. Paleas glabrous between the keels. Lemmas glabrous.....20
20. Lemmas cordate at the base, with a slightly distinct gibbous back, muticous. Spikelets elliptic.....*C. calotheca*
- 20'. Lemmas non-cordate at the base, without a gibbous back, mucronate, rarely muticous. Spikelets lanceolate or oblong.....21
21. Lemmas chartaceous, lower lemma 1 – 1.6 mm wide. Spikelets lanceolate, laterally compressed.....*C. ambiguum*
- 21'. Lemmas herbaceous, lower lemma 1.4 – 2.5 mm wide. Spikelets oblong, subcylindrical.....*C. latifolium*
-

1. *Chascolytrum altiplanense* Essi, Longhi-Wagner & Souza-Chies, unpublished.

Plants 45 - 77 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, smooth, margins non-overlapping. Leaf-blades linear, 7 - 45 cm x 0.8 - 2.2 mm, flat or convolute, glabrous. Ligule 1.8 - 2.5 mm, acute.

Panicle contracted, erect, 3 - 7 cm long. Pedicels smooth. Spikelets 5.5 - 6 x 4 mm, 5 - 8-flowered, cylindrical, oblong. Florets imbricate, obscuring the rachilla. Glumes herbaceous, convex, glabrous, smooth, acute or obtuse, subequal; lower glume 2.3 - 3 x 0.9 - 1.5 mm, 5-nerved; upper glume 2.3 - 3 x 1 - 1.5 mm, 5-nerved. Lemmas chartaceous to coriaceous, dorsi-ventrally compressed, with a gibbous pale back, back strongly distinct from the broad margins, cordate or not at the base, 7-nerved, midvein non-salient on the back, glabrous or with sparse flattened trichomes on the back, truncate or acute at the apex, muticous or mucronate, margins with dense flattened trichomes at the base, inrolled or not at the base, without oil glands; lower lemma 2.5 - 3.8 x 1.2 - 2 mm. Paleas elliptic-orbicular to orbicular, coriaceous, with capitate trichomes between the keels, occasionally deciduous at maturity, keels ciliolate at the upper half; lower palea 1.5 - 1.8 x 1 - 1.2 mm. Lodicules flabelliform. Stamens 1 - 2. Caryopsis sub-orbicular, plano-convex. Hilum elliptic to linear-elliptic.

Fig. 5.A-D.

DISTRIBUTION. Restricted to Bolivia. Map 5.

BOLIVIA. Chuquisaca. on E side of Cerro Chataquila near Punilla (Sucre-Ravelo), 9 April 1995, *Wood 9665* (LPB); Tomina, Lampacillas, C 30 km S. of Padilla towards

Monteagudo, 31 Dec. 1994, *Wood 9068* (K); Lampacillas, ca. 30 km S of Padillas towards Monteagudo, 31 Dec. 1994, *Wood 9068* (LPB); Vamporz, Sucre On a grassy bank on sandstone rocks by a seasonal Watercourse, SE of the town, 17 April 1994, *Wood 8314* (K); Yamparaez, on the ascent, c 3 km E. of La Cienaga towards, 16 March 1996, *Wood 10841* (K, LPB). **Cochabamba.** Mizgue a 10 km S of Totora towards Aiquile, 19 Dec. 1995, *Wood 9461* (K); Tapacarí, entre Parotani y Challa, 21 Feb. 1979, *Ceballos, Charpin, Casas & Bermejo BO-445* (G); Tiraque, on pass just W of Sacabambilla on old Cochabamba-Santa Cruz road, 2 Feb. 1996, *Wood 10486* (LPB); Prov. Chapare, Parque Tunari, bosque de Polylepsis, 23 Feb. 1991, *Hensen 998* (LPB); 51 Km W of Cochabamba, 3100 m Schub Slender, loosely tufted perennial, 25 March 1981, *Renvoize & Cope 4082* (K). **Santa Cruz.** Caballero, at summit of Road on Loma Grande, c. 6 - 8 km W of Comarapa on road to Siberia, 13 March 2002, *Wood 17802* (K); Vallegrande, c. 16 km from Vallegrande on road to Masucuri, 11 Feb. 1996, *Wood 10601* (LPB); Voladerogebirge, 29 Jan. 1928, *Troll 1101* (M, B).

HABITAT. Highlands, between 2200 and 3600 m.

ETIMOLOGY. The epithet *altiplanense* refers to the species habitat.

NOTE. Many herbarium specimens of *C. altiplanense* have been identified either as *C. subaristatum* or *C. paleopiliferum*. Indeed, *C. altiplanense* is morphologically related to both species, the three sharing similar palea and lemma shapes, and coriaceous palea. Renvoize (1998) cited some specimens of *C. altiplanense* under *Briza subaristata* Lam. [= *C. subaristatum*], emphasizing the presence of capitate hairs on the palea back, typical of

C. paleapiliferum. However, *C. altiplanense* differs from both mainly by the presence of dense flattened trichomes on the lemma, at the base. In addition, *C. altiplanense* differs from *C. paleapiliferum* by the distinct spikelet shape, glume shape and lemma size. One collection of *C. altiplanense* (Wood 10768, LPB) was included in a phylogenetic molecular approach (Essi, Longhi-Wagner & Souza-Chies, in preparation), and turned out to be genetically distinct from *C. subaristatum* and *C. paleapiliferum*.

2. Chascolytrum ambiguum (Hack.) Essi, Souza-Chies & Longhi-Wagner comb. nov.

Type: Brasilia, in provincia Rio de Janeiro, *Glaziou 17956*. (holotype W!; isotypes: K! G! MO!)

Briza ambigua Hack. in Oesterr. Bot. Z. 52: 308. (1902)

Poidium ambiguum (Hack.) Matthei (1975: 121)

Plants 35.5 - 95 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, smooth, margins non-overlapping. Leaf-blades linear-lanceolate, 9 - 26 cm x 2 - 7 mm, flat, glabrous. Ligule 1.1 – 5 mm, truncate to obtuse. Panicle open, erect, 8.5 - 19 cm long. Pedicels smooth. Spikelets 3.9 - 6 x 1.9 - 4 mm, (3) 4 - 6-flowered, laterally compressed, lanceolate. Florets imbricate to loosely imbricate, obscuring or not the rhachilla. Glumes herbaceous to chartaceous, navicular, glabrous, smooth, acute, subequal; lower glume 2.5 - 3.1 x 0.5 - 0.8 mm, 3-nerved; upper glume 2.9 – 4 x 0.5 - 1 mm, 3-nerved. Lemmas chartaceous, laterally compressed, pale to pale green, without a gibbous back, non-cordate at the base, (3) 5 - 7-nerved, midvein non-salient on the back, glabrous, smooth, acute or bidentate at the apex, mucronate, rarely muticous, margins glabrous, non-inrolled at the base, without oil glands; lower lemma 3 – 4.5 x 1 –

1.6 mm. Paleas elliptic-lanceolate, membranaceous, smooth, glabrous between the keels, keels ciliolate; lower palea 1.8 - 3 x 0.5 – 1.1 mm. Lodicules linear-lanceolate or flabelliform. Stamens 1. Caryopsis elliptic, concavo-convex. Hilum elliptic.

Fig. 6.A-D.

DISTRIBUTION. Brazil. Map 6.

BRAZIL. Minas Gerais. Parque Nacional do Caparaó, 1 May 1988, *Krieger et al.* 22314 (MO); Pontão do Cristal - tufts, in mon, rocks, steep slope of Pontão Crystal, 30 April 1925, *Chase 9704* (W). **Santa Catarina.** Urubici, Serra do Corvo Branco, 4 Dec. 2003, *Essi et al.* 190, 191 (ICN); Urubici, Serra do Corvo Branco, 13 Dec. 2004, *Essi & Lüdtke* 325 (ICN). **Rio de Janeiro.** Serra dos Órgãos, Centralstock, Jan. 1916, *Luetzelburg* 12224 (M); Itatiaia, 1 May 1950, *Brade & Vianna* 20381 (RB); Itatiaia, Parque Nacional, 9 Jan. 1981, *Winge et al.* 1544 (ICN); Nova Friburgo, Pico da Caledônia, Jan. 1996, *Longhi-Wagner* 4006, 4007 (ICN); Petrópolis – Cascatinha, 19 Aug. 1949, *Góes & Dionisis* 471 (RB); Petrópolis, entre Araras e Vale das Videiras, Morro do Cuca, Pico do Pindoba, campo de altitude, 2 June 1984, *Martinelli et al.* 9883 (RB); Serra da Gramma, 25 April 1925, *Chase 9566* (RB). **Rio Grande do Sul.** Cambará do Sul, Itaimbezinho, 16 Dec. 1971, *Normann et al.* (BLA).

HABITAT. Highlands of Southern and Southeastern Brazil. Rocky slopes and grasslands.

ETIMOLOGY. The epithet refers to the shape of spikelets of the species, considered by Hackel (1902) as intermediate between the genera *Poa* L. and *Briza*.

NOTE. *Chascolytrum ambiguum* is morphologically related to *C. calotheca*, especially due to the similar shape and color of the spikelets, and to the lemmas smooth and shiny. However, the plants of *C. calotheca* are usually more robust, the inflorescences usually pendulous, and its lemmas are wider and cordate at the base, with a index length/width 1.7 – 1.9 (2.5 – 3 in *C. ambiguum*).

3. *Chascolytrum bidentatum* (*Roseng. Arrill. & Izag.*) *Essi, Souza-Chies & Longhi-Wagner comb. nov.* Type: Brasilia, Rio Grande do Sul, Município Rio Pardo, prope Fazenda Soledade, 70 m.s.m., Dec. 1905, *C. Jürgens* (holotype W!; isotypes US n.v., BAA n.v.)

Eragrostis monandra Hack. in Repert. Spec. Nov. Regni Veg. 7: 320. (1909)

Briza bidentata Roseng., Arrill. & Izag. (1968: 24)

Rhombolytrum monandrum (Hack.) Nicora & Rúgolo (1981: 302)

Plants 60 – 80 (120) cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous or pilose, smooth or scabrous, margins non-overlapping. Leaf-blades linear-lanceolate, 5.2 – 30 cm x 2 – 5.2 mm, flat, glabrous or pilose on the abaxial surface. Ligule 0.3 - 1 mm, truncate.

Panicle open or contracted, erect or pendulous, 6.5 – 15 (54) cm long. Pedicels smooth.

Spikelets 3.2 - 5 x 1.1 – 2.8 mm, 4 - 10-flowered, laterally compressed, oblong to rhomboid. Florets imbricate, obscuring the rhachilla. Glumes chartaceous, navicular, glabrous, scabrous, subequal, acute; lower glume 1.6 – 3.3 x 0.4 – 0.7 mm, 3-nerved; upper glume 1.5 – 3 x 0.5 – 0.7 mm, 3 (5)-nerved. Lemmas chartaceous to herbaceous, dorsi-

ventrally compressed, pale, without a gibbous back, margins non-distinct from the back, non-cordate at the base, 5-nerved, midvein non-salient on the back, pilose, obtuse or acute at the apex, mucicous, margins ciliate at the lower third, trichomes longer than 0.3 mm, inrolled at the base, without oil glands; lower lemma 2 – 2.7 x 0.6 – 0.9 mm. Paleas elliptic-lanceolate, membranaceous, pilose between the keels, keels ciliate; lower palea 1.6 – 2.3 x 0.4 – 0.7 mm. Lodicules linear. Stamens 1 - 2. Caryopsis oblong, concavo-convex. Hilum punctiform.

Fig. 7. A-D.

DISTRIBUTION. Brazil, Uruguay. Map 7.

SELECTED COLLECTIONS. BRAZIL. Paraná. Horizonte [Novo Horizonte], 2 Km de Palmas, 24 Nov. 1981, *Hatschbach & Hirzinker 44415* (MBM); Palmas, Loc. Santo Agostinho, 13 Dec. 1966, *Hatschbach 15424* (MBM). **Rio Grande do Sul.** Bagé, Minas de Camaquã, 28 Nov. 1998, *Longhi-Wagner 5041* (ICN); Caçapava do Sul, BR 153, 21 Nov. 2003, *Essi et al. 106* (ICN); Canela, Cascata do Caracol, 20 Oct. 1971, *Valls 1851* (ICN); Esmeralda, Estação Ecológica de Aracuri (Capão da Fonte), 17 Jan. 1979, *Sampaio & Lemos 749* (ICN); Faxinal do Soturno, Cerro Comprido, Nov. 1988, *Sobral 5969* (ICN); Invernardinha, 1 Jan. 1956, *Mattos 3970* (BLA); Lagoa Vermelha, Escola Técnica Rural, 08 Nov. 1962, *Rosengurtt 9089* (BLA); Lavras do Sul (estr. Bagé-Lavras, 51 km após trevo entrada, após Arroio Cardoso), 16 Nov. 1975, *Longhi et al. 270* (ICN); Passo Fundo, Nov. 1986, *Valls et al. 10514* (ICN); Piratini, morro em frente da ponte do Arroio Piratini, 16 Nov. 2003, *Essi et al. 55* (ICN); Três Passos – Criciumal, 1 Nov. 1971, *Valls et al. 1796* (BLA, ICN). **Santa Catarina.** Celso Ramos, 14 Dec. 2004, *Essi & Lüdtke 356* (ICN);

Curitibanos, 16 km hacia Lebon Régis, Rio Marombas, 9 Dec. 1992, *Rúgolo et al.* 1563 (MO); São Joaquim, paredão arenítico ao lado do Rio Canoas, 12 Dec. 2004, *Essi & Lüdtke* 318 (ICN). **URUGUAY. Rivera.** Tranqueras, ruta 30, 4 Dec. 1957, *Rosengurtt B-6864* (BLA).

HABITAT. Shaded habitats. Frequent in the limit between grasslands and forests.

NOTE. *Chascolytrum bidentatum* is morphologically related to *C. rhomboideum*, due to the chartaceous to herbaceous lemmas, ciliate at the lower third, chartaceous glumes and rhomboid spikelets. *Chascolytrum rhomboideum* differs by the glabrous paleas between the keels, wider spikelets and longer glumes, almost as long as the first florets.

4. *Chascolytrum brachychaetum* (Ekman) Essi, Longhi-Wagner & Souza-Chies comb.

nov. Type: Brasil: in altoplanitie, Curytiba, in campo, 15 Feb 1911, caespium unicum, *P.*

Dusén 13390 (holotype S!; isotypes L! BM! K! L! B n.v., BAA n.v, US n.v.)

Briza brachychaete Ekman in Ark. Bot. 13 (10): 60, t. 4, f. 3. (1913)

Poidium brachychaetum (Ekman) Matthei (1975: 107)

Microbriza brachychaete (Ekman) Parodi ex Nicora & Rúgolo (1981: 292)

Plants 24 - 130 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths pilose, scaberulous, margins non-overlapping. Leaf-blades linear, 4 - 23 cm x 1 - 5 mm, flat, pilose only at the base, usually at the adaxial surface. Ligule 0.9 - 2.2 mm, obtuse to truncate.

Panicle open, erect, 10 – 19.5 cm long. Pedicels smooth. Spikelets 1.9 – 2.5 x 1.4 – 3.8 mm, 2 - 4-flowered, laterally compressed, ovate. Florets imbricate, obscuring the rachilla. Glumes herbaceous, convex or navicular, glabrous, tuberculate towards the apex, equal to subequal, acute; lower glume 1.9 – 2.4 x 0.4 – 0.6 mm, 1 - 3-nerved; upper glume 1.9 – 2.6 x 0.5 – 0.7 mm, 3-nerved. Lemmas herbaceous, laterally compressed, pale to purplish, without a gibbous back, margins non-distinct from the back, non-cordate at the base, 3-nerved, midvein non-salient on the back, glabrous, tuberculate, obtuse at the apex, mucicous, margins glabrous, smooth or scabrous, non-inrolled at the base, without oil glands; lower lemma 1.6 - 2 x 0.6 – 0.8 mm. Paleas elliptic-lanceolate, membranaceous, smooth or punctuated, glabrous or sparsely pilose between the keels, keels ciliate; lower palea 1.3 – 1.7 x 0.5 – 0.7 mm. Lodicules lanceolate. Stamens 1 - 2. Caryopsis elliptic, concavo-convex. Hilum punctiform.

Fig. 8.A-D.

DISTRIBUTION. Southern Brazil. Map 8.

SELECTED COLLECTIONS. BRAZIL. Paraná. Campina Grande do Sul, 20 Nov. 1965, *Hatschbach 13152* (MBM, L, P). **Rio Grande do Sul.** Cambará do Sul, Itaimbezinho, 4 Jan. 1974, *Hickenbick 109, 115, 132, 133, 134, 135, 136, 140, 141, 142, 143, 150, 151* (ICN); Ibidem, 17 Nov. 2004, *Essi 297, 298, 300* (ICN); São José dos Ausentes, Monte Negro, 2 Dec. 2003, *Essi et al. 151* (ICN). **Santa Catarina.** Bom Jardim da Serra, at the head of the Rio do Rastro, 14 Dec. 1971, *Smith & Klein 15787* (HBR, K); Lauro Muller, a 25 km pasando la Serra do Rio do Rastro, 7 Dec. 1992, *Rúgolo et al. 1475* (ICN, MO); Morro da Igreja, São Joaquim [Urubici], 22 Jan. 1960, *Mattos 8317* (HBR).

HABITAT. Southern Brazil highlands. Wet places in open grasslands and rocky slopes.

5. Chascolytrum brasiliense (Nees ex Steud.) Essi, Longhi-Wagner & Souza-Chies **comb.**

nov. Type: Brasil: civit. Paraná, in parte septentrionali civitatis, *Sellow 4848* (lectotype BAA! (**designated here**); isoelectotype US).

Poidium brasiliense Nees ex Steud. in Syn. Pl. Glumac. 1: 288 (1854: 288)

Poa poidium Döll (1878: 119) Type: the same as *Poidium brasiliense* Nees ex Steud.

Briza brasiliensis (Nees ex Steud.) Ekman (1913: 57)

Plants 30 - 70 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, smooth, margins non-overlapping. Leaf-blades linear, 5.2 – 24 cm x 1.3 - 2 mm, convolute, glabrous or pilose on the abaxial surface. Ligule 1.8 – 4.8 mm, truncate to obtuse.

Panicle open, erect, 5 – 12 cm long. Pedicels smooth. Spikelets 2.5 – 3.8 x 1.4 – 3.6 mm, 3 - 5-flowered, laterally compressed, ovate. Florets loosely imbricate, rhachilla apert.

Glumes herbaceous, convex or naviculate, glabrous, smooth, subequal; lower glume 1.7 – 2.6 x 0.4 – 0.5 mm, 3-nerved; upper glume 1.9 – 2.6 x 0.4 – 0.6 mm, 3 - 5-nerved. Lemmas herbaceous, laterally compressed, pale to purplish, without a gibbous back, margins non-distinct from the back, non-cordate at the base, 5 – 7-nerved, midvein non-salient on the back, glabrous, scaberulous, acute at the apex, muticous, margins glabrous, inrolled or not at the base, without oil glands; lower lemma 1.9 – 2.3 x 0.4 – 0.6 mm. Paleas lanceolate to linear-lanceolate, membranaceous, smooth or sparsely punctuated, glabrous between the

keels, keels ciliate; lower palea 1.9 – 2.3 x 0.4 – 0.6 mm. Lodicules linear. Stamens 1.

Caryopsis elliptic, concavo-convex. Hilum elliptic to linear-elliptic.

Fig. 9.A-D.

DISTRIBUTION. Restricted to Brazil. Map 6.

SELECTED COLLECTIONS. BRAZIL. Minas Gerais. Serra do Caparaó, Pontão Cristal, 30 April - 4 May 1925, *Chase 9204* (MO). **Paraná.** Morretes, Serra Marumbi, subida do Pico Olimpo, 20 Oct. 1982, *Hatschbach 78202* (MBM); Ibidem, 20 Oct. 1988, *Hatschbach 45684* (MBM, K). **Rio de Janeiro.** Itatiaia, 28 Dec. 1936, *Pilger & Brade 113* (RB); Itatiaia, Parque Nacional do Itatiaia, campo de altitude, Vale do Lírio, 1 Nov. 2003, *Moura et al. 349 & 357* (ICN); Itatiaia, Planalto 2000 m, 22 - 28 Nov. 1958, *Markgrat & Brade 3686* (RB); Itatiaia, Planalto – 2100 m, March 1937, *Brade 15622* (B). **Rio Grande do Sul.** Cambará do Sul, Itaimbezinho, 23 Dec. 1975, *Sampaio et al. 183* (ICN). **São Paulo.** Campos do Jordão, 22 Oct. 1938, *Hashimoto 130* (RB); Campos do Jordão, Umuarama, 23 Nov. 1949, *Kuehn & Kuhlmann 2092* (K); São Paulo, Moóca 6 Oct. 1912, *Brade 6162* (BM).

HABITAT. Highlands of Southern and Southeastern Brazil. Grasslands and stony slopes.

ETIMOLOGY. The epithet is related to the geographic distribution of the species that is endemic to Brazil.

NOTE. Matthei (1975) selected a neotype for *Poidium brasiliense* Nees ex Steud. and included *Briza itatiaiae* Ekman in its synonymy. However, this specimen belongs to *Chascolytrum itatiaiae* (syn. *Briza itatiaiae* Ekman), which is here accepted as a different species, as proposed by Longhi-Wagner (2001). The holotype of the “true” *Poidium brasiliense*, from B Herbarium, was destroyed. However, there are some fragments of the holotype in other herbaria, one of them at BAA, that is designated here as the lectotype of *P. brasiliensis*.

The two species present small ovate spikelets and narrow herbaceous lemmas, without distinction between back and margins. *Chascolytrum itatiaiae* can be distinguished by its wider lemmas (0.7 - 1 mm), imbricated florets (obscuring the rhachilla) and wider (3 - 4 mm) and flat leaf-blades.

6. *Chascolytrum brizoides* (Lam.) Essi, Longhi-Wagner & Souza-Chies comb. nov. Type:

Uruguay: E. Monte-Video, *Commerson*. (holotype P-LAM!)

Bromus brizoides Lam. Tabl. Encycl. 1: 193, n. 1060 (1791)

Calotheca brizoides (Lam.) Desv. (1810: 190)

Briza brizoides (Lam.) Kuntze (1898: 341)

Briza patula Phil. (1896: 163) Type: Supra Angol. Nov. 1887 (holotype SGO n.v.)

Briza tandilensis Parodi (1920: 132) Type: Argentina: Buenos Aires, cerros del tandil, Nov. 1919, Parodi (holotype BAA n.v.; isotype US n.v.)

Plants (6) 10.5 - 72 cm high. Basal internodes of the culms non-thickened. Basal innovations intra-vaginal. Leaf-sheaths glabrous, margins non-overlapping. Leaf-blades

linear, 4 – 14 (26) cm x 0.6 – 2.4 mm, conduplicate, glabrous. Ligule 0.7 - 2.5 mm, obtuse or truncate.

Panicle open, erect, (2) 5 – 8.5 cm long. Pedicels smooth. Spikelets 5 - 10 x 2.2 - 6 mm, 4 - 8-flowered, laterally compressed, oblong. Florets imbricate, obscuring the rhachilla.

Glumes chartaceous, naviculate, glabrous, subequal; lower glume 2.8 – 5.8 x 0.6 – 1.2 mm, 3-nerved; upper glume 3.4 – 6.8 x 0.7 – 1.7 mm, 5-nerved. Lemmas chartaceous, slightly laterally compressed to dorsi-ventrally compressed, pale, without a gibbous back, with broad margins non-distinct from the back, non-cordate at the base, 5-nerved, midvein non-salient on the back, glabrous on the back, with a crown of long trichomes originated at the callus, truncate at the apex, awned, awn 1.3 – 4.4 mm, margins glabrous, scaberulous, non-inrolled at the base, without oil glands; lower lemma 5 - 7 x 2.5 – 3.6 mm. Paleas lanceolate, membranaceous, pubescent between the keels, keels ciliate; lower palea 2.4 – 4.1 x 1 – 1.4 mm. Lodicules bilobate. Stamens 3. Caryopsis oblong to elliptic, concavo-convex. Hilum elliptic.

Fig. 10.A-D.

DISTRIBUTION. Argentina, Brazil, Chile and Uruguay. Map 9.

SELECTED COLLECTIONS. ARGENTINA. Buenos Aires. Tornquist, Sierra de la Ventana, 17 Nov. 1972, *Gómez et al.* (ICN). **BRAZIL. Rio Grande do Sul.** Bagé, estrada para o Passo do Viola, entre a Estância Santa Genoveva e São Francisco, 3 Oct. 1985, *Girardi-Deiro et al.* (CNPO). **CHILE. Colchagua.** Picholemu, 31 Oct. 1967, *Montero 8041* (CONC). **Colleco.** Angol, Deneo - coloaur camino a Angol, 6 Nov. 1977, *Montero 10707* (CONC). **Concepción.** Cerros de La Toma, 12 Oct. 1976, *Fister*

(CONC). **Ñuble**. Camino a San Nicolas, 5 Nov. 1961, *Matthei* 263 (CONC). **URUGUAY**. **Cerro Largo**. Rio Negro, Parcela 7- portera, estancia Palleros, 6 Dez. 1937, *Gallinal et al.* 1606 (BLA). **Florida**. Parcela S. Nicolás, Estancia San Pedro del Timote, 10 Dez. 1936, *Gallinal et al.* 3142 (BLA). **Maldonado**. Ruta 29, Aiguia a San Carlos, 12 Out. 1965, *Rosengurtt et al.* 9797 (BLA). **Soriano**. Estancia Monzón-Heber, Estacion Jackson, Dec. 1937, *Gallinal et al.* 7956 (BLA). **Tacuarembó**. Clanqueal, pasando A. Jaguarí, 7 Out. 1961, *del Puerto* 217 (BLA). **Lavalleja**. Minas, Parque UTE, 6 Nov. 1955, *Rosengurtt B-* 6366 (BLA, P). **Montevideo**. 152 km na saída para Punta del Este, 13 Feb. 1977, *Sampaio* 662, 673 (ICN).

HABITAT. Open grasslands, dry and wet soils, more frequent in rocky grasslands.

ETIMOLOGY. The epithet referred to the similarity of the species with the genus *Briza*.

7. Chascolytrum bulbosum (*Parodi*) *Essi, Longhi-Wagner & Souza-Chies* **comb. nov.**

Type: Uruguay: Minas: cerro Arequita, frecuente en pedregal arbustivo y pratense, 15 Oct 1939, *B. Rosengurtt* 3154 (holotype BAA n.v.)

Erianthecium bulbosum Parodi in *Notas Mus. La Plata, Bot.* 8: 77, f.1 (1943)

Plants 30 - 45 cm high. Basal internodes of the culms thickened, bulbous. Basal innovations extravaginal. Leaf-sheaths glabrous or pilose, smooth, margins non-overlapping. Leaf-blades linear to linear-lanceolate, 4.5 - 20 cm x 2.8 - 5 mm, flat, pilose on both surfaces. Ligule 1.8 - 4 mm, truncate to acute.

Panicle sub-contracted or contracted, erect, 3 - 12 cm long. Pedicels smooth. Spikelets 7 - 8 x 2.5 - 4 mm, 3 - 11-flowered, sub-cylindrical to laterally compressed, lanceolate. Florets imbricate, obscuring the rhachilla. Glumes chartaceous, convex, glabrous on the surface and ciliate at the margins, subequal or unequal, acute; lower glume 3.8 - 4.3 x 1 - 2 mm, 3 - 5-nerved; upper glume 4 - 5 x 1.3 - 3 mm, 7-nerved. Lemmas chartaceous, slightly laterally compressed, pale to pale green, without a gibbous back, margins non-distinct from the back, non-cordate at the base, 7 - 9-nerved, midvein non-salient on the back, surface pubescent, bidentate at the apex, awned, awn 1 - 3 mm, margins non-inrolled at the base, without oil glands; lower lemma 4 - 5.7 x 3.5 - 4 mm. Paleas elliptic, truncate or lobed, chartaceous, pubescent between the keels, keels ciliate; lower palea 3 - 3.5 x 1.3 mm. Lodicules flabelliform. Stamens 3. Caryopsis elliptic, concavo-convex. Hilum punctiform.

Fig. 11.A-D.

DISTRIBUTION. Southern Brazil, also Uruguay, according to Parodi (1943). Map 10.

BRAZIL. Rio Grande do Sul. Canguçu, estrada de terra no Bairro Teixeira, 14 Nov. 1975, *Longhi et al.* 182 (ICN); Pelotas, Campus IPEAS-UFPEL 13 Nov. 1972, *Felisbino et al.* 35 (ICN); *Ibidem*, 9 Oct. 1972, *Luz et al.* 37 (ICN); Pelotas, Instituto Agrônômico do Sul, 16 Oct. 1962, *Kappel & Froner* (BLA 2364); *Ibidem*, 5 Oct. 1954, *Sacco* (PACA 63392); Pelotas, Instituto de Agricultura, 25 Aug. 1952, *Beetle* 2226 (PACA); Pinheiro Machado, BR-293- Bagé, 15 km antes do Arroio Lajeado, 15 Nov. 1975, *Longhi et al.* 224 (ICN); Pinheiro Machado, Granja Pedras Altas, 22 Oct. 1957, *Capparelli* 908 (BLA); Piratini, logo após Ponte do Arroio Piratini Menor, 15 Nov. 1975, *Longhi et al.* 208, 209 and 212 (ICN); *Ibidem*, 15 Nov. 1975, *Sampaio et al.* 49 (ICN); Piratini, beira do Arroio

Piratini Menor, após ponte saindo de Piratini, 11 Oct. 2004, *Longhi-Wagner & Essi* 9292 (ICN); Piratini, BR 392, morro ao lado do rio, 11 Oct. 2004, *Essi & Longhi-Wagner* 284 (ICN); Piratini, estrada Canguçu a Piratini, 52 km após Canguçu, 11 Oct. 1972, *Valls et al.* 2239 (ICN); Piratini, Fazenda Santa Fé, 4 Oct. 2005, *Guglieri et al.* 641a (ICN); Piratini, morro em frente da ponte do Arroio Piratini, próximo a gruta com Santa, 16 Oct. 2003, *Essi et al.* 50 (ICN); Piratini, morro em frente da ponte do Arroio Piratini, próximo a gruta com Santa, 16 Oct. 2003, *Essi et al.* 60 (ICN).

HABITAT. Grasslands and rocky slopes.

ETIMOLOGY. The epithet refers to the culms of the species, with the basal internodes thickened, bulbous.

8. *Chascolytrum calothea* (Trin.) Essi, Longhi-Wagner & Souza-Chies comb. nov. Type:

V. spp. Bras. Sto. Paulo, in fruticeis humidis prope Jundiahy, Riedel (holotype LE n.v.; isotypes LE n.v., US n.v.)

Eragrostis calothea Trin. in Mem. Acad. Imp. Sci. St. Petersburg, Ser. 6, Sci. Math. 1 (4): 414 (1830)

Poa calothea (Trin.) Kunth (1833: 340)

Briza calothea (Trin.) Hack. (1904: 282)

Poidium calothea (Trin.) Matthei (1975: 116)

Briza poa Nees (1855: 283) Type: Brasilia, *Sellow* (lectotype CGE n.v. (designated by Matthei, 1975))

Briza neesii Döll (1878: 132) Type: Rio de Janeiro: St. Luiz im Serra dos Órgãos, *Glaziou* 1639 (lectotype W! (**designated here**))

Plants 36 - 150 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, smooth or scaberulous, margins non-overlapping.

Leaf-blades linear to linear-lanceolate, 10 - 50 cm x (1) 3 – 9.3 mm, flat, glabrous. Ligule 0.7 – 3.5 mm, obtuse.

Panicle open, rarely sub-contracted, pendulous, less frequently erect, 5 - 24 cm long.

Pedicels smooth. Spikelets 2.9 – 7.5 x 2 - 5 mm, 4 - 9-flowered, laterally compressed, elliptic. Florets imbricate, obscuring the rachilla. Glumes herbaceous, convex to navicular, glabrous, smooth, subequal; lower glume 1.8 – 3.6 x 0.7 – 1.1 mm, 3-nerved; upper glume 2.1 - 4 x 0.9 – 1.6 mm, 3 – 5 - 7-nerved. Lemmas chartaceous to coriaceous, laterally compressed, pale, with a slightly distinct gibbous back, distinct from the broad margins, cordate at the base, 5-nerved, midvein non-salient on the back, glabrous, smooth, acute, muticous, margins glabrous, non-inrolled at the base, without oil glands; lower lemma 2.3 – 3.8 x 1.3 – 2 mm. Paleas lanceolate, membranaceous, glabrous between the keels, keels ciliate; lower palea 1.5 – 2.5 x 0.6 – 1.1 mm. Lodicules linear. Stamens 1 - 3. Caryopsis elliptic, concavo-convex. Hilum elliptic.

Fig. 12.A-D.

DISTRIBUTION. Argentina, Brazil, Paraguay and Uruguay. Map 11.

SELECTED COLLECTIONS. ARGENTINA. Corrientes. Santa Rosa, 18 Nov. 87,

Quarín et al. 3891 (K). **BRAZIL. Minas Gerais.** Poço de Caldas, 5 Nov. 1980, *Garielli et*

al. 389 (UEC). **Paraná.** 5 km do trevo de início da estrada da Graciosa em direção a Morretes, 30 Oct. 89, *Pedralli et al.* 3008 (MO); Serra do Ibitiraquire, 24 Oct. 2000, *Cordeiro; Barbosa & Costa* 1779 (K); Serra do Itararé, município de Guaratuba, 24 Nov. 81, *Klein et al.* 12032 (B). **Rio de Janeiro.** Itatiaia, 12 Nov. 1975, *Camerik* 168 (BLA). **Rio Grande do Sul.** São José dos Ausentes, Monte Negro, 2 Dec. 2003, *Essi et al.* 154 (ICN); São José dos Ausentes, Serra da Rocinha, 11 Dec. 1996, *Longhi-Wagner* 3394 (ICN). **Santa Catarina.** São Joaquim, caminho entre a sede da Vila São Sebastião do Arvoredo e São Joaquim, 3 Dec. 2003, *Essi et al.* 184 (ICN). **São Paulo.** Campos do Jordão, Horto Municipal, 28 Oct. 1992, *Wasum et al.* (MO 4574399). **PARAGUAY.** **Caaguazú.** Caaguazu, Arroyo Guaranungua, 28 km S of Arroyo Yhu. Savanna, 1 Dec. 1990, *E. Zardini & C. Velásquez* 24664 (MO); **Guaira.** 15 km N of Tebicuary. Inundated Savanna, 16 Nov. 1990, *E. Zardini & C. Velásquez* (MO 3879835). **URUGUAY.** **Tacuarembó.** 6 Dec. 1957, *Rosengurtt* B-6919 (P).

HABITAT. Wet grasslands and swamps with *Sphagnum* spp., less common in dry grasslands.

NOTES.1. *Chascolytrum calotheca* is a polymorphic species, especially concerning the size of the spikelets, leaf-blades and height. The plants from populations that occur in swamps with different moss species (*Sphagnum* spp.), are much more robust and taller, up to 150 cm high, than the plants from dry grasslands. It is morphologically close to *C. juergensii*, *C. uniolae* and *C. ambiguum*, being distinguished from the former by its glabrous paleas (pilose in *C. juergensii*), from the second especially by its smooth lemmas (papillose on the back in *C. uniolae*), and from the last by its cordate lemmas with slightly

distinct gibbous back (non-cordate and lemmas without distinction between back and margins in *C. ambiguum*).

2. Although any specimen of the type collection of this species was examined, several specimens of *Eragrostis calotheca* Trin. identified by Riedel were analysed in different European herbaria.

3. Matthei (1975) designated a destroyed lectotype to *Briza neesii* Döll, indicating syntypes. Several syntypes were analysed at W herbarium, and one is designated here as a new lectotype.

9. *Chascolytrum itatiaiae* (Ekman) Essi, Longhi-Wagner & Souza-Chies **comb. nov.**

Type: Bras., Serra do Itatiaia, in monticulo Serra da Pedra Assentada, 2400 m.s.m, 19 May 1902, *Dusén 292*. (lectotype S! (designated by Matthei, 1975); isolectotype S! R n.v.)

Briza itatiaiae Ekman (1913: 58)

Poidium itatiaiae (Ekman) Nicora & Rúgolo (1981: 306)

Plants 55 - 75 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, smooth, margins non-overlapping. Leaf-blades linear, 16 - 40 cm x 3 - 4 mm, flat, glabrous. Ligule 1 - 2 mm, truncate.

Panicle open, erect, 15 - 20 cm long. Pedicels smooth. Spikelets 2.3 - 3 x 2.3 - 3.1 mm, 3 - 4-flowered, laterally compressed, ovate. Florets imbricate, obscuring the rachilla. Glumes herbaceous, naviculate, non-cordate at the base, glabrous, smooth or scaberulous towards the apex, subequal, acute; lower glume 1.1 - 3 x 0.4 - 0.7 mm, 3 (5)-nerved; upper glume 1.1 - 2.6 x 0.7 - 0.9 mm, 3 (5)-nerved. Lemmas herbaceous, laterally compressed, pale to purplish, without a gibbous back, margins non-distinct from the back, non-cordate at the

base, 5 - 7-nerved, midvein non-salient on the back, glabrous, smooth or scaberulous, obtuse or acute at the apex, mucicous, margins glabrous, non inrolled at the base, without oil glands; lower lemma 1.8 – 2.1 x 0.7 - 1 mm. Paleas elliptic-lanceolate, membranaceous, glabrous and smooth between the keels, keels ciliolate; lower palea 1.7 - 2 x 0.6 – 0.9 mm. Lodicules elliptic-lanceolate. Stamens 1 - 2. Caryopsis elliptic, concavo-convex. Hilum elliptic.

Fig. 13.A-D.

DISTRIBUTION. Brazil, in highlands of the States of Rio de Janeiro and São Paulo, in Southeastern Brazil, and a single collection in Santa Catarina. Map 6.

SELECTED MATERIAL. BRAZIL. Rio de Janeiro. Macieiras, Parque Nacional do Itatiaia, 9 Jan. 1981, *Longhi-Wagner et al. 1548* (ICN); Itatiaia, Agulhas Negras, 2 Aug. 1977, *Winge 1039, 1041, 1042, 1045* (ICN); Prateleira, 2 Aug. 1977, *Winge 1079* (ICN); Macieiras, Parque Nacional do Itatiaia, 9 Jan. 1981, *Winge et al. 1547* (ICN); Itatiaia, Parque Nacional, 16 April 1967, *Lindeman & Haas 5173* (MBM, K); Itatiaia, March 1937, *Brade 15622* (RB); Itatiaia, Planalto 2200 – 2400 m, 6 March 1962, *Pereira 7059* (MO); Itatiaia, próx. Prateleiras, 13 March 1960, *Castellanos 22674, 22575* (GUA). **Santa Catarina.** Urubici, Morro da Igreja, 22 Jan. 1997, *Longhi-Wagner et al. 3602* (ICN).

HABITAT. Grasslands in high altitude.

ETIMOLOGY. The epithet refers to the region of “Itatiaia”, in Rio de Janeiro State, Brazil, where the holotype was collected.

10. Chascolytrum juergensii (Hack.) Essi, Longhi-Wagner & Souza-Chies **comb. nov.**

Type: Rio Grande do Sul: Pinheiral, Município Rio Pardo, C. Jürgens (holotype W!)

Briza juergensii Hack. in Verh. K. K. Zool. Bot. Ges. Wien 65: 76 (1915)

Poidium juergensii (Hack.) Matthei (1975: 114)

Plants 28 - 150 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, rarely pilose, margins non-overlapping. Leaf-blades linear-lanceolate, 7 - 60 cm x 2 - 7 mm, flat, glabrous or pilose on the abaxial surface.

Ligule 1 - 4.5 mm, obtuse to truncate.

Panicle open, erect or pendulous, 4.5 - 33 cm long. Pedicels smooth. Spikelets 3 - 6 x 2 - 4.5 mm, 4 - 9-flowered, sub-cylindrical to laterally compressed, elliptic-lanceolate. Florets imbricate, obscuring the rhachilla, or loosely imbricate, rhachilla aparent. Glumes herbaceous, convex to naviculate, glabrous, smooth, subequal; lower glume 1.7 - 3 x 0.5 - 1.1 mm, 3-nerved; upper glume 2 - 3.3 x 0.9 - 1.2 mm, 3 - 5-nerved. Lemmas chartaceous or coriaceous, laterally compressed or dorsi-ventrally compressed, pale, with a slightly distinct gibbous back or without a gibbous back, margins broad or narrow, distinct or not from the back, 5-nerved, midvein non-salient on the back, pilose, occasionally glabrous, acute at the apex, muticous, margins glabrous or sparsely pilose, inrolled or not at the base, without oil glands; lower lemma 2.5 - 3.9 x 1 - 1.8 mm. Paleas lanceolate, membranaceous, pilose between the keels, keels ciliate; lower palea 1.5 - 2.5 x 0.7 - 1 mm. Lodicules linear-lanceolate. Stamens 1 - 3. Caryopsis elliptic, concavo-convex. Hilum elliptic.

Fig. 14.A-J.

Key to varieties:

1 - Lemmas wide (1.4 – 1.8 mm), with a gibbous back, slightly distinct from the margins, surface pilose or glabrous.....*C. juergensii* var. *juergensii*

1' – Lemmas narrow (1 –1.3 mm) ,without a distinction between back and margins, surface pilose.....*C. juergensii* var. *angustilemma*

Chascolytrum juergensii (Hack.) Essi, Longhi-Wagner & Souza-Chies var. **juergensii**

DISTRIBUTION. Brazil and Colombia. Map 12.

SELECTED COLLECTIONS. COLOMBIA. Antioquia. San Pedro, km 13 of road San Pedro – Entrerriós, 28 Sept. 1987, *Zarucchi et al.* 5870 (MO). **BRAZIL. Minas Gerais.** Camanducaia, Serra da Mantiqueira, Vila Monte Verde, 26 Nov. 2000, *Windisch et al.* 9627 (ICN); Monte Verde, *Longhi-Wagner & Witten* 5005 (ICN). **Paraná.** Contenda, Rodovia do Xisto, 22 Oct. 1967, *Hatschbach* 17553 (MBM, K); General Carneiro, Iratim, 6 Dec. 1971, *Hatschbach* 28326 (MBM, K); Piraquara, Loc. Borda do Campo, Passo do Cercado, 30 Oct. 1949, *Hatschbach* (MBM 25810). **Rio Grande do Sul.** Cristal, divisa com São Lourenço, Km 349.6, 16 Nov. 2003, *Essi et al.* 41 (ICN); Pelotas, Instituto Agronômico do Sul, 27 Oct. 1945, *Swallen* 7195 (NY, MO); São Leopoldo, Oct. 1942, *Leite* 3080 (NY). **Santa Catarina.** Florianópolis, antiga estrada p/o Balneário Daniela, Ilha de Santa Catarina, 2 Oct. 1987, *Souza & Araújo* 1151 (MBM); Urubici, 4 Dec. 2003, *Essi et al.* 195, 197 (ICN). **São Paulo.** Itatiaia, Planalto de Agulhas Negras, perto do

Abrigo Rebouças, 24 Nov. 1999, *Longhi-Wagner et al. 7204* (ICN); Pindamonhangaba, divisa com Campos do Jordão, 9 Dec. 1995, *Longhi-Wagner & Witten 2802* (ICN).

HABITAT. Open grasslands, wet soils.

ETIMOLOGY. The epithet is dedicated to Jürgens, botanist and collector of the holotype of the species.

Chascolytrum juergensii var. angustilemma *Essi, Longhi-Wagner & Souza-Chies*, unpublished

DISTRIBUTION. Brazil, mainly Southern Brazil, in Rio Grande do Sul and Santa Catarina States, with only one record for Minas Gerais State and another for São Paulo State. Map 12.

BRAZIL. Minas Gerais. Camanducaia, Monte Verde, 22 Jan. 2002, *Longhi-Wagner 8009* (ICN). **Rio Grande do Sul.** Bom Jesus - sede - 300m após entroncamento p/ Vacaria, 29 Nov. 1975, *Sampaio et al. 102* (ICN); Bom Jesus, Aparados da Serra, Dec. 1954, *Barreto* (BLA); Cambará do Sul - Itaimbezinho, Bela Vista, 1 Dec. 1981, *Sacchet 28* (ICN); Cambará do Sul, Fortaleza dos Aparados, 28 Nov. 1975, *Longhi et al. 315, 325, 328* (ICN); Cambará do Sul, Itaimbezinho, 1 Dec. 1981, *Longhi-Wagner et al. 957, 959, 960* (ICN); Cambará do Sul, Itaimbezinho, 15 Jan. 1979, *Lemos & Sampaio 33, 36* (ICN); Cambará do Sul, Itaimbezinho, 28 Nov. 1975, *Sampaio et al. 58, 59, 61, 62* (ICN); Cambará do Sul, Itaimbezinho, 4 Jan. 1974, *Hickenbick 92* (ICN); Cambará do Sul,

Itaimbezinho, 7 Jan. 1977, *Sampaio et al.* 406, 411 (ICN); Cambará do Sul, Itaimbezinho, Bela Vista, 1 Dec. 1981, *Longhi-Wagner et al.* 949 (ICN); Cambará do Sul, Parque Nacional da Serra Geral, S29°03'43.9" W049°57'23.7", 1 Dec. 2003, *Essi et al.* 135, 136, 139 (ICN); Cambará do Sul, a caminho do Parque, em basalto com *Sphagnum* sp., na beira da estrada, 17 Nov. 2004, *Essi*, 295 (ICN); Itaimbezinho, 100 m p. S. Francisco de Paula, in campestribus subumidis, 20 Feb. 1953, *Rambo* 54022 (B); Itaimbezinho - São Francisco de Paula, 20 Feb. 1953, *Rambo* (PACA); São José dos Ausentes, Monte Negro, S28°37'00.6" W049°47'42.1", 2 Dec. 2003, *Essi et al.* 152 (ICN). **Santa Catarina.** Água Doce, 6 Nov. 1971, *Smith et al.* 15700 (HBR); Bom Jardim, Curral Falso, 19 Nov. 1959, *Reitz & Klein* 8405 (HBR); Bom Jardim, Serra do Oratório, 9 Dec. 1958, *Reitz & Klein* 7642 (HBR, HB); Caçador, 2 Dec. 1964, *Smith & Klein* 13367 (HBR, MO, S); Campo dos Padres, 1700 m. In subhumidis graminosis, 22 Jan. 1957, *Rambo* (B 1001176910); Lauro Müller, a 25 km passando la Serra do Rio do Rastro, 7 Dec. 1992, *Rúgolo et al.* 1478 (MO); Porto União, 27 Oct. 1962, *Reitz & Klein* 13670 (HBR); Porto União, S. Miguel, 29 Dec. 1936, *Rambo* (HBR 2643); Serra do Oratório, Bom Jardim campo alt.1400 m, 9 Dec. 1958, *Reitz & Klein* 7642 (L). **São Paulo.** Campos do Jordão, 9 Dec. 1995, *Longhi-Wagner & Witten* 2793 (ICN).

HABITAT. Open grasslands in high altitude, in wet and rocky soils, and in *Araucaria angustifolia* Kuntze forests.

ETIMOLOGY. The epithet *angustilemma* refers to the narrow lemmas, typical of this variety.

11. Chascolytrum koelerioides (*Trin.*) *Essi, Longhi-Wagner & Souza-Chies comb. nov.*

Syntypes: Chile: Quillota: in pascuis sylvaticis collium editiorum, Oct. 1829, *Bertero 947* (syntype LE n.v.; isosyntype P!); Chile, *Cuming dl. amic. [Trinius] Prescott 1832* (syntype LE n.v.)

Poa koelerioides Trin. in *Mem. Acad. Imp. Sci. Saint-Petersbourg, Ser. 6, Sci. Math., Seconde Pt. Sci. Nat.* 4, 2 (1): 62. (1836)

Gymnachne koelerioides (Trin.) Parodi (1962: 19)

Poa tenuiculmis Steud. (1854: 259), nom. illeg. superfl.

Rhombolytrum berteroanum E. Desv. (1854: 387) Type: Chile: Quillota, in parcuis sylvaticis colium editivum, *Bertero 947* (lectotype P! (**lectotype designated here**))

Gymnachne jaffuelii Parodi (1938: 30) Type: Chile: Concepción, *Jaffuel 1884* (holotype BAA n.v.)

Plants 43 - 78 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, margins non-overlapping. Leaf-blades linear, 13 - 35 cm x 1 - 4 mm, flat or convolute, glabrous. Ligule 1 – 1.5 mm, truncate.

Panicle contracted, erect, 7 - 11 cm long. Pedicels scabrous. Spikelets 4.2 – 7 x 1.5 – 2.2 mm, 4 - 6-flowered, laterally compressed, rhomboid. Florets imbricate, obscuring the rachilla. Glumes chartaceous, convex to navicular, scabrous, equal to subequal; lower glume 3 – 3.2 x 0.4 – 1.3 mm, 3-nerved; upper glume 3 – 3.2 x 0.4 – 1.3 mm, 3-nerved.

Lemmas herbaceous to chartaceous, dorsi-ventrally compressed or slightly laterally compressed, pale to pale green, without a gibbous back, margins non-distinct from the back, non-cordate at the base, 3 (5)-nerved, midvein non-salient on the back, glabrous, scaberulous, obtuse or bilobate at the apex, muticous or mucronate, margins glabrous,

smooth or scaberulous, non-inrolled at the base, without oil glands; lower lemma 2.6 – 3.2 x 0.6 – 0.9 mm. Paleas lanceolate to linear-lanceolate, membranaceous, glabrous and smooth between the keels, keels ciliate; lower palea 2.9 – 3.1 x 0.5 – 0.6 mm. Lodicules lanceolate. Stamens 1. Caryopsis oblong to elliptic, concavo-convex. Hilum punctiform. Fig. 15.A-D.

DISTRIBUTION. Central Chile. Map 13.

SELECTED COLLECTIONS. CHILE. Arauco. Boca Lebu Norte, 22 Nov. 2000, *Baeza & Rodriguez 1904* (CONC). **Concepción.** Ramuncho, 3 Dec. 1960, *Matthei 158* (CONC). **Countin.** Cerro Nielol, Temuco, 16 Dec. 1941, *Montero 4516* (CONC). **Linares.** Camino entre Chanco y Constitución, 11 Jan. 1983, *Matthei y Bustos 24* (B). **Quillota.** Valparaíso, 1832, *Cuming 464* (BM); Puerta Ocoa, Cerro La Campana, 23 Nov. 1986, *Zoellner 13137* (CONC). **Valdivia.** S. Juan, Feb. 1887, *Philippi* (BM); Valdivia, 1861, *Philippi* (K). **San Fernando,** Termas del Flaco, 17 Nov. 1996, *Ryves & Clement 96CE/167* (K).

HABITAT. Open grasslands.

12. Chascolytrum lamarckianum (*Nees*) *Matthei* (1975: 74) Type: Brazil: Rio Grande do Sul, in finibus regni Paraguayani, *Sellow*. (lectotype L n.v. (designated by *Matthei*, 1975); isolectotypes BAA n.v., LE n.v., US n.v.)
Briza lamarckiana *Nees* in *Agrost. Bras. Enumeratio Plantarum* 2(1): 481 (1829)

Briza lindmanii Ekman (1913: 54) Type: Bras. Civ. Rio Grande do Sul, Porto Alegre, in fruticeis, locis graminosis minus apricis, 21 Sept. 1892, Exp. 1. Regn. A275 ½, *Lindman* (holotype S!)

Briza subaristata var. *fusca* Parodi (1920: 127) Type: Uruguay, campos, Nov. 1903, *Arechavaleta* (holotype MVM n.v.)

Briza fusca (Parodi) Parodi (1922: 93)

Plants 20 - 100 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous or sparsely pilose, margins non-overlapping. Leaf-blades linear, 6.5 - 39 cm x 1.6 - 4.8 mm, flat, glabrous. Ligule 0.4 - 1.7 mm, truncate. Panicle open, pendulous, 3 - 25 cm long. Pedicels smooth. Spikelets 3.2 - 5 x 1.9 - 3.5 mm, 3 - 6-flowered, cylindrical, oblong. Florets imbricate, obscuring the rachilla. Glumes herbaceous, convex, glabrous, smooth or scaberulous, equal to subequal; lower glume 1.7 x 0.8 - 1.5 mm, (3) 5 - 7-nerved; upper glume 2.3 - 3.2 x 1 - 1.5 mm, (5) 7-nerved. Lemmas coriaceous, dorsi-ventrally compressed, with a rufous, less frequently yellow-gold gibbous back, strongly distinct from the broad margins, cordate at the base, 5-nerved, midvein non-salient on the back, glabrous, smooth, rarely pilose on the back, acute at the apex, muticous or mucronate, margins glabrous, non-inrolled at the base, without oil glands; lower lemma 2.4 - 3.4 x 1.3 - 2.1 mm. Paleas elliptic to orbicular, coriaceous, glabrous and smooth between the keels, rarely pilose, keels glabrous; lower palea 1.2 - 2 x 0.9 - 1.4 mm. Lodicules flabelliform. Stamens 1 - 3. Caryopsis sub-orbicular, plano-convex. Hilum punctiform or elliptic.

Fig. 16.A-D.

DISTRIBUTION. Argentina, Southern Brazil and Uruguay. Map 14.

SELECTED COLLECTIONS. ARGENTINA. Entre Rios. Federacion Santa Ana, 5 Oct. 1978, *Renvoize, Wilmot-Dear & Zuloaga 2940* (K). **BRAZIL. Rio Grande do Sul.** Soledade, BR 386, km 259, 31 Oct. 1993, *Boechat* (ICN). **Santa Catarina.** Pericó, SC 430, 7 Dec. 1992, *Rúgolo et al. 1510* (ICN). **Paraná.** Guarapuava, Jan. 2004, *Essi & Longhi-Wagner 260* (MBM). **URUGUAY. Maldonado.** Abra de Perdomo, 21 Nov. 1948, *Rosengurtt 5259* (ICN); **Rocha.** Parque Santa Teresa, 17 Dec. 1969, *Rosengurtt 11139* (MSB); **Tacuarembó.** Arenal Granja Municipal, 18 Oct. 1952, *Rosengurtt B-6225* (P).

HABITAT. Open grasslands and road sides. Wet soils.

ETIMOLOGY. The epithet dedicated to Lamarck, naturalist and botanist.

13. *Chascolytrum latifolium* *Essi, Longhi-Wagner & Souza-Chies*, unpublished.

Plants 27 - 87 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, margins non-overlapping. Leaf-blades linear-lanceolate, 7.5 - 40 cm x (4) 7 - 12 mm, flat, glabrous. Ligule 1.5 - 4 mm, truncate. Panicle open, pendulous, 9 - 15 cm long. Pedicels scabrous. Spikelets 5 - 6 x (1.5) 4 - 5.1 mm, 4 - 8-flowered, subcylindrical, oblong. Florets imbricate, obscuring the rhachilla. Glumes herbaceous, convex to naviculate, non-cordate at the base, glabrous, smooth, subequal; lower glume 3.2 - 3.9 x 0.6 - 1.5 mm, 3-nerved; upper glume 3.2 - 4 x 1.1 - 1.5 mm, 3 - 5-nerved. Lemmas herbaceous, slightly laterally compressed, without a gibbous

back, with broad margins non-distinct from the back, non-cordate at the base, 7-nerved, midvein non-salient on the back, glabrous, acute, truncate or bidentate at the apex, mucronate, margins glabrous, smooth or scaberulous at the apex, non-inrolled at the base, without oil glands; lower lemma 3.9 – 4 x 1.4 – 2.5 mm. Paleas elliptic-lanceolate, membranaceous, glabrous and smooth between the keels, keels ciliate; lower palea 2.8 – 3.2 x 1.1 – 1.7 mm. Lodicules linear. Stamen 1. Caryopsis sub-orbicular, plano-convex. Hilum elliptic.

Fig. 17.A-D.

DISTRIBUTION. Brazil, in highlands of Rio Grande do Sul and Santa Catarina States, between 800 - 1580 m. Map 15.

BRAZIL. Rio Grande do Sul. São José dos Ausentes, Monte Negro: na beira de lageado, próximo a mata de Araucária e de ponte de madeira, na descida do Monte Negro, 2 Dec. 2003, *Essi et al. 159* (ICN). **Santa Catarina.** Bom Retiro, Campo dos Padres, 16 Dec. 1948, *Reitz 2389* (HBR); Bom Retiro, Campo dos Padres, 18 Dec. 1948, *Reitz 2553* (HBR, S); Bom Retiro, Campo dos Padres, 23 Jan. 1957, *Smith & Reitz 10311* (HBR, RB); Bom Retiro, Campo dos Padres, between Fazenda Campo dos Padres and Fazenda Santo Antônio, 21 Nov. 1956, *Smith & Klein 7804* (HBR, NY); São Joaquim [Urubici] Morro da Igreja, Fazenda Morrinhos, Campestre do Malacara, 22 Jan. 1960, *Mattos 7448* (BLA); São Joaquim, Campestre do Malacara, Faz. de Morrinhos, 22 Jan. 1960, *Mattos 8462* (HAS); Parque Nacional São Joaquim, 2001, *Longhi-Wagner & Garcia 7377* (ICN); Urubici, Fazenda Arno Philippi, 7 Dec. 2006, *Longhi-Wagner et al. 10228, 10229, 10230* (ICN);

Urubici, Morro da Igreja: em frente à Cavalgada, próximo do quartel, 4 Dec. 2003, *Essi et al.* 201 (ICN).

HABITAT. Open grasslands with wet soils and swamps, usually associated with *Sphagnum* spp.

ETIMOLOGY. The epithet refers to the remarkable wide leaf-blades of the species, easily to recognize in the field.

14. Chascolytrum monandrum (*Hack.*) *Essi, Longhi-Wagner & Souza-Chies comb. nov.*

Type: prope Cutervo, *Jelski 402* (holotype W!; isotypes W! US n.v.)

Poa monandra Hack. in Oesterr. Bot. Z. 52(10): 376 (1902)

Briza monandra (Hack.) Pilg. (1929: 725)

Poidium monandrum (Hack.) Matthei (1975: 103)

Calotheca stricta var. *mandoniana* Griseb. (1879: 289) Type: Bolivia, *Mandon* [1356]

(holotype GOET!; isotypes G! GH n.v. L n.v. LE n.v. S n.v. P! W!)

Briza mandoniana (Griseb.) Henrard (1921: 70)

Briza lilloi Parodi (1920: 133) Type: Argentina, Tucuruan, La Cienaga, 1905, *Lillo 4094*.

(holotype: BAA n.v.)

Briza mandoniana var. *herzogiana* Henrard (1921: 71) Type: Auf Alpenwiesen bei

Choquetanga grande, Oct. 1911, *Herzog 3017* (holotype L! isotype L!)

Briza mandoniana var. *tuberculata* Henrard (1921: 71) Type: In vicinis Sorata, cerro del

Iminapi in humo pingui, Jan-March 1860, *Mandon 1355* (holotype L!; isotypes BM! G!

NY, S!, K! P! W!)

Briza mandoniana var. *vallegrandensis* Henrard (1921: 71) Type: In subalpinen Gebüsch des Berges über Vallegrande, March 1911, *Herzog 1879* (holotype L!; isotype L! S!)

Plants 10 - 65 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous or pilose, margins non-overlapping. Leaf-blades linear, 7 - 22 cm x 1 - 3 mm, flat or convolute, glabrous, pilose on the adaxial surface, or pilose on both surfaces. Ligule 0.6 – 2.2 mm, truncate.

Panicle open, erect, 3.5 - 18 cm. Pedicels scaberulous. Spikelets 3 - 6 x 1 – 3.5 mm, 3 - 6-flowered, laterally compressed, elliptic-lanceolate. Florets imbricate, obscuring the rachilla. Glumes herbaceous, convex to naviculate, glabrous, scaberulous or smooth, subequal; lower glume 1.5 - 3 x 0.5 – 0.8 mm, 3-nerved; upper glume 1.5 - 3 x 0.4 – 0.9 mm, 3-nerved. Lemmas herbaceous, laterally compressed, pale, purplish or pale green, without a gibbous back, margins broad or narrow, non-distinct from the back, non-cordate at the base, 5-nerved, midvein non-salient on the back, glabrous, smooth or tuberculate, obtuse at the apex, muticous, margins ciliate at the lower third, trichomes longer than 0.3 mm, inrolled or not at the base, without oil glands; lower lemma 2 – 2.3 x 0.7 – 0.9 mm. Paleas elliptic to elliptic-lanceolate, membranaceous, glabrous, smooth or sparsely punctuated between the keels, keels ciliate; lower palea 1.5 – 2 x 0.4 – 0.7 mm. Lodicules lanceolate. Stamens 1 (3). Caryopsis elliptic, concavo-convex. Hilum elliptic or punctiform.

Fig. 18.A-D.

DISTRIBUTION. Argentina, Bolivia, Brazil, Colombia, Ecuador and Peru. Map 16.

SELECTED COLLECTIONS. ARGENTINA. Tucumán. La Quenoa [San Javier], 2 April 1912, *Rodriguez 508* (P). **BOLIVIA. La Paz.** Nor Yungas, c. 3-4 km above Unduavi on road to La Paz/El Cumbre, 3 May 1997, *Wood 12049* (LPB). **Santa Cruz.** Vallegrande, on road to Masucuri, 11 Feb. 1996, *Wood 10602* (LPB). **Cochabamba.** Ayopaya, c. 1 km above Independencia, 11 March 2000, *Wood 16013* (LPB). **Tarija.** Cuesta Sama, 28 Feb. 1979, *Coro 1275* (LPB). **Larecaja.** Larecaja, Vicinis Sorata, Cerro del Iminapi, in humo pingui 2650 m, Jan.-March 1860, *Mandon 1355* (L). **Murillo.** La Paz, 131 km al este (debajo) de La Cumbre, por el camino a Unduavi, 1 May 1988, *Solomon 18307* (LPB).

Brazil. Paraná. Horizonte (Palmas), 24 Nov. 1981, *Hatschbach & Hirzinker 44415* (ICN). **Rio Grande do Sul.** Bom Jesus, perto da Serra da Rocinha, 29 Nov. 1977 *Mattos 17815* (HAS). **Santa Catarina.** Água Boa, Nov.-Dec. 1987, *Valls et al.* 11499 (ICN).

COLOMBIA. Grap. the flat cerntry near Laicto, 1846-69, *Jamerson 779* (BM).

EQUADOR. Chimborazo, Urbina, Mt. Chimborazo, 27 July 1939, *Asplund* (P).

Pichincha. Quito, Panecillo, 15 May 1939, *Asplund, 6048* (G, K). **Chimborazo.** from camp site above Rio Alao, 20 May 1990, *Peterson et al. 9193* (K). Paramo, grassland, Totarococha Mazan valley, 12 Sept. 1987, *Ramsay & Merrow-Smith 543* (K). **Imbabura.** Near Lago Cuicocha, 14 March 1988, *Lalgaard & Renvoize 70874* (K). **PERU.**

Paucartambo. Cuzco, entre Sunchubamba y Challabamba, 12 April 1967, *Vargas 19261* (B). **Ancash.** Carlos Fermin Fitzcarrold, Cordillera Blanca, 6 km SE of San Luiz, 23 March 1997, *Peterson & Rodriguez 13890* (K). **Cajamarca.** Hualgayoc, 4 km NE of Hualgayoc on HWY 3N towards Bambamarca, 17 March 2000, *Peterson & Rodriguez 14943* (K); Loja, 02 Juny 1990, *Peterson & Judziewicz 9460* (MO). **Chota.** 19 March 2000, *Peterson & Rodriguez 14978* (MO); **Sihaus.** 24 March 1997, *Peterson & Rodriguez*

13902 (MO); Carlos Fermin Fitzcarrold, 23 March 1997, *Peterson & Rodriguez 13890* (MO).

HABITAT. Open grasslands in high altitude.

ETIMOLOGY. The epithet refers to the presence of a single stamen, mentioned in the protologue of the species.

NOTE. *Chascolytrum monandrum* is a very polymorphic species, especially concerning the spikelets size and lemma width. The Bolivian specimens present a wider spectrum of variation. In the populations from Southern Brazil, the plants present smaller spikelets and narrower lemmas.

15. *Chascolytrum paleopiliferum* (Parodi) Matthei (1975: 88) Type: Argentina, Catamarca, El Rodeo, Jan. 1910, *Castillon* (lectotype LIL n.v. (designated by Matthei, 1975))

Briza paleopilifera Parodi (1920: 124)

Plants 15 - 60 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, margins non-overlapping. Leaf-blades linear to linear-lanceolate, 6 - 25 cm x 1 - 3 mm, flat or convolute, glabrous, margins scaberulous. Ligule 1 - 4 mm, obtuse or acute.

Panicle contracted, erect, 5 - 10 cm long. Pedicels smooth. Spikelets 5 - 8 x 4 - 4.5 mm, 5 - 10-flowered, subcylindrical to laterally compressed, oblong. Florets imbricate, obscuring

the rhachilla. Glumes herbaceous, naviculate, glabrous, smooth, subequal; lower glume 3.2 - 5 x 1.5 - 2 mm, 3 - 5-nerved; upper glume 3.5 - 5 x 1.5 - 2 mm, 5 - 7-nerved. Lemmas chartaceous to coriaceous, dorsi-ventrally compressed, with a gibbous pale back, strongly distinct from the broad margins, cordate at the base, chartaceous to coriaceous, 5 - 7-nerved, midvein non-salient on the back, glabrous, rarely with 2 - 4 capitate trichomes around the callus, obtuse or bidentate at the apex, muticous or mucronate, margins glabrous, non-inrolled at the base, without oil glands; lower lemma 4.5 - 5 x 2 - 2.5 mm. Paleas elliptic-orbicular, coriaceous, with capitate trichomes between the keels, keels ciliate; lower palea 2 - 3 x 1 - 1.5 mm. Lodicules linear. Stamens 3. Caryopsis sub-orbicular, plano-convex. Hilum elliptic to linear-elliptic.

Fig. 19.A-D.

DISTRIBUTION. Western Argentina. Map 17.

SELECTED COLLECTIONS. ARGENTINA. Catamarca. S. Luis - Oyola [Oyolas], 19 Dec. 1909, *Stuckert 20800* (W). **Córdoba.** Copina, 7 Dec. 1958, *Rosengurtt B-7676* (ICN); Quebrada Calderón, Estancia Pampa de San Luis, 19 Dec. 1909, *Stuckert 21018* (G); **Punilla**, Alta Gracia, 1-4 Dec. 1926, *Parodi 7429* (W); Camino de Ascochinga a La Cumbre, Arroyo Tiu Mayú, a 6,8 km del limite, 9 April, *Anton 359* (CORD); Capilla del Monte, path up Cerro Uritorco 23 Jan. 1966, *Hawkes et al. 3292* (K); Los Cocos, 7 Dec. 1907, *Stuckert 17968* (W); Los Cocos, 1 Jan. 1908, *Stuckert 18396* (W); Los Cocos - San Estevan, 29 Dec. 1906, *Stuckert 16681* (G); Los Cocos por San Esteban, 29 Dec. 1906, *Stuckert 16687* (W); Subida de Pampa de La Ollada a P. de La Esquina, 10 Jan. 1970, *Cuti 4942* (ICN). **San Luis.** Pampas S. Luis, 20 Dec. 1909, *Stuckert 21018* (W).

HABITAT. In high altitude grasslands, common in rocky soils.

ETIMOLOGY. The epithet refers to the presence of trichomes between the keels of the paleas.

NOTES.1. Nicora & Rógolo de Agrasar (1981) cited *C. paleopiliferum* (under *Briza paleopilifera* Parodi) for Bolivia, without mentioning the vouchers. However, Renvoize (1998) did not cite this species for this area. We did not find any material of this species collected in Bolivia. It is possible that the citation by Nicora & Rógolo de Agrasar (1981) was based on erroneously identified specimens of *C. altiplanense* as this species also can present capitate trichomes between the keels of the palea.

2. Although the lectotype of this species was not examined, an old isosyntyple was analysed at Kew herbarium (Argentina, Sierra de Morro, 1913, *F. Pastore*).

16. Chascolytrum parodianum (*Roseng., Arrill. & Izag.*) *Matthei* (1975: 68) Type:

Uruguay, Dep. Maldonado, Abra de Perdomo, 21 Nov. 1948, *Rosengurt B 5273* (holotype MVFA n.v.; isotype K!)

Briza parodiana Roseng., Arrill. & Izag. (1968: 26)

Plants 50 - 80 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, smooth, margins non-overlapping. Leaf-blades linear, 12.5 – 25.5 cm x 0.5 – 2.5 mm, involute, glabrous. Ligule 1.5 – 5.5 mm, acute or truncate.

Panicle open, pendulous, 7.5 – 25.5 cm. Pedicels smooth. Spikelets 3.5 - 4 x 1.5 – 2.8 mm, 3 - 6-flowered, cylindrical to subcylindrical, elliptic-lanceolate. Florets imbricate, obscuring the rhachilla. Glumes chartaceous, convex, glabrous, scaberulous on the keels, subequal; lower glume 2 - 3 x 0.6 – 0.9 mm, 3-nerved; upper glume 2.2 - 3 x 0.8 – 1 mm, 3-nerved. Lemmas chartaceous to coriaceous, slightly laterally compressed or dorsiventrally compressed, pale to purplish, without a gibbous back, margins slightly distinct from the back, non-cordate at the base, 7-nerved, midvein non-salient on the back, glabrous, acute, muticous, margins glabrous, inrolled or not at the base, without oil glands; lower lemma 2.4 - 3 x 1.1 – 1.2 mm. Paleas obovate, chartaceous, glabrous and smooth between the keels, keels glabrous or ciliolate at the upper half; lower palea 1.3 – 1.9 x 0.7 - 1 mm. Lodicules elliptic-lanceolate. Stamens 1 - 2. Caryopsis elliptic, concavo-convex. Hilum elliptic.

Fig. 20.A-D.

DISTRIBUTION. Brazil and Uruguay. Map 18.

SELECTED COLLECTIONS. BRAZIL. Rio Grande do Sul. 11 Oct. 2004, *Essi & Longhi-Wagner* 290 (ICN); 11 Oct. 2004, *Essi & Longhi-Wagner* 291 (ICN); 11 Oct. 2004, *Essi & Longhi-Wagner* 292 (ICN); 11 Oct. 1972, *Valls et al.* 2254 (ICN). **URUGUAY.** Lavalleja, 30 Dec. 1949, *Rosengurtt* 5654 (BLA); Maldonado, Nov. 1923, *Montoro* 2802 (BLA); 6 Nov. 1955, *Rosengurtt* B-6357 (ICN, P, BLA); 1816-1821, *Saint-Hilaire* (P); 21 Nov. 1948, *Rosengurtt* B-5273 (K); *Saint-Hilaire* (P).

HABITAT. Rocky grasslands and outcrops.

ETIMOLOGY. The epithet is dedicated to Parodi, Argentinean botanist.

17. Chascolytrum poaemorphum (*J. Presl*) *Essi, Longhi-Wagner & Souza-Chies comb.*

nov. Type: Hab. in montanis Peruviae huanoccensibus, *Haenke* (holotype PR!; isotypes B, BAA, US n.v.)

Panicum poaemorphum *J. Presl* in *Reliq. Haenk.* 1 (4 – 5): 282 (1830)

Briza poaemorpha (*J. Presl*) *Henrard* [*Briza* “*poimorpha*”] (1921: 36)

Poidium poaemorphum (*J. Presl*) *Matthei* (1975: 108)

Microbriza poaemorpha (*J. Presl*) *Parodi ex Nicora & Rúgolo* (1981: 295)

Isachne hackelii *Lindm.* (1900: 11) Type: Bras. Civ. Rio Grande do Sul: Porto Alegre,

Parthenon, *Lindman Exp. I. Regn. A595* (holotype W n.v.)

Briza hackelii (*Lindm.*) *Ekman* (1913: 61)

Briza hackelii f. *pseudisachne* *Ekman* (1913: 61) Type: Bras. Civ. Rio Grande do Sul: Rio Grande do Sul oppidum in campo arenoso loco subumido, 7 Nov. 1901, *Malme Exp. Regn. 250* (holotype S!)

Plants 44 - 100 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, smooth or scaberulous, margins non-overlapping.

Leaf-blades linear, 6.5 – 21.5 cm x 0.8 – 4.8 mm, flat, glabrous, rarely pilose on the abaxial surface. Ligule 1.2 – 5.2 mm, acute.

Panicle open, erect, 7 – 15.5 cm long. Pedicels smooth. Spikelets 1.2 – 1.8 x 1 - 2 mm, 2 - 3-flowered, laterally compressed, ovate to orbicular. Florets imbricate, obscuring the rhachilla. Glumes herbaceous, convex, glabrous, smooth, subequal; lower glume 1 – 1.6 x

0.3 – 0.5 mm, 3-nerved; upper glume 1 – 1.7 x 0.4 – 0.6 mm, 3-nerved. Lemmas coriaceous, slightly laterally compressed, pale, without a gibbous back, margins non-distinct from the back, non-cordate at the base, 5-nerved, midvein non-salient on the back, glabrous, surface echinulose, obtuse at the apex, muticous, margins glabrous and echinulose, laterally inrolled, without oil glands; lower lemma 1 – 1.5 x 0.5 – 0.7 mm. Paleas elliptic, coriaceous, glabrous and echinulose between the keels, keels glabrous or ciliolate; lower palea 0.7 – 1.2 x 0.4 – 0.6 mm. Lodicules linear. Stamens 1. Caryopsis elliptic, concavo-convex. Hilum punctiform or elliptic.

Fig. 21.A-E.

DISTRIBUTION. Argentina, Brazil, Paraguay and Uruguay. Map 19.

SELECTED COLLECTIONS. ARGENTINA. Misiones. Apostoles San Jose, Escuela Agrotecnico Don Bosco Grassland with niced scrub forest mostly along the river courses, 12 Dec. 1978, *Renvoize 3056* (P, B). **BRAZIL. Rio Grande do Sul.** São Francisco de Paula, CPCN Pró-Mata, 11 Nov. 2002, *Caporal* (MPUC). **Santa Catarina.** Bom Jardim, 15 Dec. 1971, *Smith & Klein 15791*(HBR). **PARAGUAY.** Villa Rica, Dans les praires marécogeuses, 10 Oct. 1874, *Balansa 263* (L). **URUGUAY.** Cerro Lago - Paso Tia Lucia, 11 Nov. 1964, *Arrillaga et al. 2454* (K); Arroyo de Pintos, Artilleros, near Puerto Plateo, Dep. de Colonia, 8 Aug. 2005, *Bartlett 20781* (P).

HABITAT. Open grasslands. Dry and wet soils, more common in damps and swamps.

NOTE. The linear and narrow leaf-blades of this species are typical and remarkable in the field. However, there are several specimens with wider leaf-blades.

18. Chascolytrum rhomboideum (*Link*) *Essi, Longhi-Wagner & Souza-Chies comb. nov.*

Type: Habitat in Chile, e seminibus ideallitis enata. (holotype B n.v.; isotypes BAA n.v. P n.v.)

Rhombolytrum rhomboideum Link [“Rhombolytrum”] in Hort. Berol. 2: 296 (1833)

Poa tumidula Steud. (1854: 258) Type: Chile: Cachapuel Rancagua, *Bertero 277* (lectotype P! (**designated here**); isolectotype US n.v.).

Rhombolytrum quinquenervium Phil. (1864: 293 - 294) Type: Prope “los Molles” in prov. Aconcagua. *Philippi* (holotype SGO n.v.; isotypes P n.v. US n.v.).

Plants 30 - 53 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous or pilose, margins non-overlapping or overlapping.

Leaf-blades linear, 6 - 26 cm x 0.6 – 4.5 mm, flat, glabrous or pilose on both surfaces.

Ligule 1.5 - 4 mm, truncate.

Panicle contracted, erect, 4.5 - 10 cm long. Pedicels scabrous. Spikelets 5 - 8 x 3 - 4 mm, 4 - 10-flowered, laterally compressed, rhomboid. Florets imbricate, obscuring the rachilla.

Glumes chartaceous, navicular, scabrous, subequal; lower glume 4.5 – 5 x 0.9 - 1.1 mm, 3 – 5 -nerved; upper glume 5 – 5.3 (8) x 0.9 - 1.1 mm, 3 - 5-nerved. Lemmas chartaceous to herbaceous, dorsi-ventrally compressed, pale, without a gibbous back, margins non-distinct from the back, non-cordate at the base, 5 - 9-nerved, midvein non-salient on the back, glabrous, smooth or scaberulous on the back, obtuse or acute at the apex, muticous, margins ciliate at the lower third, trichomes longer than 0.3 mm, non-inrolled at the base, without

oil glands; lower lemma 3.5 – 4 x 0.9 – 1.1 mm. Paleas lanceolate to linear-lanceolate, membranaceous, glabrous and smooth between the keels, keels ciliate; lower palea 3 – 3.2 x 0.6 – 1.1 mm. Lodicules linear. Stamens 1. Caryopsis n.v. Hilum n.v.

Fig. 22.A-D.

DISTRIBUTION. Restricted to Chile. Map 20.

SELECTED COLLECTIONS. CHILE. Santiago, Quebrada La Plata, 740 msm (33° 29' S - 70° 54'W), 15 Dec. 1960, *Schlegel 3325* (CONC); Santiago, La Obra, 20 Nov. 1927, *Montero 488* (CONC, K); El Olivar, Parque Fauna, 100 msm (33° 02' S - 71° 30'W), Oct. 86, *Poblete 58* (CONC); El Olivar, Parque Fauna, *Poblete58* (CONC); Limache, Nov. 31, *Garaventa 2523* (CONC); Nov. 1931, *Garaventa 2523* (CONC).

HABITAT. Rocky grasslands.

ETIMOLOGY. From Greek, *rhombos*, rhomboid, referring to the shape of the spikelets.

19. Chascolytrum rufum *J. Presl* (1830: 282) Type: Hab. In montanis Peruviae huanoccensibus, *Haenke* (holotype PR!)

Briza rufa (J. Presl) Steud. (1840: 225)

Poidium rufum (J. Presl) Matthei (1975: 98)

Lombardochloa rufa (J. Presl) Roseng. & Arrill. (1979: 254)

Briza glomerata Arechav. (1897: 469) Type: Uruguay, vive en parajes húmedos.

Arechavaleta 48 (lectotype MVM n.v. (designated by Rosengurtt et al., 1968);

isolectotypes LE n.v. US n.v.))

Briza glomerata Kuntze (1898: 342) nom. illeg. hom.

Plants 10 - 120 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, smooth, margins non-overlapping. Leaf-blades linear-lanceolate, 7 - 47 cm x 2 - 12.5 mm, flat, glabrous. Ligule 1.2 - 3 mm, truncate.

Panicle contracted, rarely open, erect, 3 - 13 cm long. Pedicels smooth. Spikelets 2 - 3.4 x 1.4 - 2.5 mm, 3 - 6-flowered, laterally compressed, elliptic-lanceolate. Florets imbricate, obscuring the rhachilla. Glumes herbaceous, convex, glabrous or pilose, ciliate or not on the margins, equal to subequal; lower glume 1.4 - 2.2 x 0.6 - 1 mm, 3-nerved; upper glume 1.4 - 2.3 x 0.7 - 1.2 mm, 3 - 5-nerved. Lemmas coriaceous, slightly laterally compressed, with a rufous gibbous back, strongly distinct from the broad margins, non-cordate at the base, 7-nerved, midvein non-salient on the back, glabrous on the back, with a crown of trichomes originated at the callus, obtuse at the apex, muticous, margins glabrous or pilose, inrolled or not at the base, with one oil gland (or its scar) on each side, at the base; lower lemma 1.6 - 2.4 x 0.9 - 1.5 mm. Paleas lanceolate, membranaceous, glabrous and smooth between the keels, keels ciliate; lower palea 1.1 - 1.7 x 0.5 - 0.9 mm. Lodicules linear to elliptic-lanceolate. Stamens 1 - 2. Caryopsis elliptic, concavo-convex. Hilum punctiform or elliptic.

Fig. 23.A-H.

HABITAT. Open grasslands, in dry and wet soils.

NOTE. We adopted the two varieties, based on previous works on genetic diversity, and on recent results from ISSR markers (Essi, Longhi-Wagner & Souza-Chies, in preparation) which support the two varieties.

Key to varieties:

- 1 . Lemmas and glumes glabrous..... *C. rufum* var. *rufum*
1' . Lemmas pilose on the margins, glumes pilose or
glabrous..... *C. rufum* var. *sparsipilosum*
-

Chascolytrum rufum *J. Presl* var. **rufum**

DISTRIBUTION. Argentina, Brazil and Uruguay. Map 21.

SELECTED COLLECTIONS. ARGENTINA. Buenos Aires. Campana, Otamendi, *Longhi-Wagner 905, 906* (ICN). **BRAZIL. Rio Grande do Sul.** Canoas, 6 Oct. 1997, *Rodrigues*, (ICN). **URUGUAY. Rocha.** Parque Santa Teresa, 18 Dec. 1969, *Rosengurtt 11151* (MSB). **PARAGUAY.** In regione colluim, Cerros de Tobaly, Oct. 1900, *Jürgens 6264* (W).

ETIMOLOGY. The epithet refers to the color of the lemma back, typically rufous.

Chascolytrum rufum var. sparsipilosum (Roseng., Arrill. & Izag.) Essi, Longhi-Wagner & Souza-Chies **comb. nov.** Type: Brazil: Rio Grande do Sul, 13 km de Cruz Alta a Panambi, 3 Nov. 1962, Rosengurtt & Del Puerto 8924. (holotype MVFA n.v.; isotype K!)
Briza rufa var. *sparsipilosa* Roseng., Arrill. & Izag. in Bol. Fac. Agron. Montevideo 105: 30 (1968)
Lombardochloa rufa var. *sparsipilosa* (Roseng., Arrill. & Izag.) Roseng. & Arril. (1979: 260)

DISTRIBUTION. Brazil and Argentina. Map 21.

SELECTED COLLECTIONS. ARGENTINA. Córdoba. W of Tanti at Rancho Alegre, 12 Dec. 1978, Renvoize 3741 (K). **BRAZIL. Paraná.** Piraquara, Autodromo Pinhares, 26 Dec. 1973, Hatschbach 33569 (MBM). **Rio Grande do Sul.** São Francisco de Paula – Tainhas, 29 Nov. 2002, Longhi-Wagner & Essi 8061 (ICN). **Santa Catarina.** Abelardo Luz, 15 Nov. 1964, Smith & Klein 13320 (HBR).

ETIMOLOGY. The epithet *sparsipilosa* refers to the presence of sparse trichomes on the lemma margins, which characterize the variety.

20. Chascolytrum scabrum (Nees ex Steud.) Matthei Type: Rio Grande do Sul, Campo entre Panambi y Palmeiras, 3. 11. 1962, Rosengurtt & Del Puerto 8984. (neotype MVFA n.v. (designated by Matthei, 1975); isoneotype ICN!)

Chondrachyrum scabrum Nees ex Steud. (1854: 288)

Chondrachyrum scabrum Nees (1836: 449) nom. nudum.

Panicum chondrachyrum Trin. ex Steud. (1841: 254) nom. nudum.

Briza scabra (Nees ex Steud.) Ekman (1913: 53), nomen, sed non planta. (non *B. scabra* Nees ex Steud. = *Chascolytrum calothea* (Trin.) Essi, Longhi-Wagner & Souza-Chies)

Plants 49 - 65 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, scabrous, margins non-overlapping. Leaf-blades linear, 5 - 16 cm x 2.2 - 3.4 mm, flat, glabrous. Ligule 1.4 - 2.5 mm, obtuse.

Panicle contract, erect, 4 - 10.5 cm long. Pedicels smooth. Spikelets 2.4 - 3 x 2.5 - 3.4 mm, 2 - 3-flowered, laterally compressed, ovate. Florets imbricate, obscuring the rhachilla.

Glumes herbaceous, convex, glabrous, subequal, acute; lower glume 1.9 - 2.2 x 0.8 - 1.1 mm, 5-nerved; upper glume 2.2 - 2.5 x 1 - 1.4 mm, 5-nerved. Lemmas coriaceous, slightly laterally compressed, with a pale yellow to yellow-gold gibbous back, strongly distinct from the narrow margins, non-cordate at the base, 5-nerved, midvein non-salient on the back, glabrous, obtuse at the apex, muticous, margins glabrous, inrolled at the base, without oil glands; lower lemma 2 - 2.3 x 1.2 - 1.4 mm. Paleas elliptic to orbicular, coriaceous, glabrous and smooth between the keels, keels glabrous, rarely ciliolate at the upper half; lower palea 1.6 - 1.9 x 0.9 - 1.2 mm. Lodicules linear or lanceolate. Stamens 1 - 3.

Caryopsis sub-orbicular, plano-convex. Hilum elliptic.

Fig. 24.A-D.

DISTRIBUTION. Restricted to Rio Grande do Sul State, Southern Brazil. Map 8.

BRAZIL. Rio Grande do Sul. Campo entre Panambi e Palmeiras, estrada ao norte de Santa Maria, 3 Nov. 1962, *Rosengurtt 8984* (ICN); Jaquirana, estrada para Várzea do

Cedro, 6 Oct. 2006, *Longhi-Wagner et al. 10201* (ICN); Pelotas, Instituto Agronômico do Sul, 30 Oct. 1945, *Swallen 7238* (ICN); Soledade, BR 386, km 252, 20 Nov. 2003, *Essi et al. 101* (ICN); Vacaria, BR-116, km 205.5, 16 Sept. 1971, *Valls 1578* (ICN); *Ibidem*, 15 Nov. 1972, *Valls et al. 2433* (ICN); Vacaria, estrada Vacaria-Porto Alegre, km 205, 6 Dec. 1981, *Winge 1634* (ICN).

HABITAT. Open grasslands, in rocky fields and wet soils.

21. *Chascolytrum subaristatum* (Lam.) Desv. (1810: 190). Type: Amer. Merid.,

Commerson (holotype P!; isotypes CP n.v. R n.v.)

Briza subaristata Lam. (1791: 187)

Briza erecta Lam. (1791: 187) Type: Montevideo, *Commerson* (holotype P! isotypes P! NY n.v. CP n.v.) **synon. nov.**

Bromus rotundatus Kunth (1815: 152) Type: Crescit in alta planitie Regni Mexicani, inter Zelaya et Queretaro [*Bonpland 4015*] (holotype P!)

Calotheca rotundata (Kunth) Roem. & Schult. (1817: 632)

Chascolytrum rotundatum (Kunth) Kunth (1829: 121)

Briza rotundata (Kunth) Steud. (1840: 225)

Briza triloba Nees (1829: 482) Type: Montevideo, *Sellow* (holotype B n.v.; isotype MO!)

Chascolytrum trilobum (Nees) Desv. (1853: 383)

Calotheca microstachya J. Presl (1830: 268) Type: Hab. In montanis Peruviae, *Haenke* (holotype PR n.v.; isotype MO!)

Calotheca macrostachya J. Presl (1830: 268) Type: in montanis Peruviae huanoccensibus, *Haenke* (holotype PR!; isotypes LE n.v. US n.v.)

Briza macrostachya (J. Presl) Steud. (1840: 225)

Calotheca reniformis J. Presl (1830: 268) Type: Hab. In montanis Peruviae, *Haenke*
(holotype PR n.v.; isotype W n.v.)

Briza reniformis (J. Presl) Steud. (1830: 225)

Calotheca stricta Hook. & Arn. (1832: 50) Type: Chile, Concepción, *Lay & Collie*
(holotype GL n.v.)

Briza stricta (Hook. & Arn.) Steud. (1840: 284)

Chascolytrum strictum (Hook. & Arn.) E.Desv. (1854: 384)

Briza violascens Steud. (1854: 283) Type: Chile, *Lechler 250* (holotype P!; isotypes P! B!
BAA n.v. (fragm.), US (fragm.) n.v.)

Chascolytrum coarctatum Phil. (1864: 293). Type: Prov. Valdivia, San Juan 1862, *Philippi*
(holotype W!)

Briza triloba f. *pumila* Hack. ex Kneuck. (1902: 97) Type: Estancia San Teodoro, Distrikt
Villamonte, Bezirk Rio Primeiro, Provinz Córdoba in Argentinien, *Teodoro Stuckert*
(holotype W!; isotypes S! K! MO! US n.v. MVM n.v.)

Briza triloba var. *interrupta* Hack. ex Stuck. (1911: 146) Type: Argentina, *Stuckert 19995*
(holotype W n.v.)

Briza subaristata var. *interrupta* (Hack. ex Stuck.) Roseng., Arrill. & Izag. (1968: 22)

Plants 27 - 100 cm high. Basal internodes of the culms non-thickened. Basal innovations
extravaginal. Leaf-sheaths glabrous and smooth, margins non-overlapping. Leaf-blades
linear-lanceolate, 9 - 50 cm x 2.4 – 6.5 mm, flat, glabrous, smooth or scaberulous on the
margins. Ligule 0.6 – 4.5 mm, truncate or obtuse.

Panicle open or sub-contracted, erect or pendulous, 4 – 21 cm long. Pedicels smooth. Spikelets 2.4 - 9 x 1.9 – 9 mm, 4 - 14-flowered, cylindrical to globose, oblong to orbicular. Florets imbricate, obscuring the rachilla. Glumes herbaceous, convex, glabrous, smooth or scaberulous, subequal, lower glume 1.5 – 4.6 x 0.6 – 1.7 mm, 3 - 9-nerved; upper glume 1.5 – 4.8 x 0.9 – 1.9 mm, 5 - 11-nerved. Lemmas coriaceous, dorsi-ventrally compressed, with a pale to cream gibbous back, strongly distinct from the broad margins, cordate at the base, 5 (12)-nerved, midvein non-salient on the back, glabrous or pilose, acute, muticous or mucronate at the apex, margins glabrous, non-inrolled at the base, without oil glands; lower lemma 1.8 – 6.5 x 0.9 - 5 mm. Paleas elliptic-orbicular to orbicular, coriaceous, glabrous between the keels, rarely pilose, keels glabrous or ciliolate; lower palea 1.3 - 3 x 0.8 – 1.9 mm. Lodicules flabelliform. Stamens 1 - 3. Caryopsis sub-orbicular, plano-convex. Hilum punctiform or elliptic.

Fig. 25.A-G.

DISTRIBUTION. Ubiquitous in temperate or subtropical areas from South America, from Argentina to Peru towards the West, occurring in Southern Brazil, Uruguay, Paraguay, Chile, Bolivia and Colombia, extending to Guatemala and Mexico. Adventitious in Australasia. Map. 22.

SELECTED COLLECTIONS. ARGENTINA. Salta. Dept. Guachipas, Cuesta del Lajan, ruta 6, 27 Oct. 1991, *Charpin 23049* (G). **AUSTRALIA.** Cumberland State Forest, west Pennant Hills, 29 Oct. 1976, *Coveny 8595* (K). **BRAZIL. Rio Grande do Sul.** Bom Jesus, Fazenda Fundo das Almas, 23 Jan. 2001, *Wasum 1269* (G). **CHILE. Concepción.** Región del Biobío, 10 Km Flussabwärts an hydragr., linken Talhang des Biobío gegen

Santa Juana, 24 Oct. 1987, *Rechinger & Rechinger 63039* (W). **GUATEMALA. Quiche.**

Bancos húmedos del bosque misto de Pascual Abaj; Chichicastenango, 1 Nov. 1965,

Standley 15299 (G). **MEXICO.** San Luis Potosé Edge of irrigation ditch, 15 July 1910,

Hitchcock 456 (L, K). **PARAGUAY.** National Park Ybyaí, NW corner of park along

Arroyo Mina Cerrado, 26° 01' S 56° 46'W, 25 Nov. 1991, *Zardini & Tilleria 28966* (K).

PERU. *Haenke* (W 1889 - 246598). **URUGUAY. Canelones.** Carrasco, 25 Nov. 1968,

Rosengurtt 10940 (BM).

HABITAT. Open grasslands, natural or disturbed, including road sides. Occur in several types of soil, including sandy soils near the coast.

ETIMOLOGY. The epithet refers to the presence of a mucron at the lemma apex.

VERNACULAR NAMES. Flor de Maria (Spanish, Mexico).

NOTES. 1. *Chascolytrum erectum*, here proposed as a synonym of *C. subaristatum*, was usually distinguished from *C. subaristatum* by its larger and cream-colored lemmas. However, *C. subaristatum* is extremely variable, and intermediates are very common, like plants with small cream spikelets and plants with larger, but non-cream spikelets. Even in the same tuft it is possible to find several sizes of spikelets. We also observed that the soil can influence the spikelets color, usually cream in sandy soils, and the size of the rhizomes, being longer and thin in the same type of soil. We believe that the features traditionally used to separate *C. erectum* from *C. subaristatum* are influenced by the environment and genetic intra-specific variability. A preliminary approach with ISSR markers (Essi, Longhi-Wagner & Souza-Chies, submitted) agrees with the proposed synonymy.

2. *Chascolytrum subaristatum* (under *Briza subaristata*) and *C. erectum* (under *B. erecta*) were published by Lamarck (1791) simultaneously, on the same page. However, *C. subaristatum* is the name accepted here, because it is already widespread in several floras, while the name *C. erectum* is usually restricted to Brazilian, Argentinian and Uruguayan floras.

22. *Chascolytrum uniolae* (Nees) Essi, Longhi-Wagner & Souza-Chies comb. nov. Type:

Habitat in confinibus regni Paraguayan, *Sellow* (holotype B n.v.; isotypes BAA (fragm.) n.v. LE n.v.)

Eragrostis uniolae Nees in Mart., Fl. Bras. Enum. Pl. 2: 494 - 495 (1829)

Briza uniolae (Nees) Nees ex Steud. (1855: 283)

Poidium uniolae (Nees) Matthei (1975: 93)

Chascolytrum spicigerum J. Presl (1830: 282) Type: Hab. in Peruviae montanis et in Chile,

Haenke (holotype PR n.v.; isotypes W! MO! M n.v. US (fragm.) n.v.)

Briza spicigera (J. Presl) Steud. (1840: 255)

Briza uniolae var. *modestior* Döll (1878: 131) Type: In insula S. Catharinae, F. Müller (holotype B!; isotype US (fragm.) n.v.)

Plants 30 - 135 cm high. Basal internodes of the culms non-thickened. Basal innovations extravaginal. Leaf-sheaths glabrous, scabrous, margins non-overlapping. Leaf-blades linear-lanceolate, 7 - 43 cm x 2 - 12 mm, flat, glabrous and smooth. Ligule 2 - 8 mm, obtuse to truncate.

Panicle contracted, rarely open, erect, 5 - 32 cm. Pedicels scabrous. Spikelets 3.2 - 6 x 2 - 4.4 mm, 4 - 11-flowered, laterally compressed, ovate to elliptic-lanceolate. Florets

imbricate, obscuring the rhachilla. Glumes herbaceous to chartaceous, naviculate, non-cordate at the base, glabrous, scabrous, subequal; lower glume 1.8 – 2.8 x 0.7 – 1.3 mm, 3 – 5 -nerved; upper glume 2 - 3 x 0.8 – 1.5 mm, 5 - 7-nerved. Lemmas chartaceous, laterally compressed, with a papillose whitish gibbous back, rarely without papillae, strongly distinct from the broad margins, non-cordate at the base, 5-nerved, midvein salient on the back, glabrous, acute or bidentate at the apex, muticous or mucronate, margins glabrous, smooth or scabrous, non-inrolled at the base, but slightly curved at the apex, without oil glands; lower lemma 2.4 – 3.4 x 1.2 - 2 mm. Paleas elliptic to elliptic-lanceolate, membranaceous, glabrous and smooth between the keels, rarely sparsely pilose, keels ciliate; lower palea 1.2 - 2 x 0.4 – 0.8 mm. Lodicules linear. Stamens 1. Caryopsis elliptic, concavo-convex. Hilum elliptic or punctiform.

Fig. 26.A-D.

DISTRIBUTION. Argentina, Brazil, Bolivia, Paraguay and Uruguay. Map 23.

SELECTED COLLECTIONS. ARGENTINA. Corrientes. 7 km E de Saladas, 18 Nov. 1987, *Quarín et al.* 3875 (K). **Entre Rios.** 14 Nov. 79, *Troncoso, et al.* 2696 (K).

BOLIVIA. Chuquisaca. Azurduy, in the gorge below La Angostura, c. 6 km below Azurduy, 5 Dec. 1999, *Wood et al.* 15325 (LPB); Chuquisaca, Boeto, Nuevo Mundo, NE of Villa Serrano, 23 Nov. 1995, *Wood* 10172 (LPB). **Cochabamba.** Carrasco, c 1 Km from Montepuncu towards Schuencas, 29 Dec. 1995, *Wood* 10315 (K). **Santa Cruz.** Vallegrande, on ascent of first range climbing out of Guadalupe on the road to Masicuri, 25 Dec. 1999, *Wood & Goyder* 15623 (LPB). **Tarija.** O' Connor, Los Canalitos area, c 55-60 km from Tarija towards Entre Rios, 23 Nov. 1996, *Wood* 11628 (LPB). **Santa Cruz.** Santa

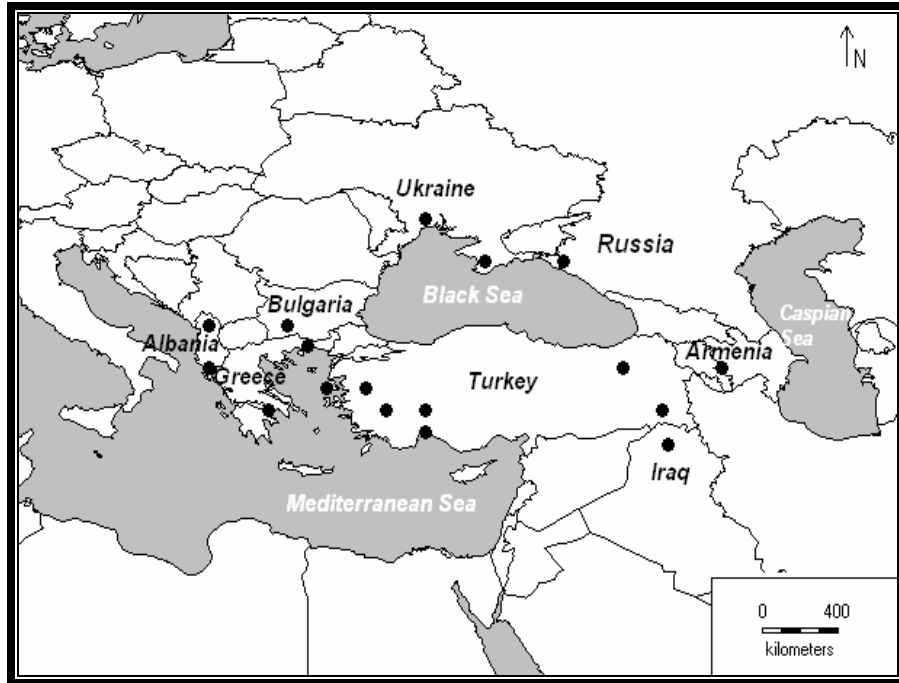
Cruz, 11 Feb. 96, *Wood 10597* (K). **Brazil. Paraná.** Curitiba, Vila Macedo, 9 Nov. 1993, *Ribas & Barbosa 598* (MBM); Curitiba, Capão da Imbia, 14 Nov. 74, *Dombrowski 5509* (K). **Rio Grande do Sul.** Cambará do Sul, estrada para Fortaleza, 24 Nov. 1994, *Hatschbach & Ribas 61310* (MBM). **Santa Catarina.** São Joaquim, 18 Dec. 71, *Smith & Klein 15872* (K); Tubarão, 21 Nov. 1986, *Nunes H 2852* (MPUC). **São Paulo.** Campos do Jordão, 1925, *Chase 9838* (MO); Itapetininga, Campo Oeste, 1 Nov. 1987, *Lofgren 411* (P). **PARAGUAY. Caaguazú.** Caaguazu, Dans les prairies, Nov. 1874, *Balansa 137a* (L). **Guaira.** Villa Rica, Dans les prairies, Sept. 1874, *Balansa 137* (L). **Itapua.** Itapua, 25 Oct. 1999, *Zardini & Rodriguez 52128* (MO). **Paraguarí.** National Park Ybicuí, 11 Nov. 1989, *Zardini & Velásquez 15940* (MO). **Misiones.** Santiago, 23 Oct. 1959, *Pedersen 5200* (K). **URUGUAY. Maldonado.** Maldonado, 21 Nov. 1948, *Rosengurtt B-5260* (K). **Montevideo.** Montevideo, Nov. 1876, *Arechavaleta 5511* (L). **Rocha.** Cerro Los Indios, 20 Dec. 1969, *Rosengurtt 11192* (M, B).

HABITAT. Open grasslands. Wet and damp soils, less frequently in dry soils.

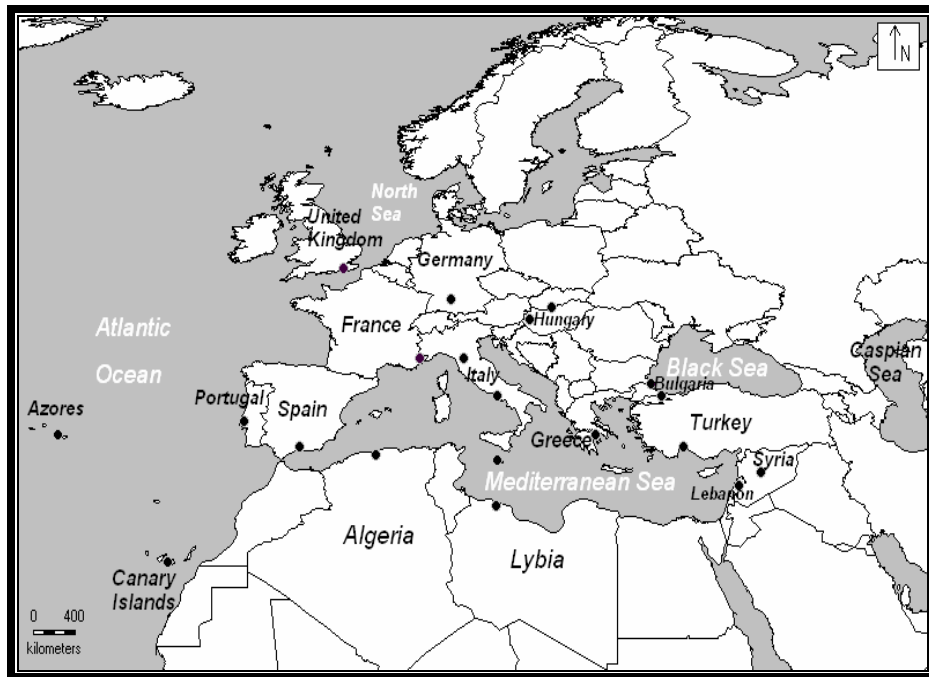
NOTE: *Chascolytrum uniolae* is usually easy to distinguish from *C. calotheca* by its generally contracted panicles, and lemmas papillose on the back. However, occasionally its plants present open panicles, and/or lemmas sparsely papillose to rarely smooth on the back. These plants, slightly different from the typical *C. uniolae*, can be differentiated from *C. calotheca* by its always erect and stiff panicle (generally pendulous or, if erect, non-stiff, in *C. calotheca*) and by the shape of the lemma, strongly gibbous and whitish on the back, with a salient midvein (lemma slightly gibbous, pale on the back, midvein non-salient in *C.*

calotheca). The inflorescences of *C. uniolae* are usually scabrous to the touch, which is noticeable in the field.

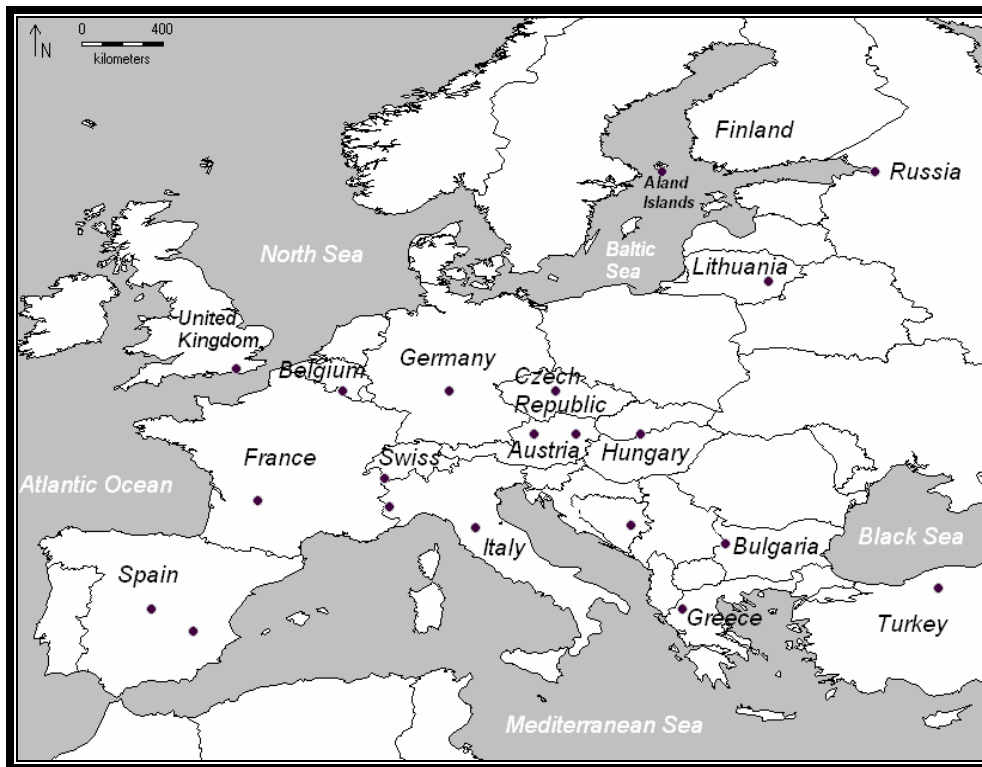
Distribution maps



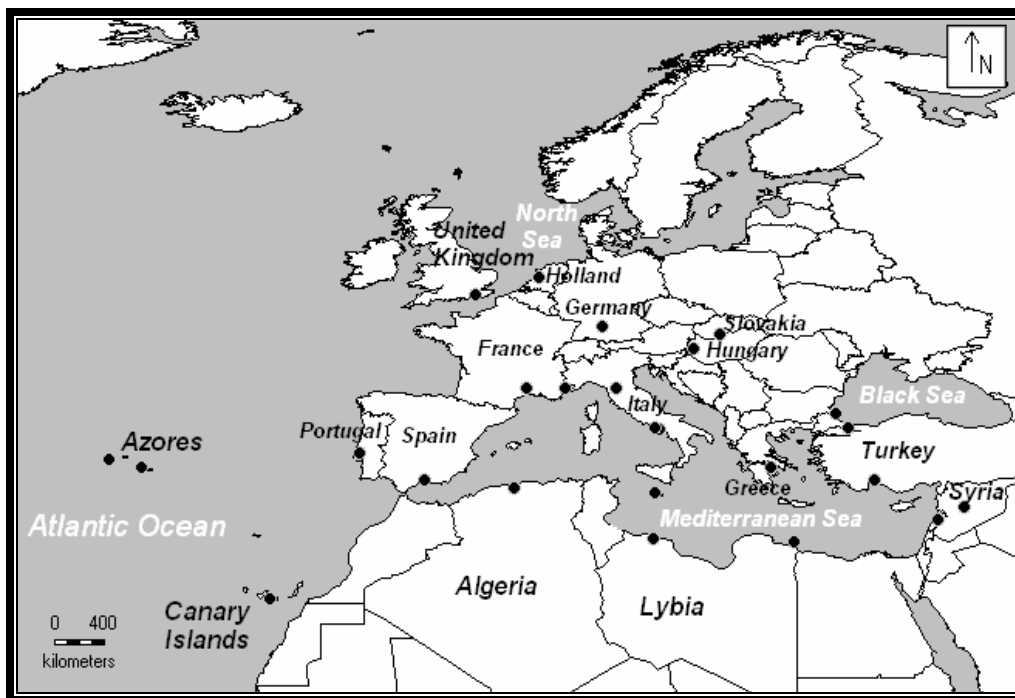
Map 1. Geographic distribution of *Briza humilis*.



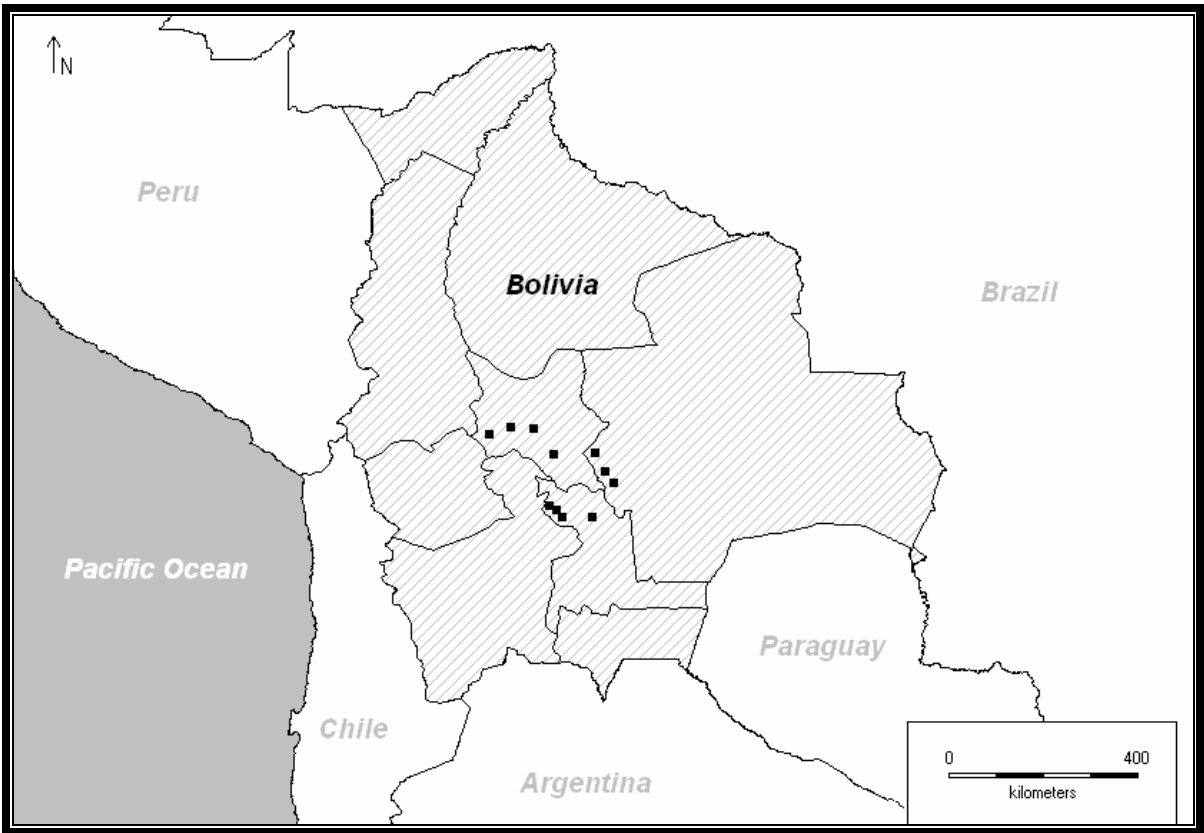
Map 2. Geographic distribution of *Briza maxima*.



Map 3. Geographic distribution of *Briza media*.



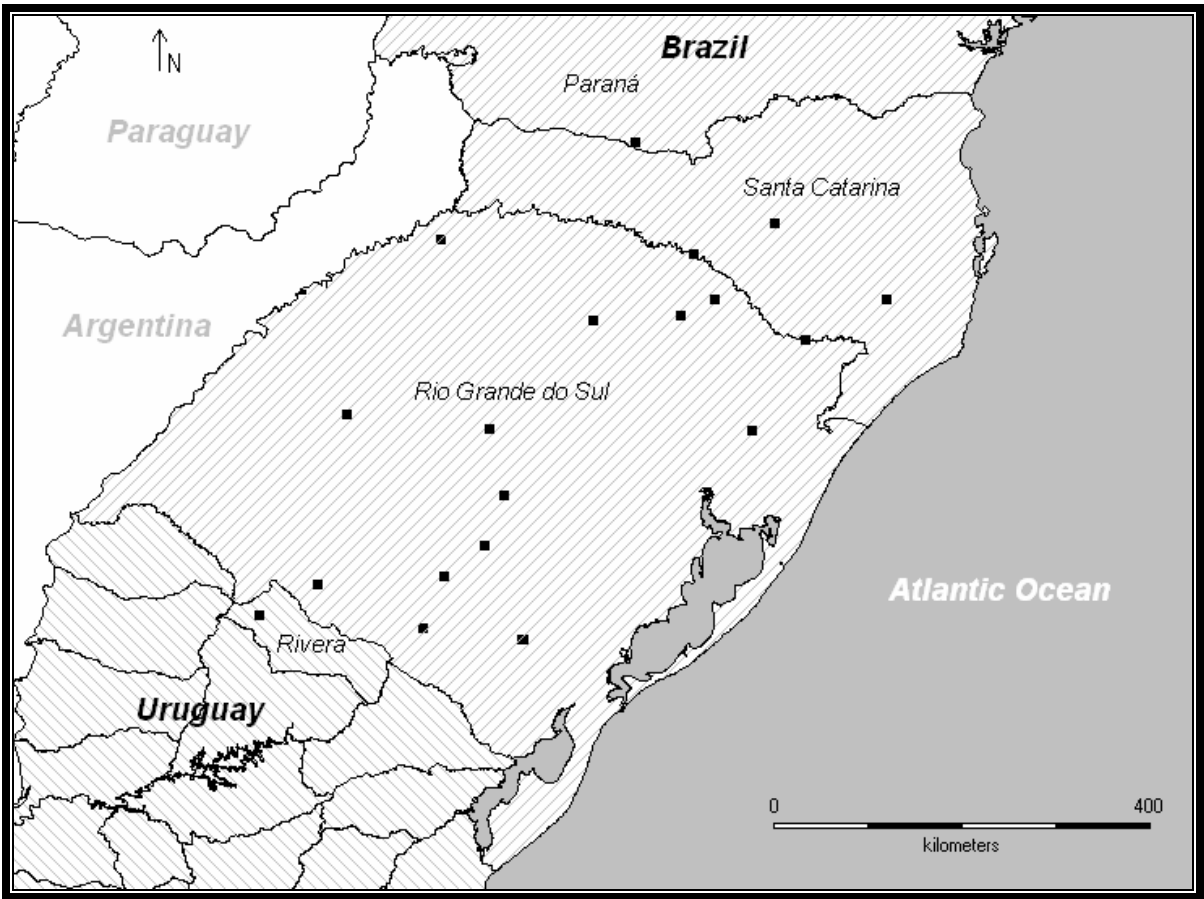
Map 4. Geographic distribution of *Briza minor*.



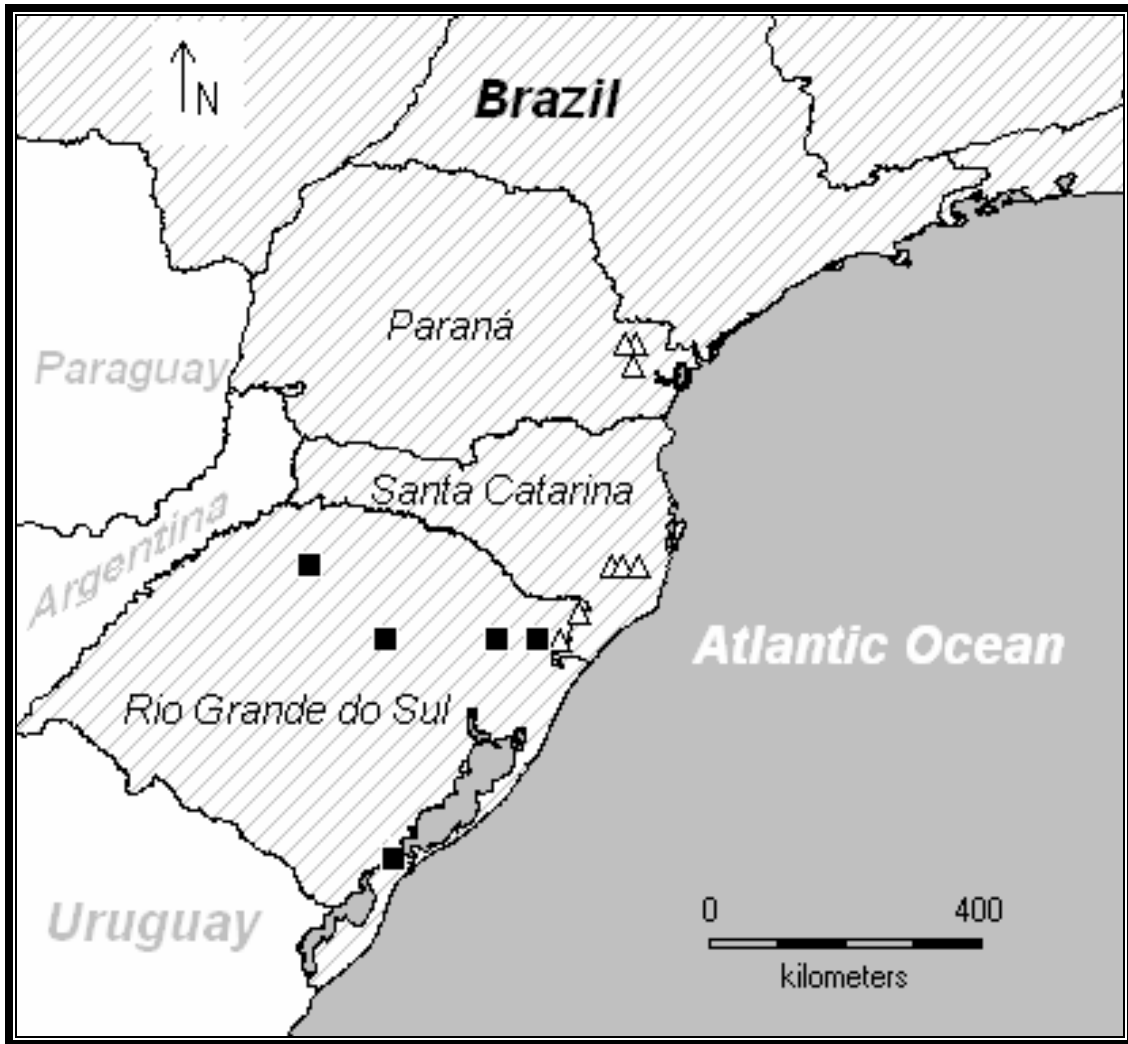
Map 5. Geographic distribution of *Chascolytrum altiplanense*.



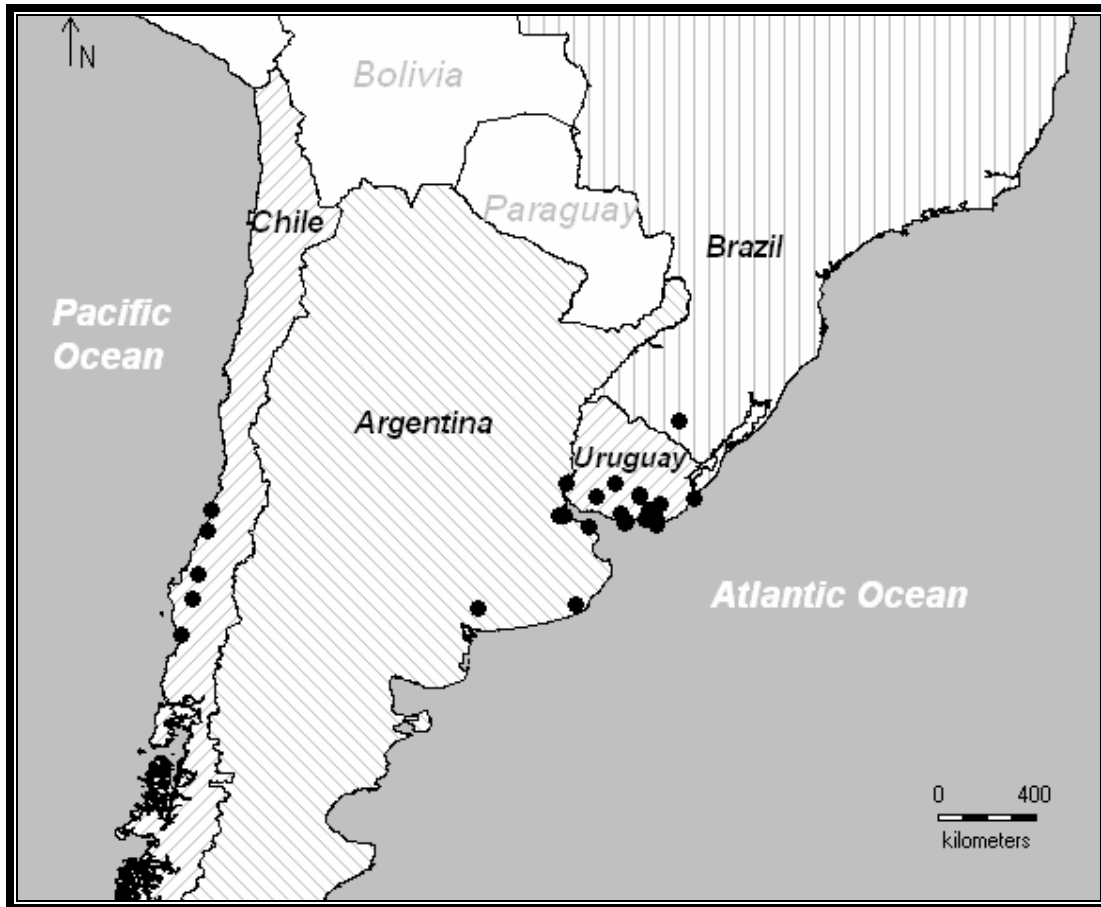
Map 6. Geographic distribution of *Chascolytrum ambiguum* (squares), *Chascolytrum brasiliense* (triangles) and *Chascolytrum itatiaiae* (crosses).



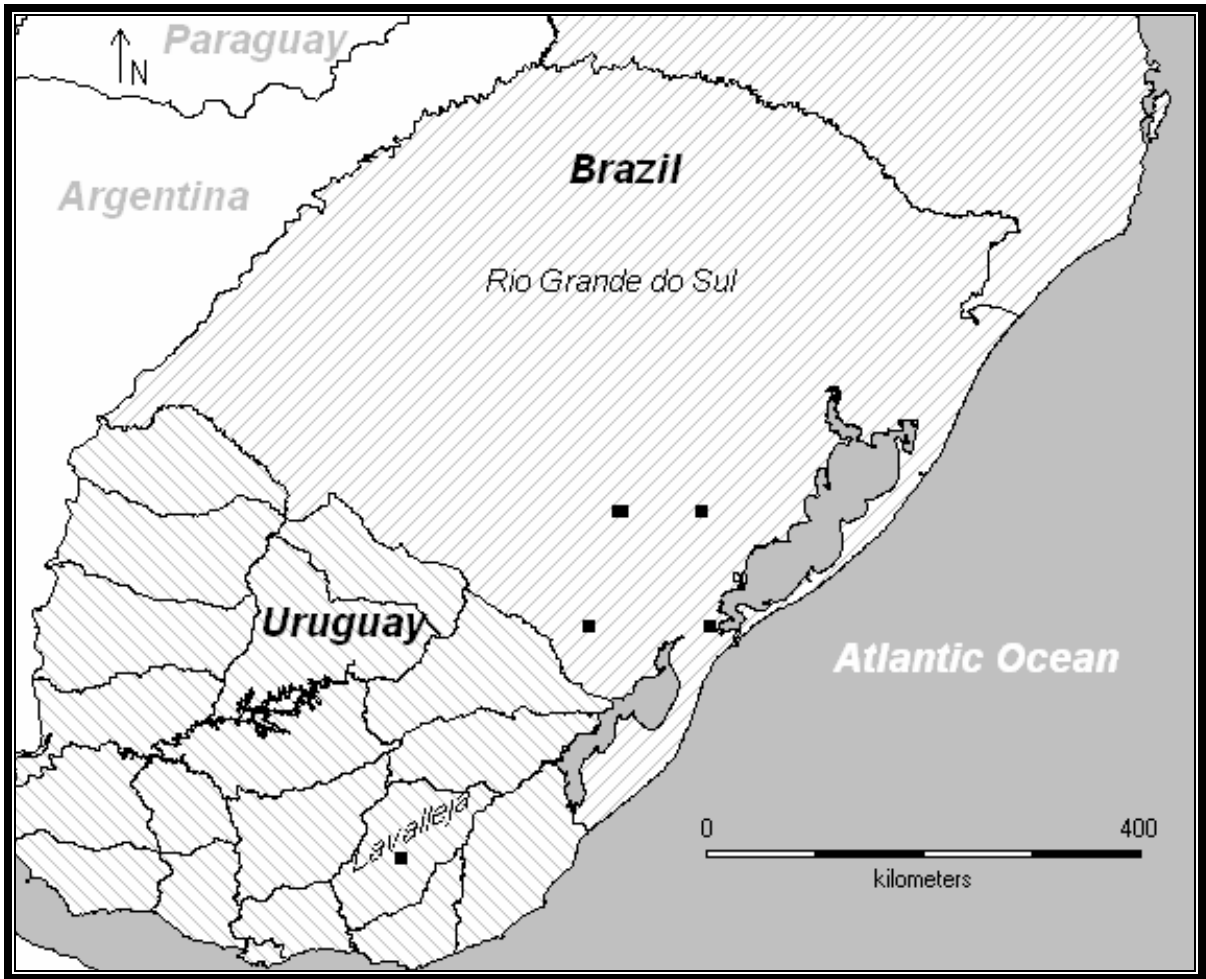
Map 7. Geographic distribution of *Chascolytrum bidentatum*.



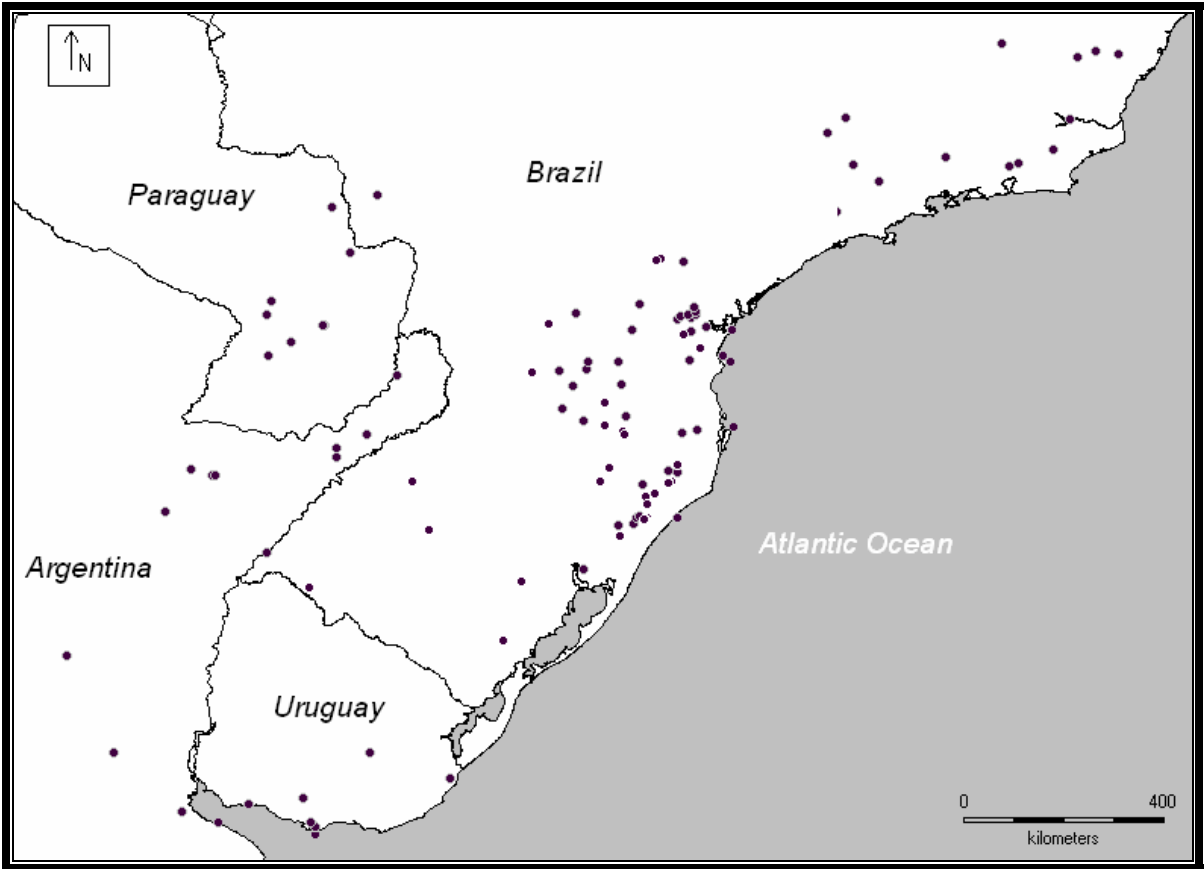
Map 8. Geographic distribution of *Chascolytrum brachychaetum* (triangles) and *Chascolytrum scabrum* (squares).



Map 9. Geographic distribution of *Chascolytrum brizoides*.



Map 10. Geographic distribution of *Chascolytrum bulbosum*.



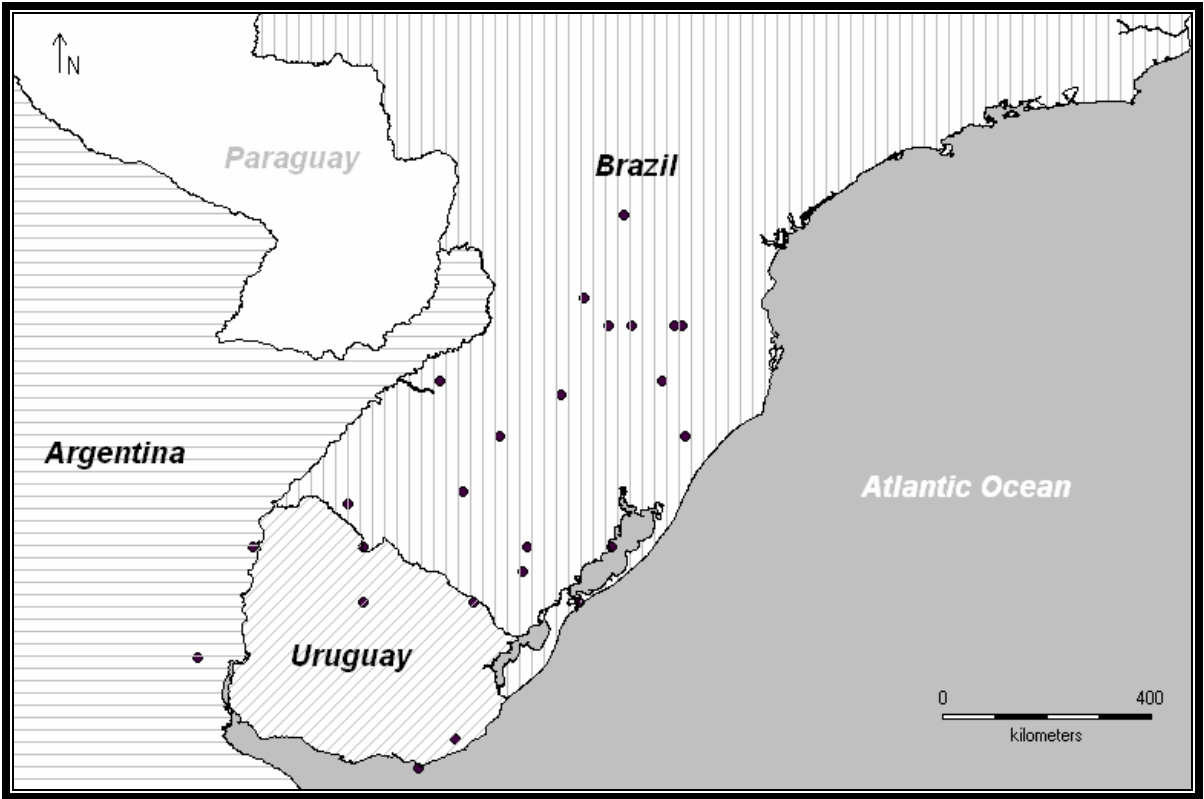
Map 11. Geographic distribution of *Chascolytrum calothea*.



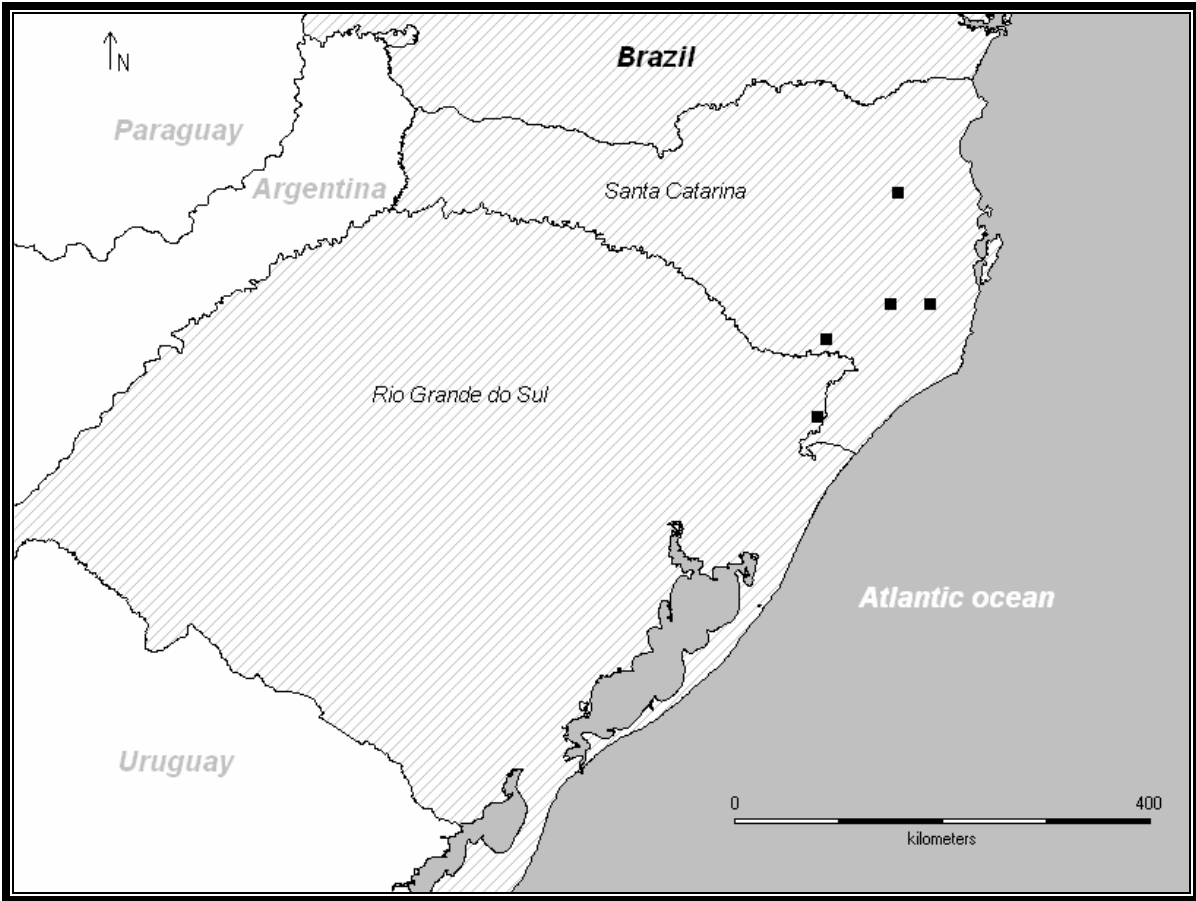
Map 12. Geographic distribution of *Chascolytrum juergensii* var. *juergensii* (points) and *Chascolytrum juergensii* var. *angustilemma* (triangles).



Map 13. Geographic distribution of *Chascolytrum koelerioides*.



Map 14. Geographic distribution of *Chascolytrum lamarckianum*.



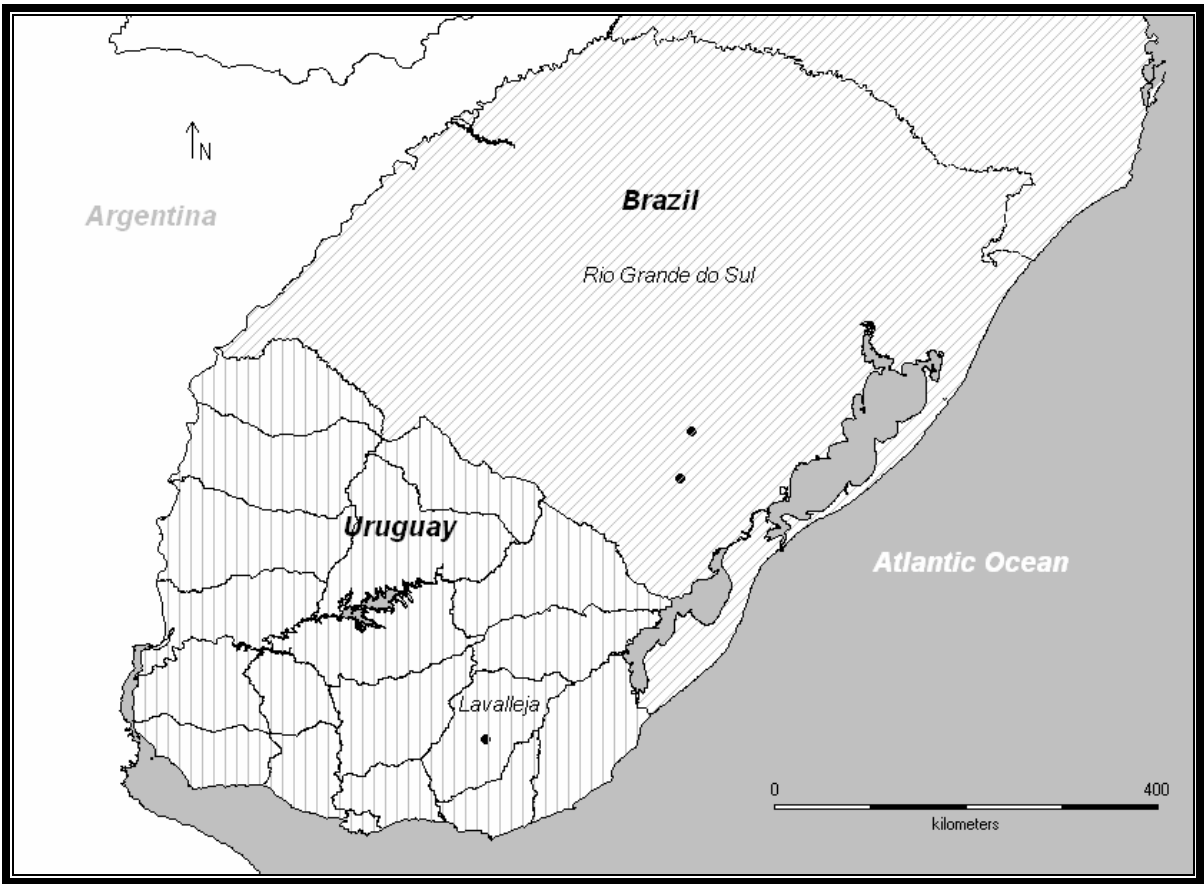
Map 15. Geographic distribution of *Chascolytrum latifolium*.



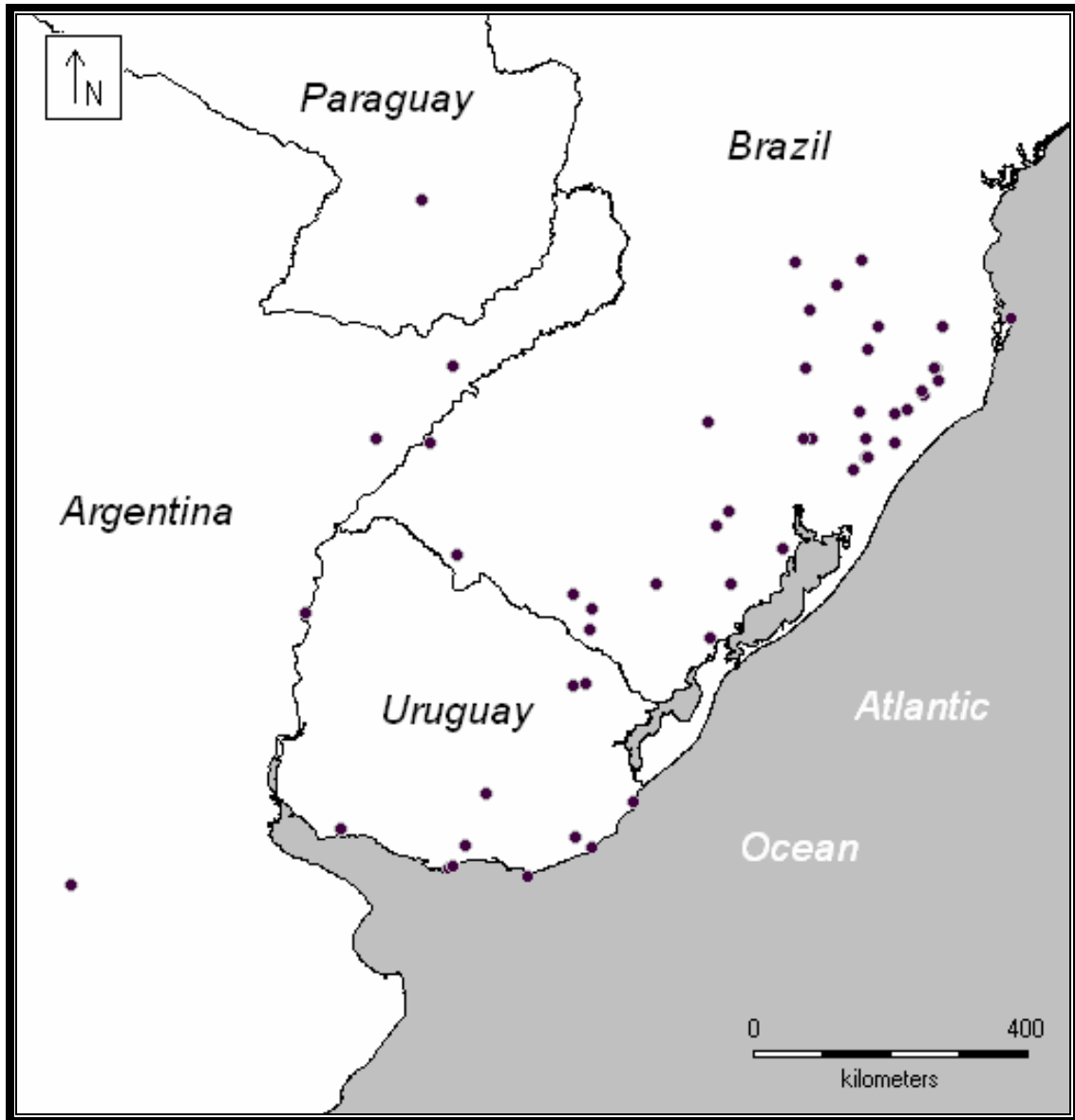
Map 16. Geographic distribution of *Chascolytrum monandrum*.



Map 17. Geographic distribution of *Chascolytrum paleopiliferum*.



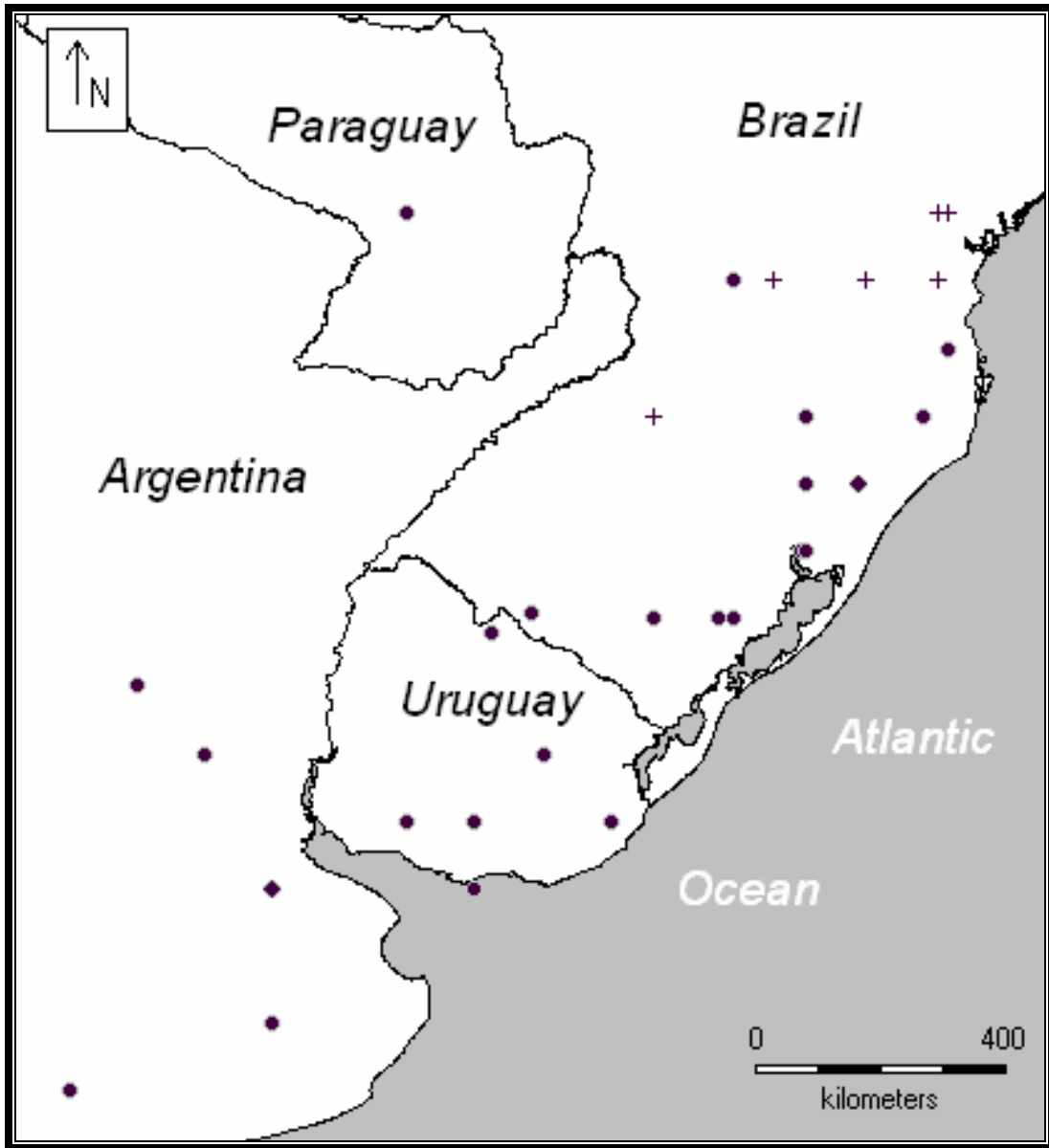
Map 18. Geographic distribution of *Chascolytrum parodianum*.



Map 19. Geographic distribution of *Chascolytrum poaemorphum*.



Map 20. Geographic distribution of *Chascolytrum rhomboideum*.



Map 21. Geographic distribution of *Chascolytrum rufum* var. *rufum* (points) and *Chascolytrum rufum* var. *sparsipilosum* (crosses).



Map 22. Geographic distribution of *Chascolytrum subaristatum*.



Map 23. Geographic distribution of *Chascolytrum uniolae*.

Figures

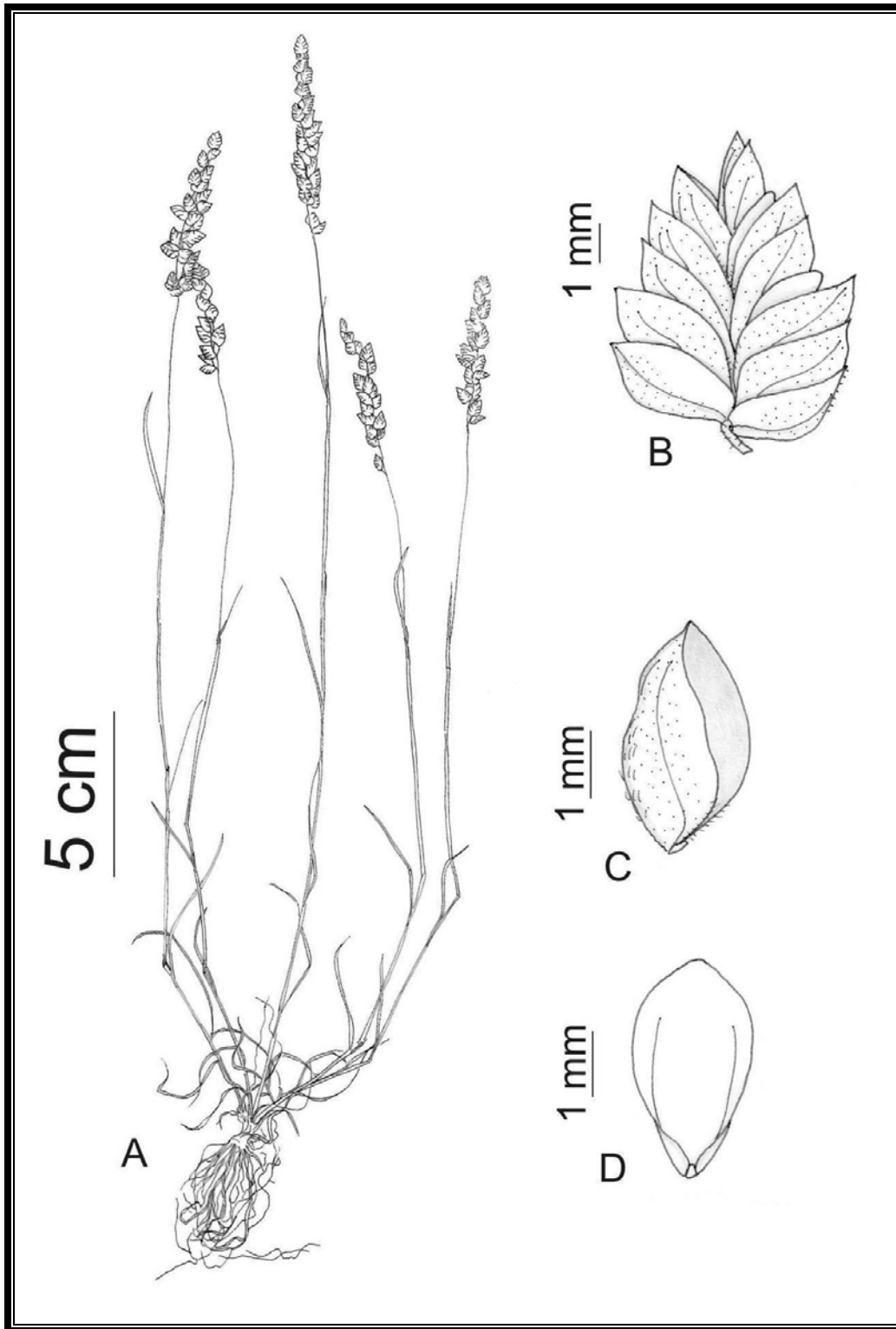


Fig. 1. *Briza humilis*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (**A.** Davis 42345, K; **B – D.** Townsend 63/104, K)

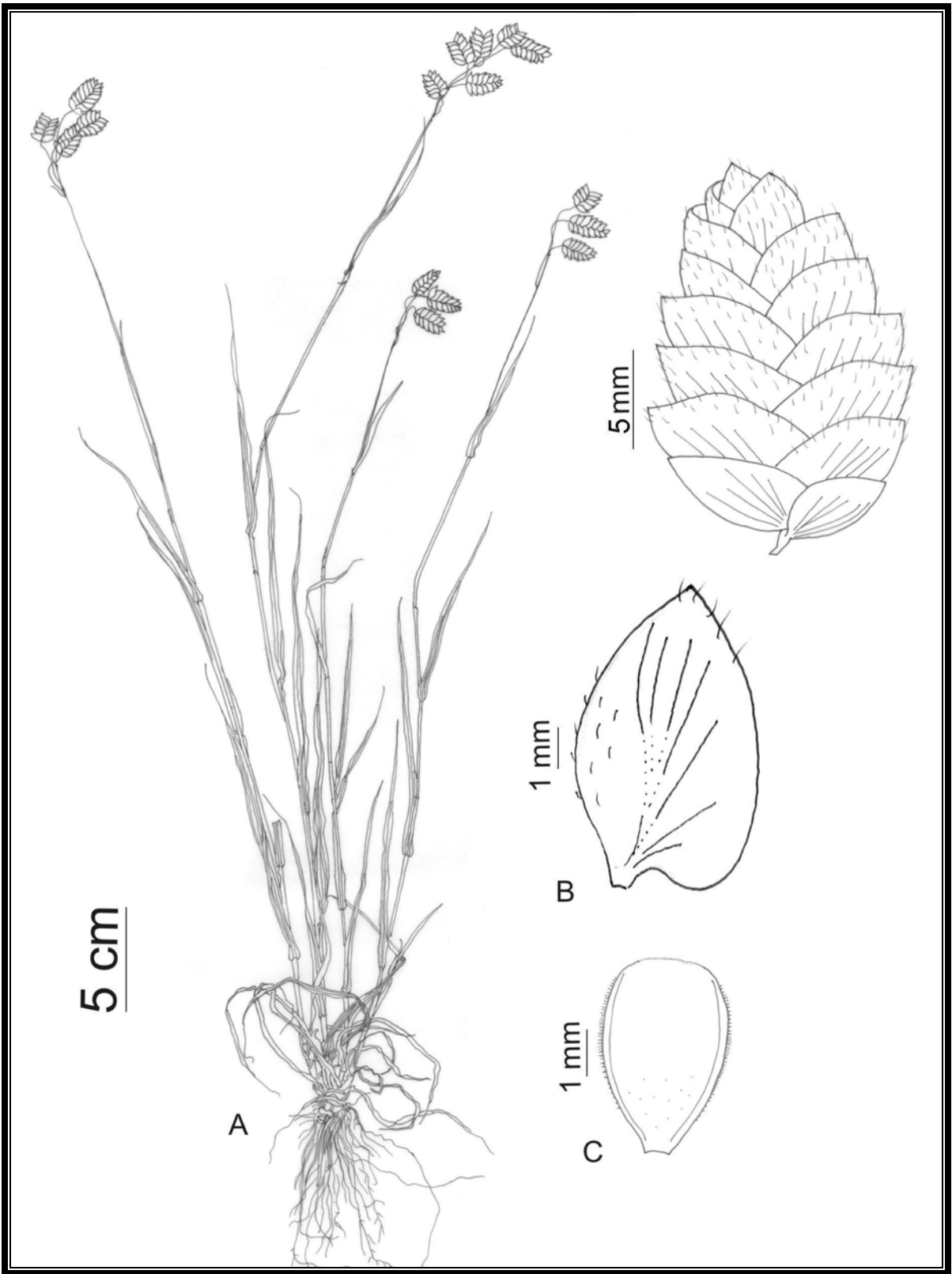


Fig. 2. *Briza maxima*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. Essi et al. 45, ICN)

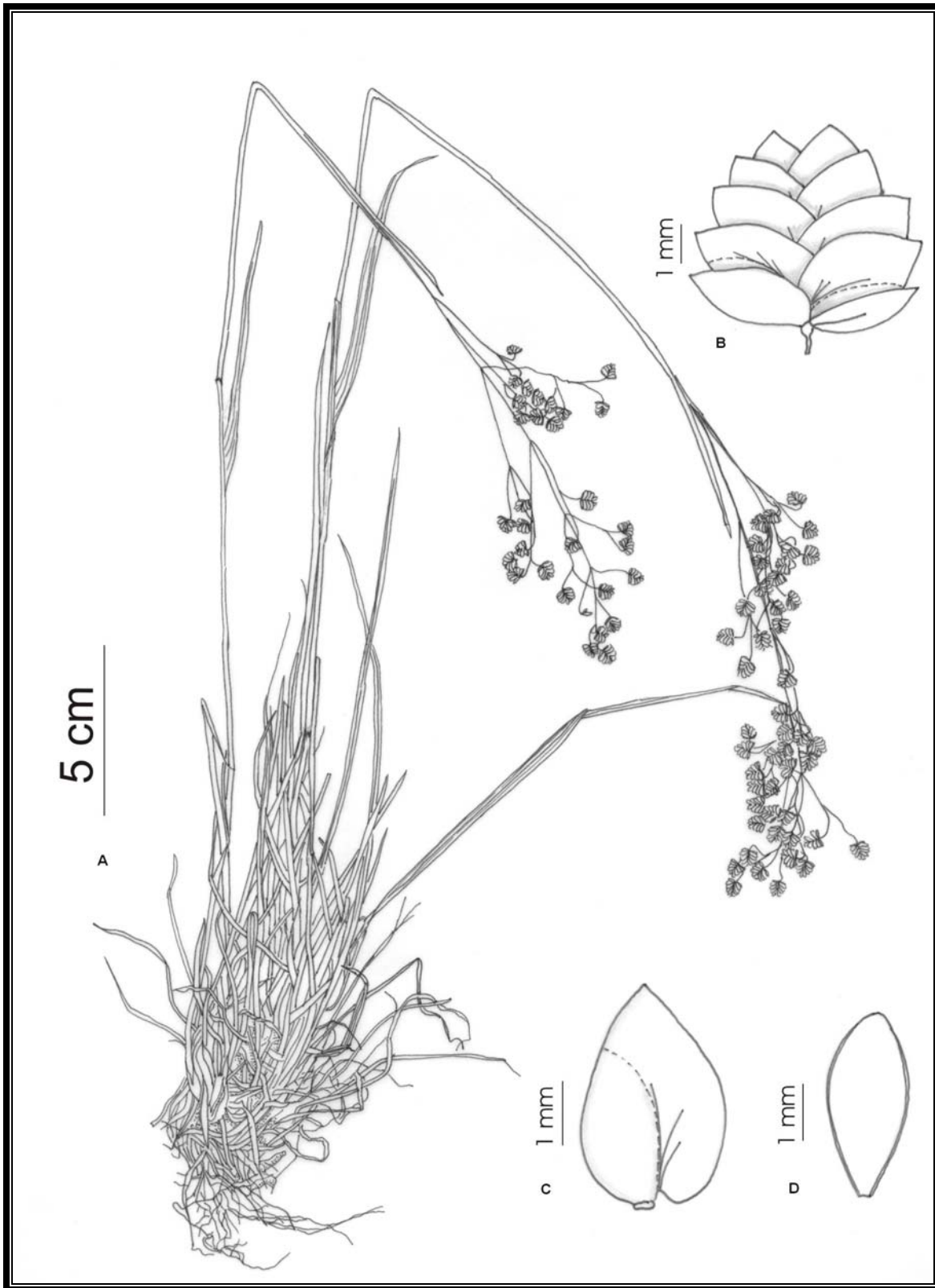


Fig. 3. *Briza media*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. Davis & Coode D37157, K)

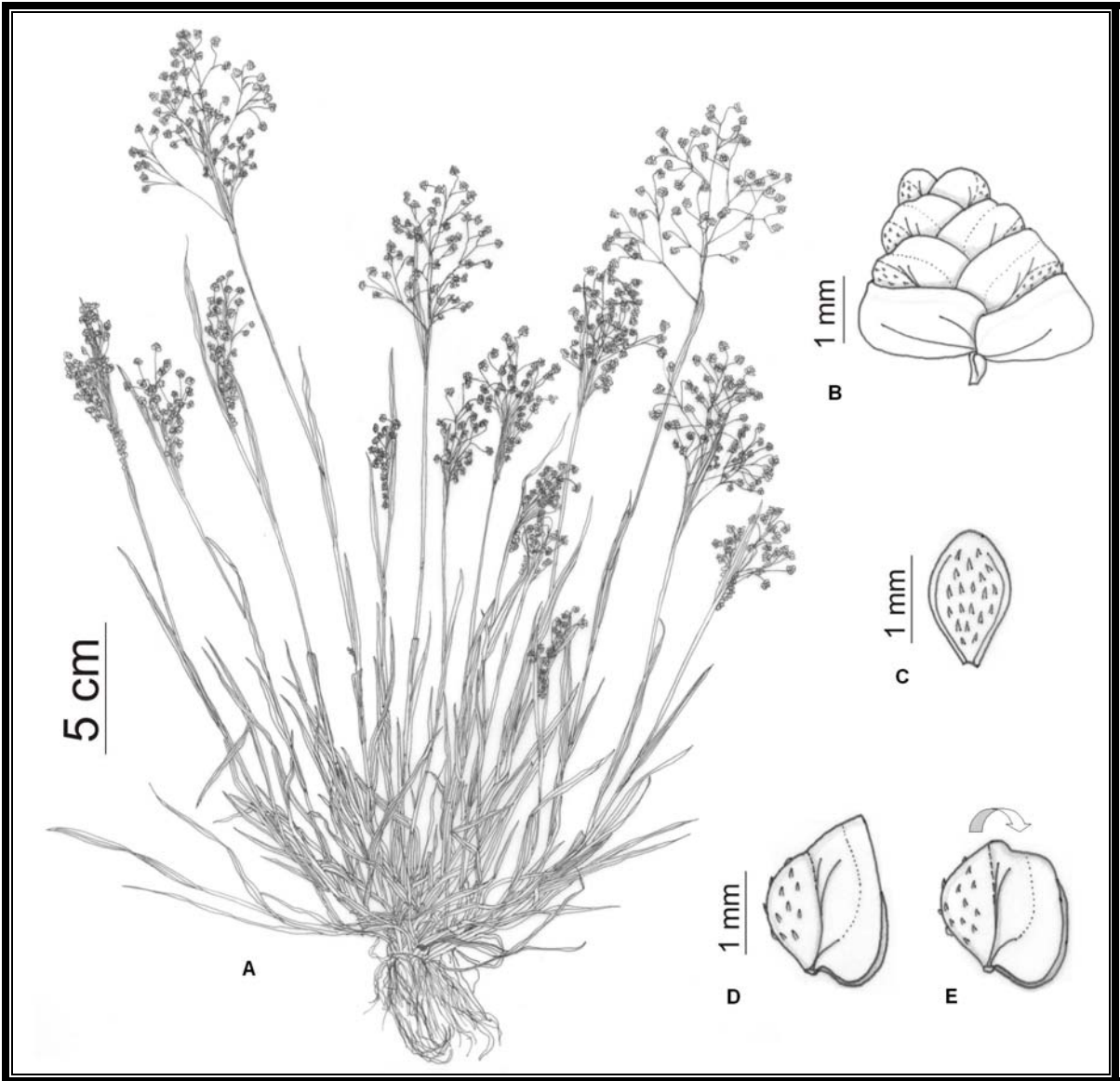


Fig. 4. *Briza minor*. **A.** Habit. **B.** Spikelet. **C.** Palea. **D.** Lemma (lateral view). **E.** Lemma (lateral view) with a curved apex. (**A.** *Essi 1*, ICN; **B – E.** *Essi et al. 63*, ICN)

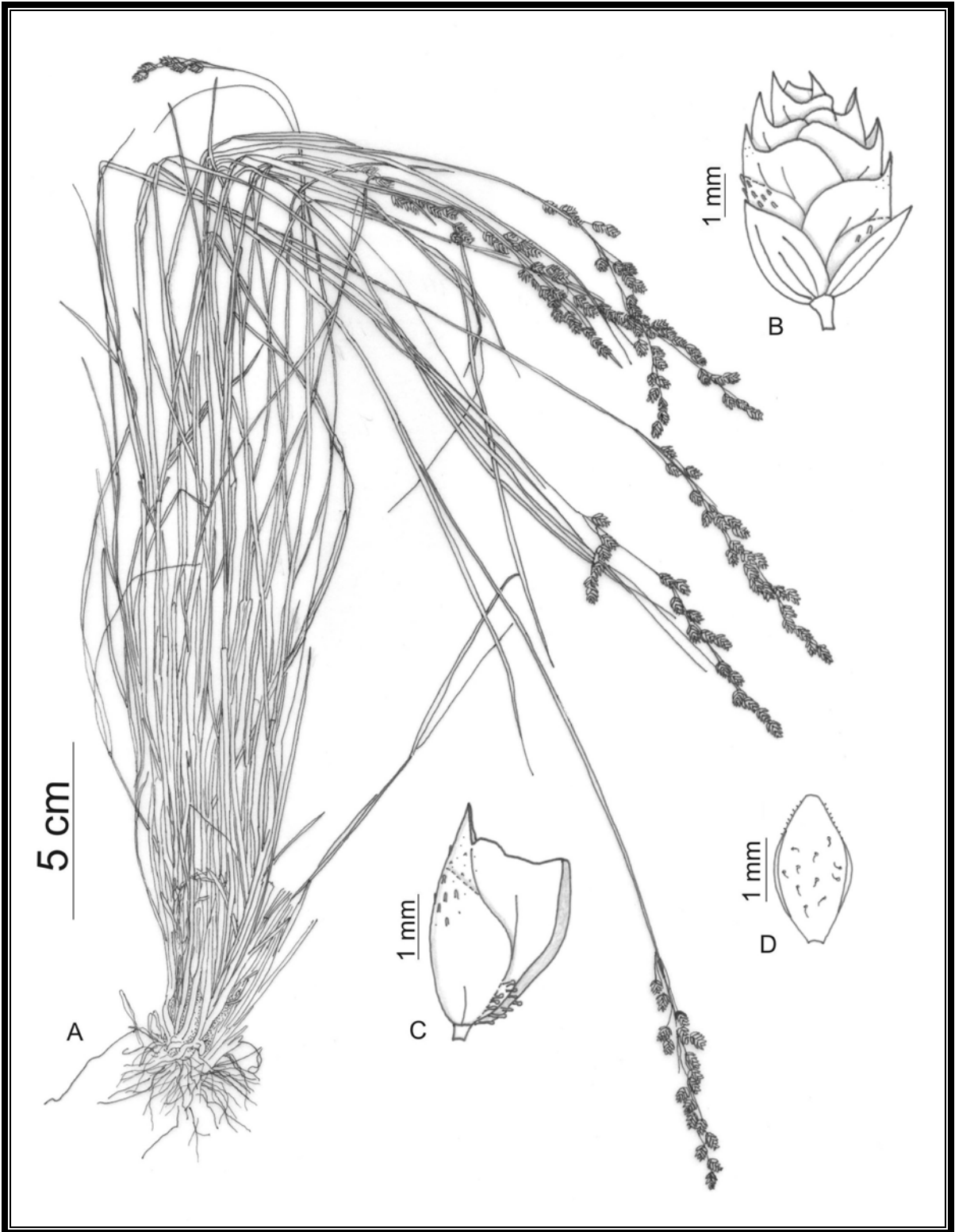


Fig. 5. *Chascolytrum altiplanense*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. Wood 10841, K)

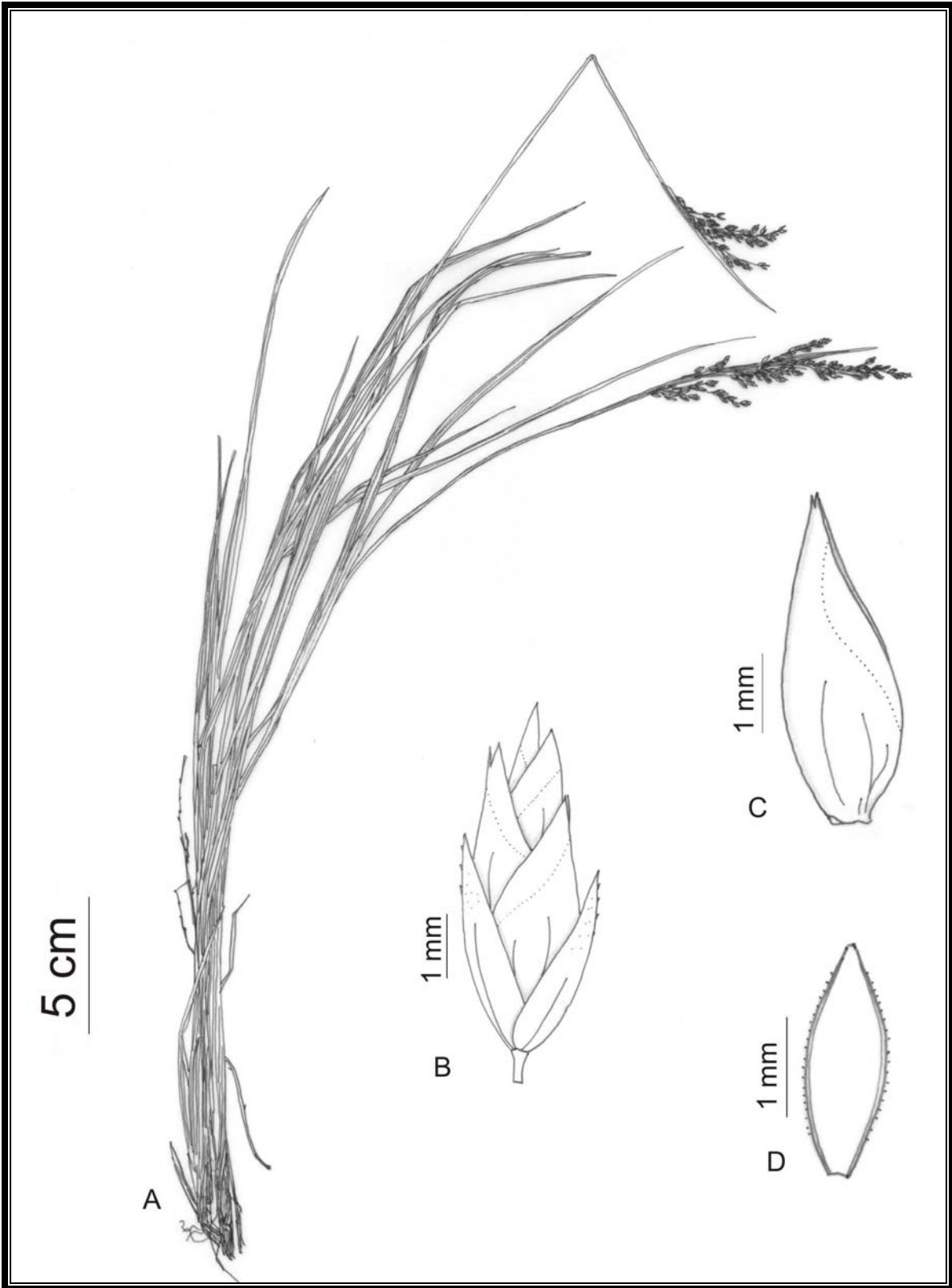


Fig. 6. *Chascolytrum ambiguum*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. Winge et al. 1544, ICN)

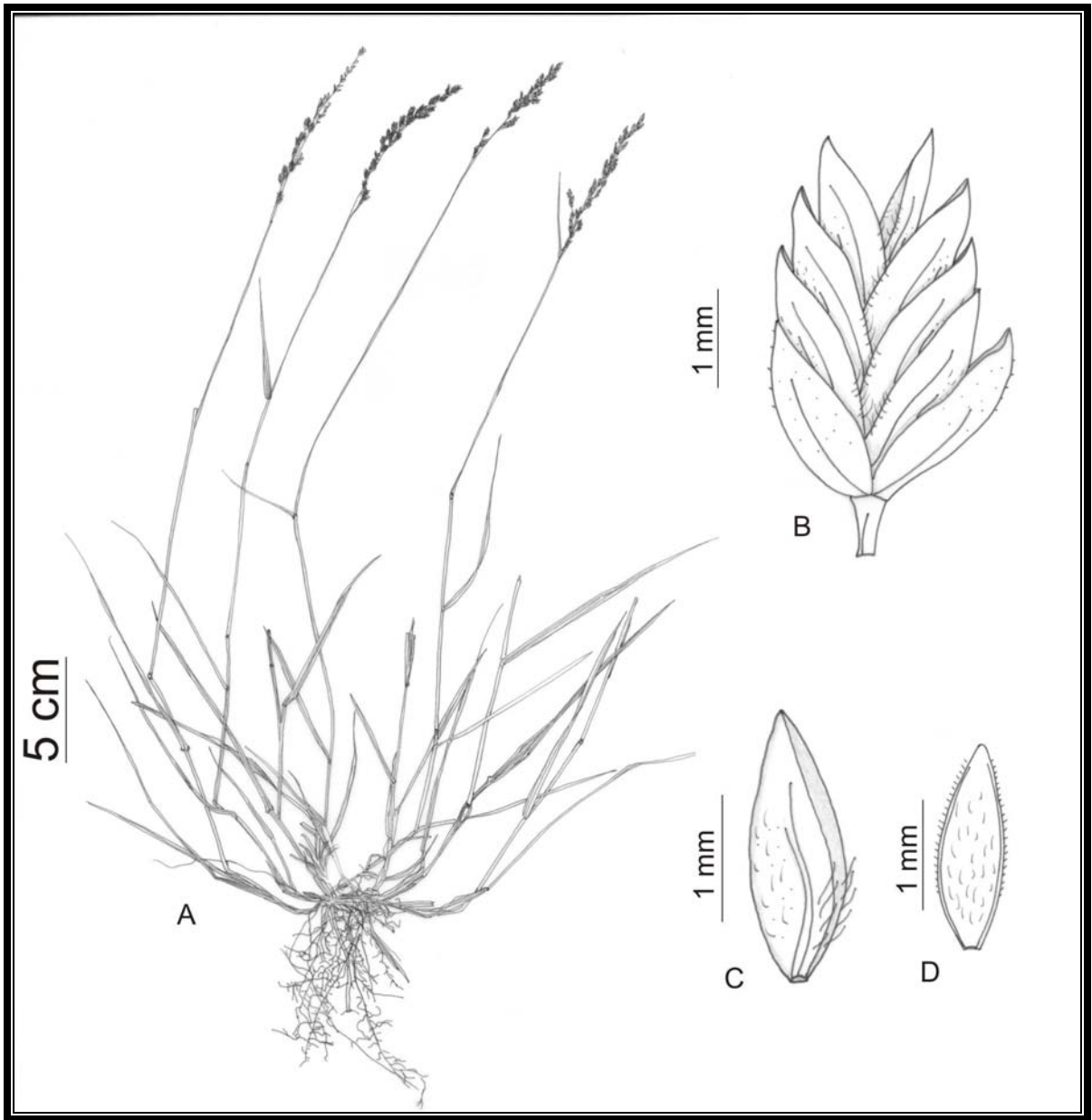


Fig. 7. *Chascolytrum bidentatum*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A. Essi et al. 318, ICN; B – D. Essi et al., 317D)

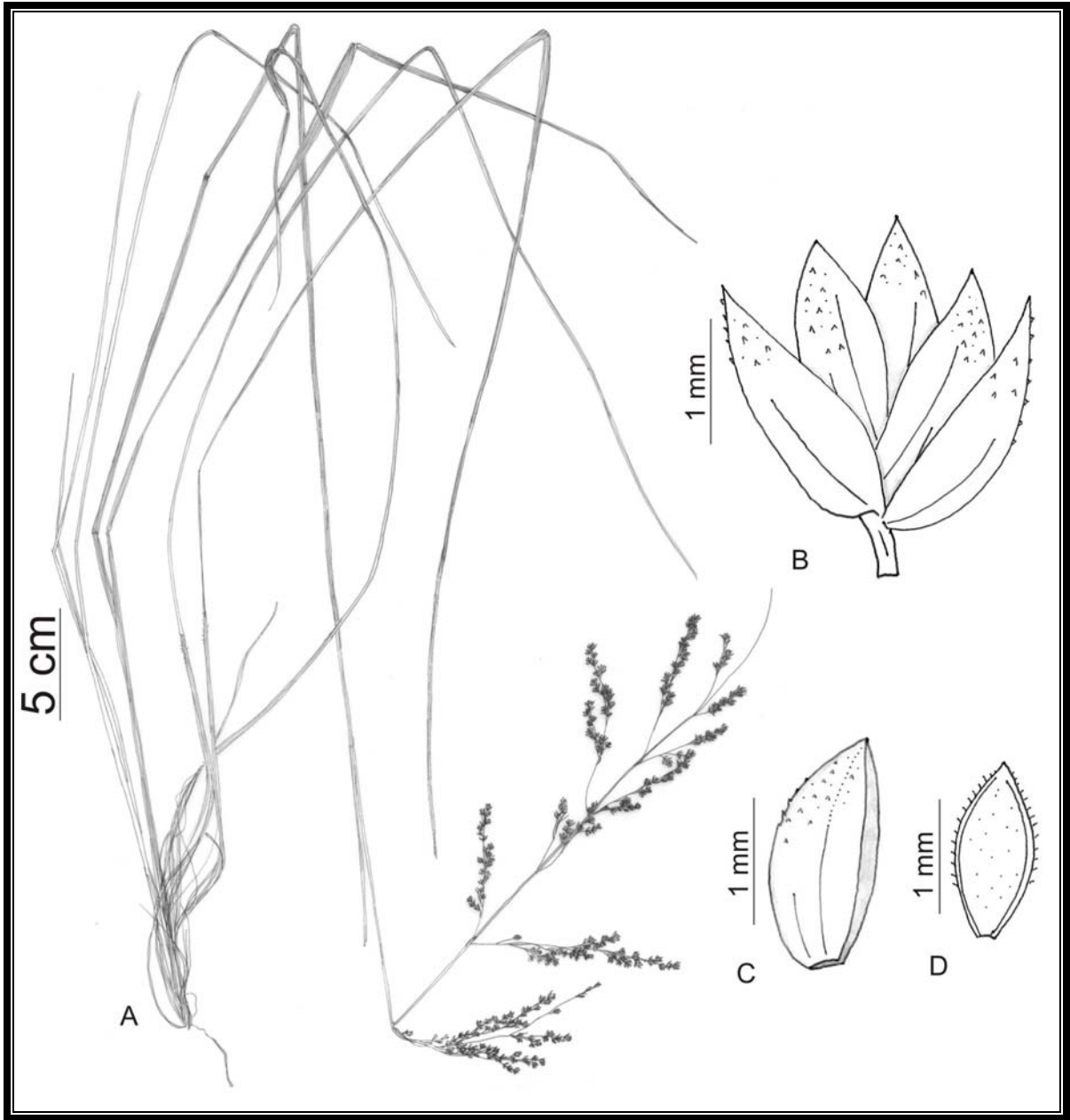


Fig. 8. *Chascolytrum brachychaetum*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (**A.** *Essi et al.* 296, ICN; **B** – **D.** *Essi* 300, ICN)

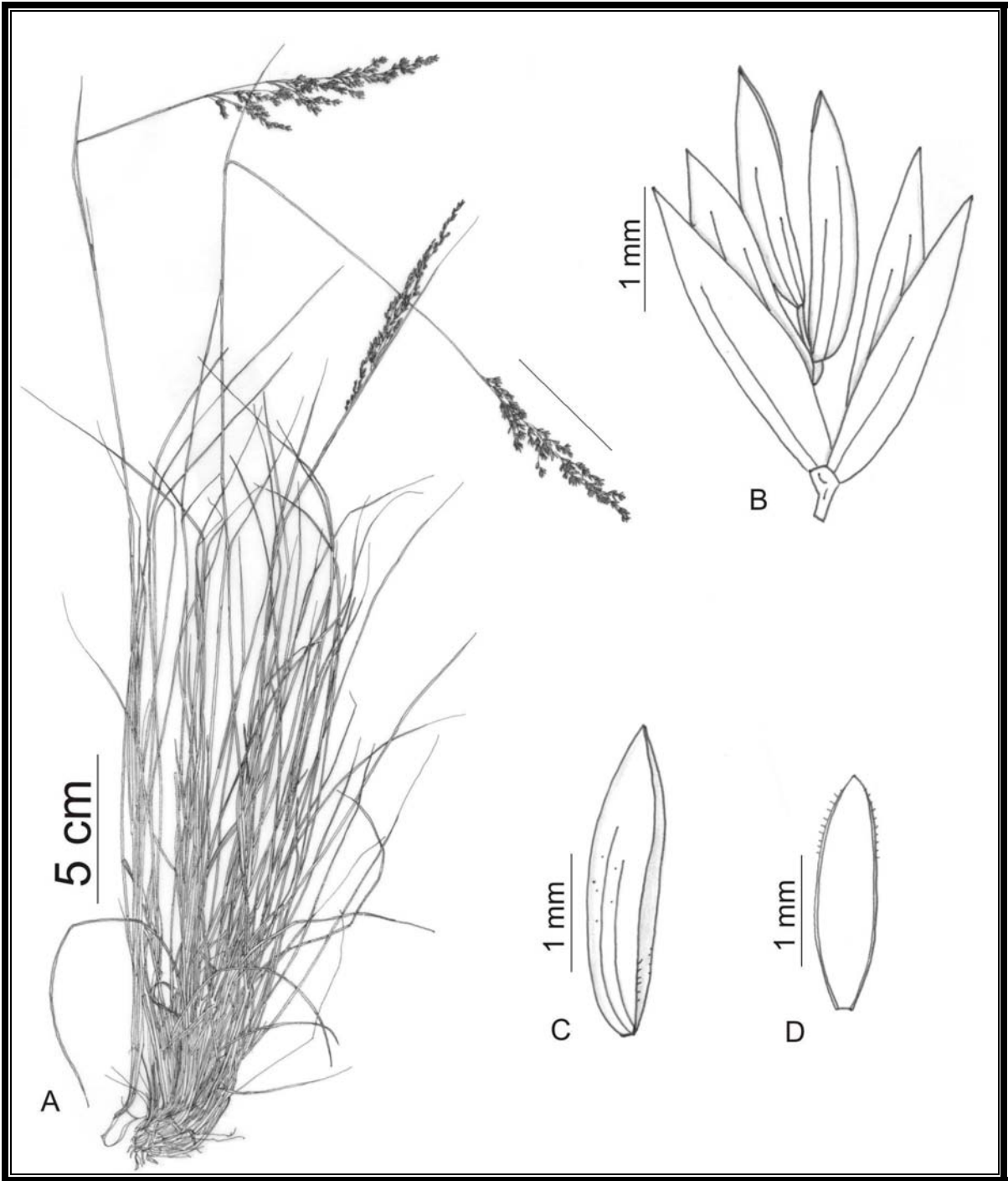


Fig. 9. *Chascolytrum brasiliense*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. Moura et al. 349, ICN)

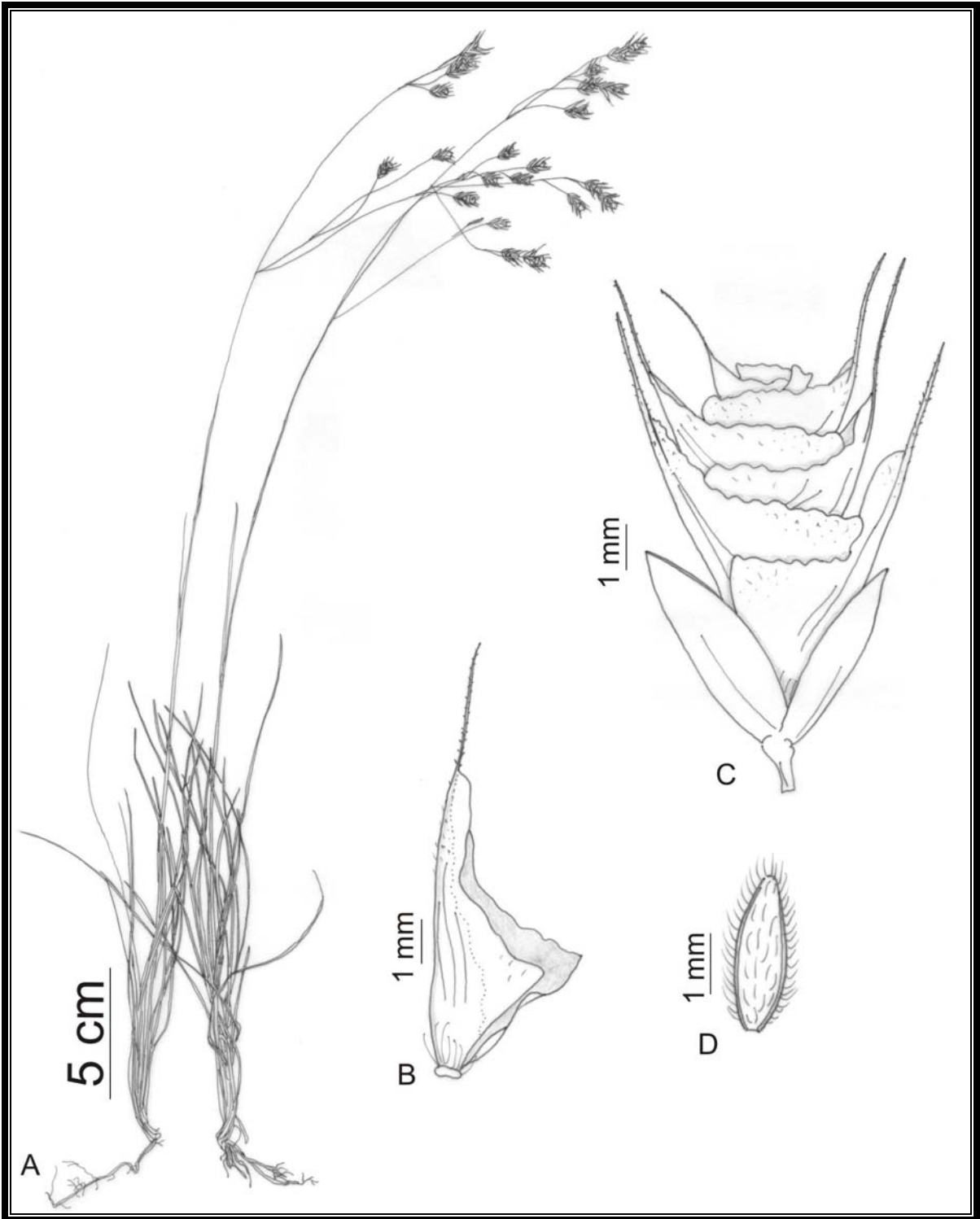


Fig. 10. *Chascolytrum brizoides*. **A.** Habit. **B.** Lemma (lateral view). **C.** Spikelet. **D.** Palea. (A – D. Gómez, Leguizamón & Martínez, ICN 25858)

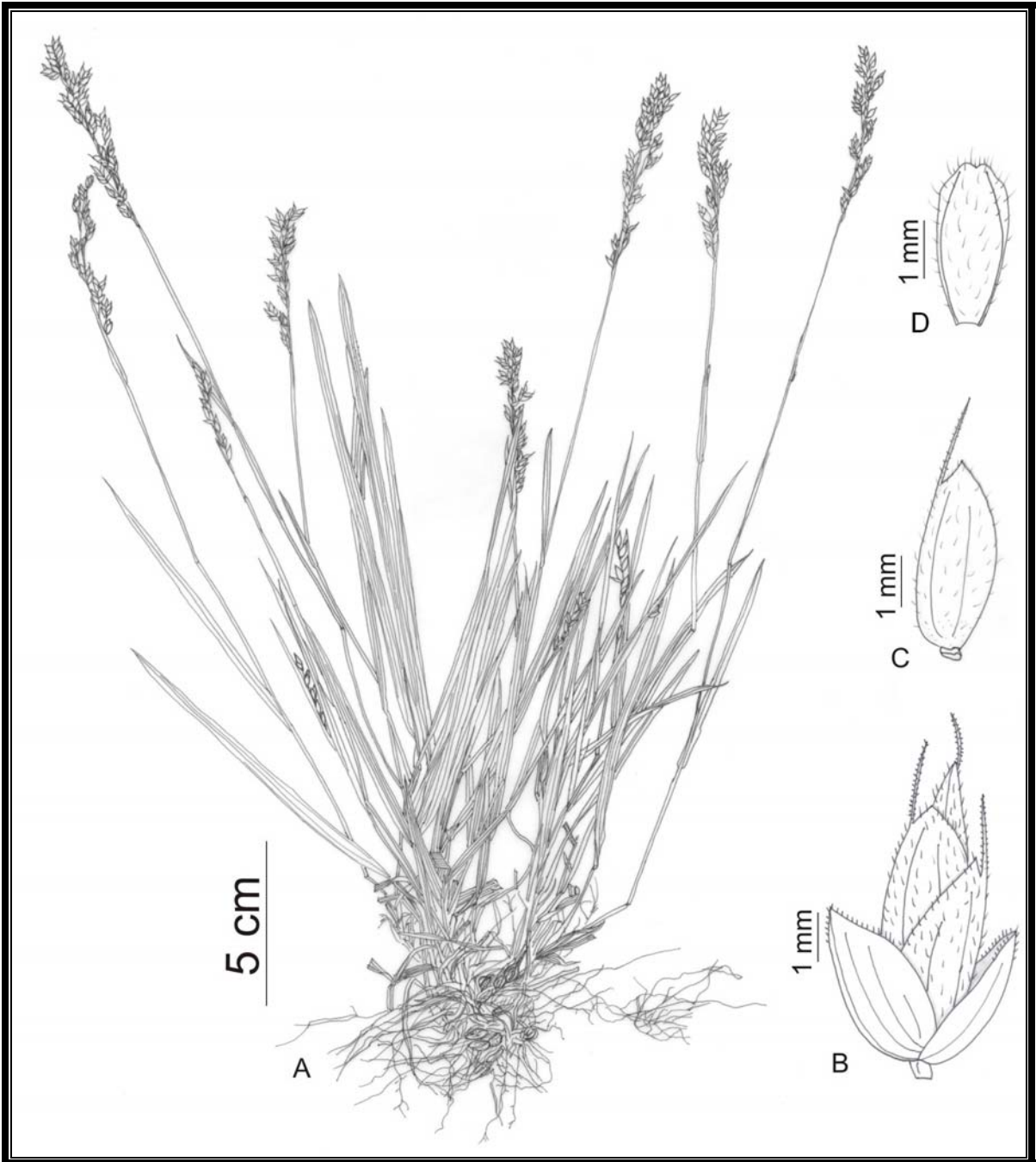


Fig. 11. *Chascolytrum bulbosum*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea..
(A – D. *Essi et al.* 284, ICN)

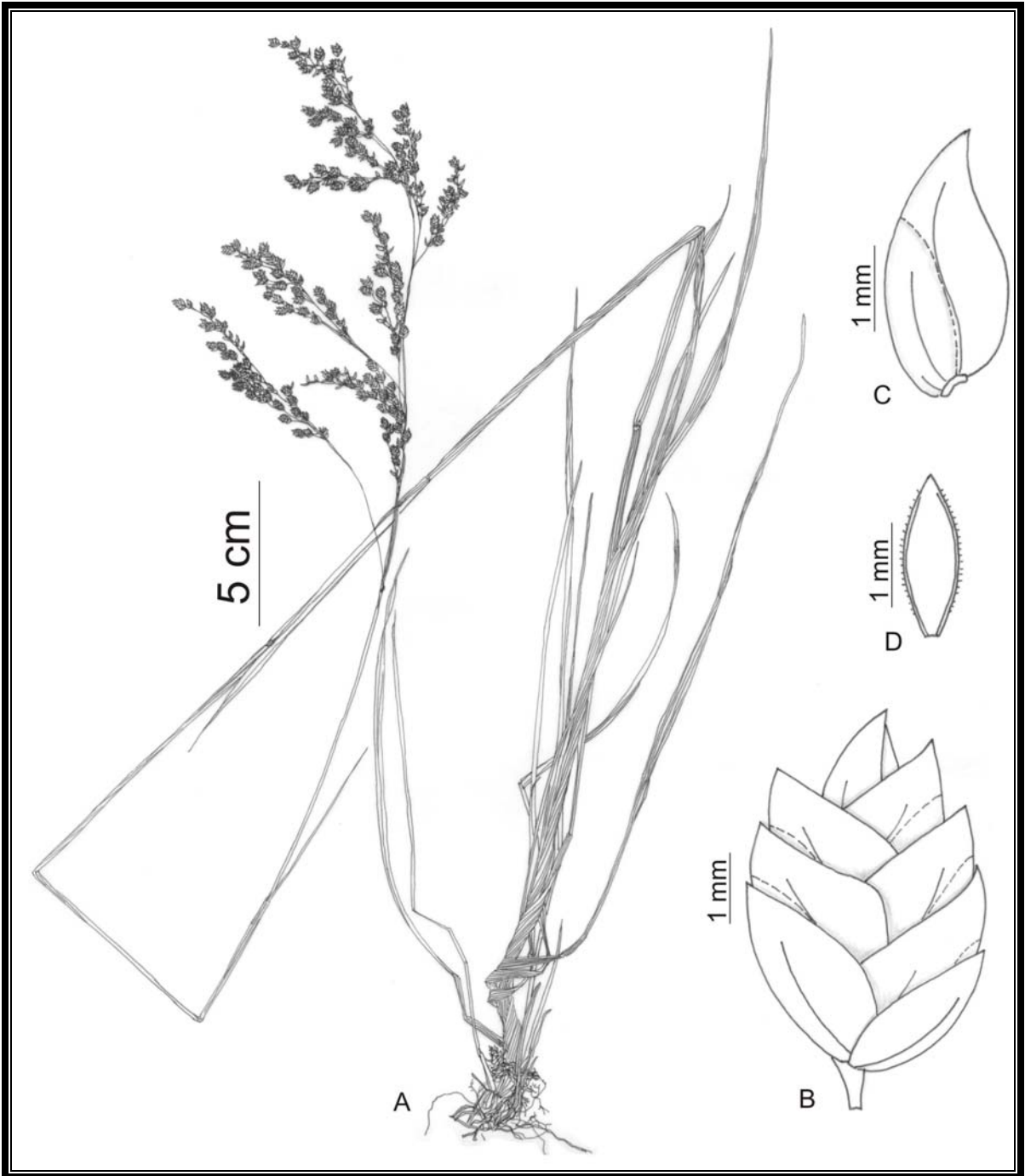


Fig. 12. *Chascolytrum calothea*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. *Essi et al.* 140, ICN)

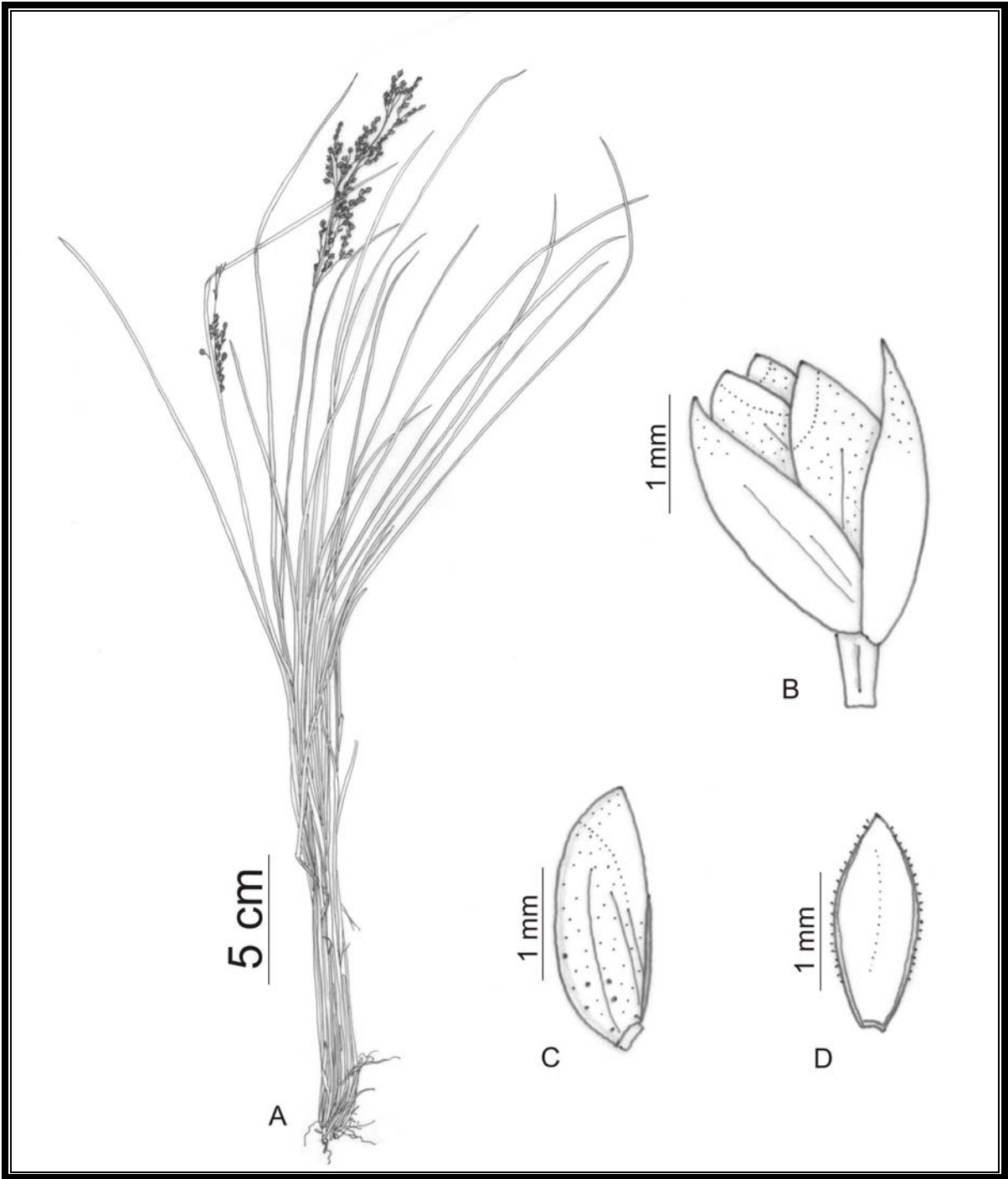


Fig. 13. *Chascolytrum itatiaiae*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. Winge et al. 1548, ICN)

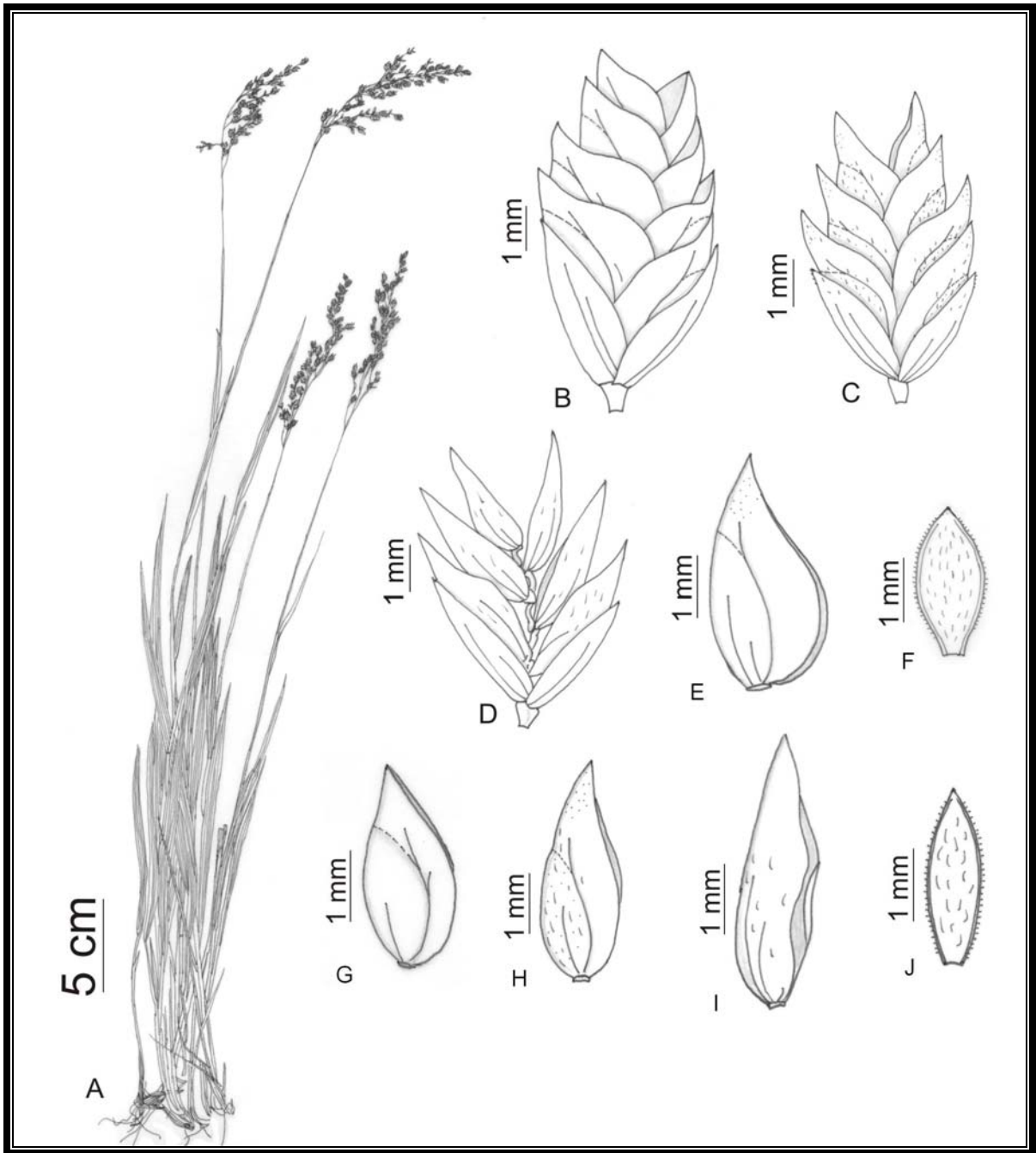


Fig. 14. *Chascolytrum juergensii*. **A.** Habit. **B - D.** Spikelet variation: **B, C.** *Chascolytrum juergensii* var. *juergensii*; **D.** *Chascolytrum juergensii* var. *angustilemma*. **E, G, H.** Lemma variation (lateral view): **E, G, H.** *Chascolytrum juergensii* var. *juergensii*; **I.** *Chascolytrum juergensii* var. *angustilemma*. **F, J.** Palea variation. (**A, C, H.** Longhi et al 7204, ICN. **B, E, F.** Essi et al. 40, ICN. **D, I, J.** Essi 122, ICN. **G.** Paz 118, ICN).

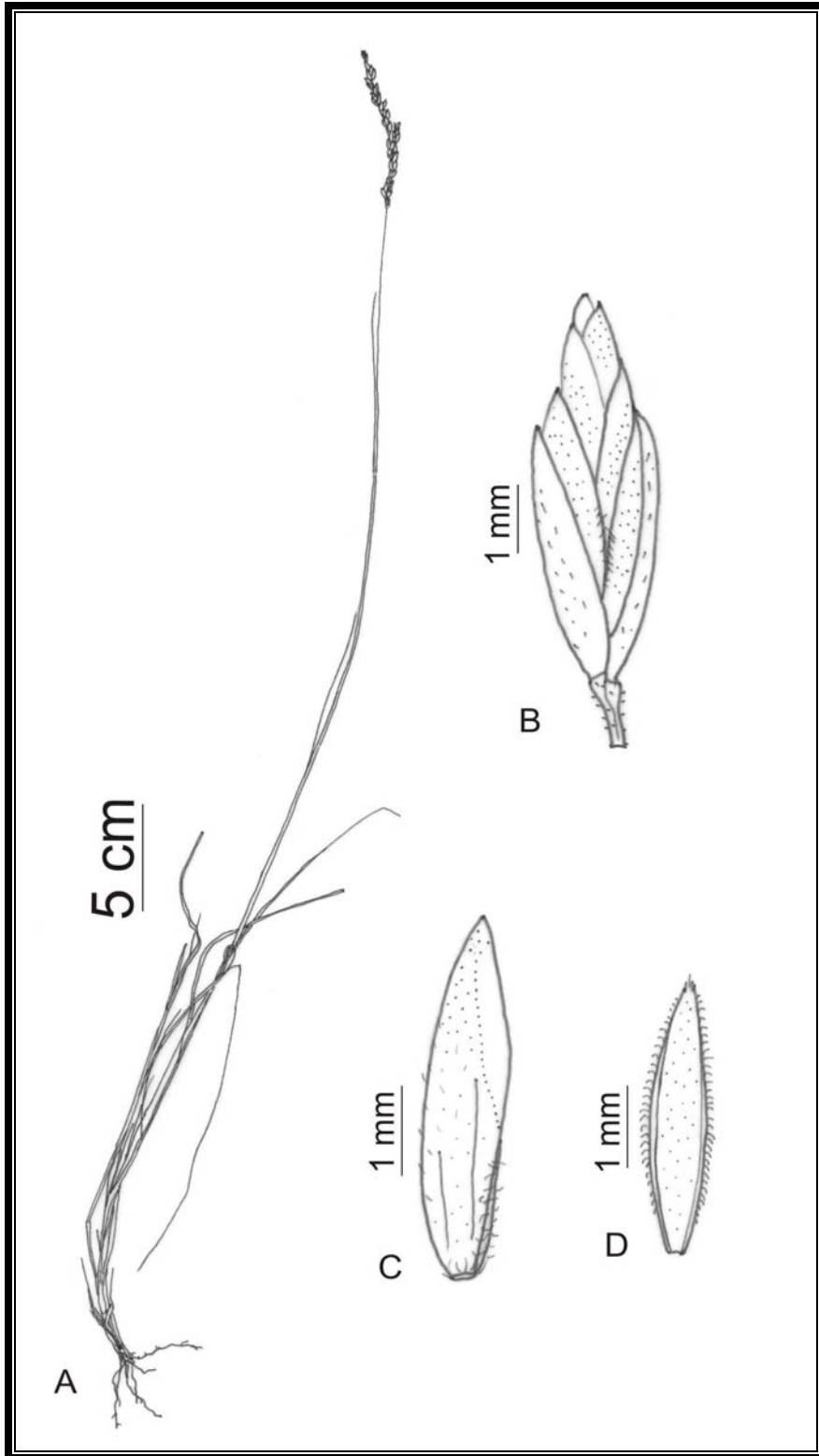


Fig. 15. *Chascolytrum koelerioides*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. Ryves & Clement 96CE/167, K).

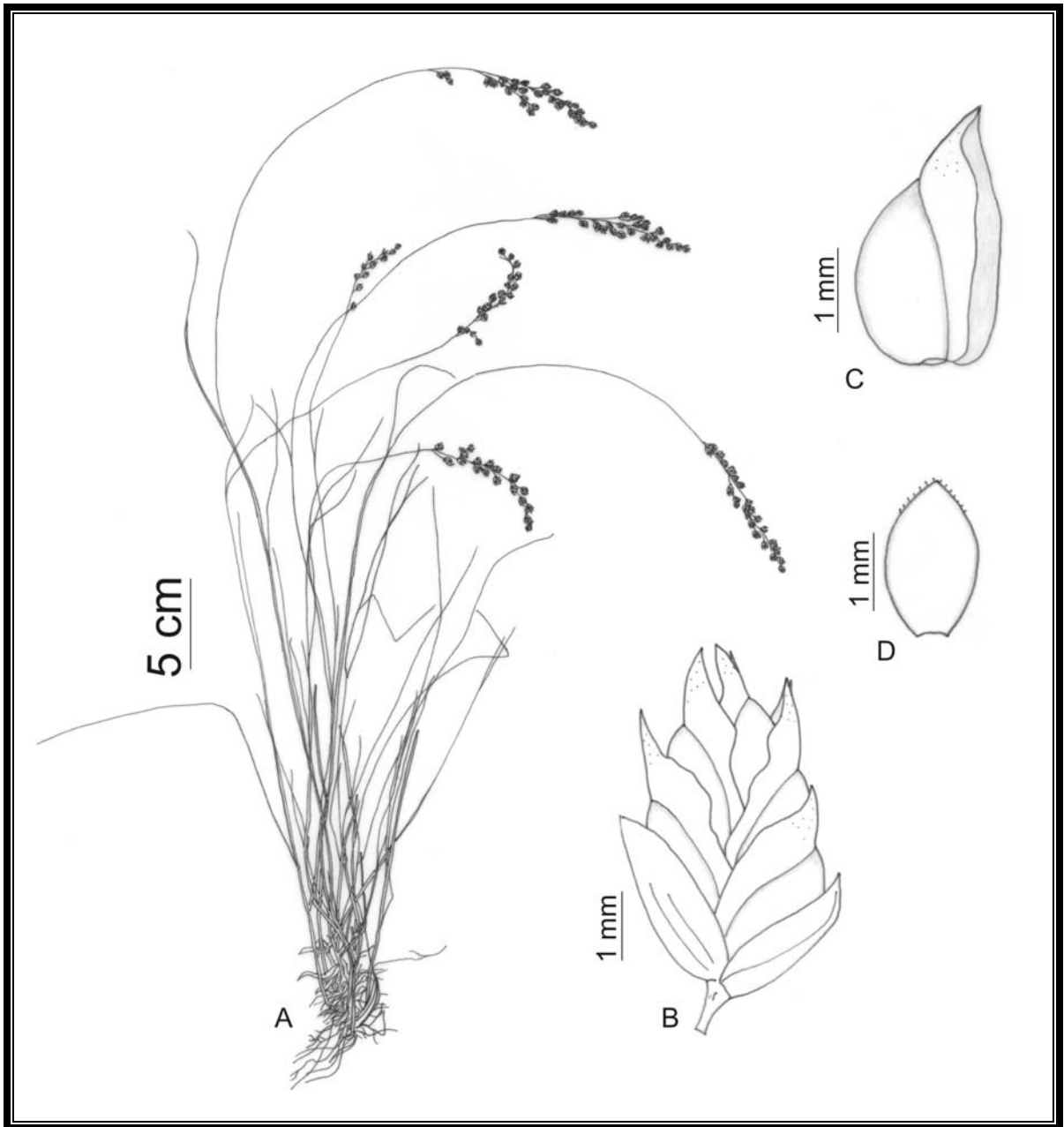


Fig. 16. *Chascolytrum lamarckianum*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. *Essi et al.* 206, ICN)

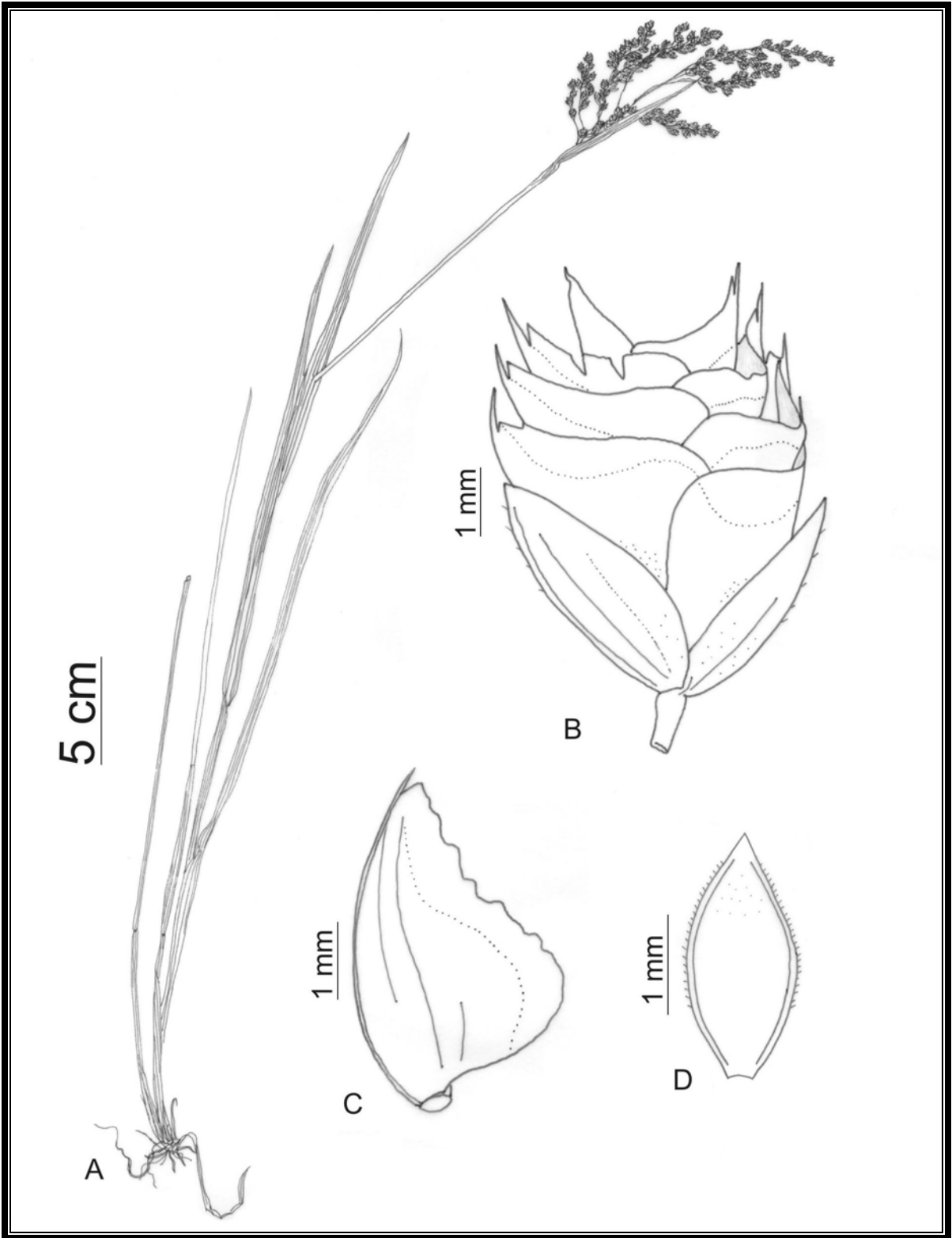


Fig. 17. *Chascolytrum latifolium*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. Longhi-Wagner et al. 10228, ICN)

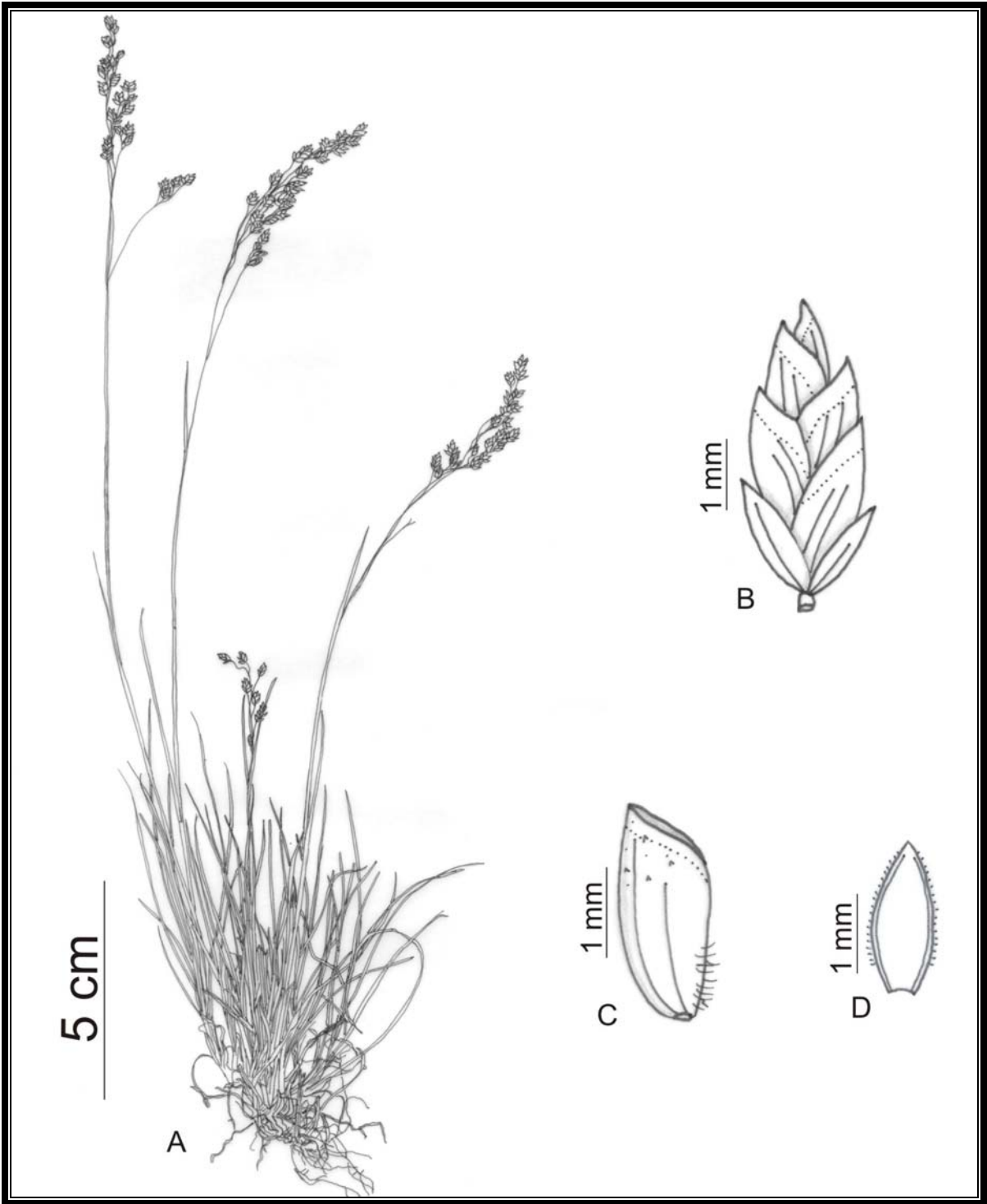


Fig. 18. *Chascolytrum monandrum*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (**A.** *Beck* 30397, ICN. **B.** *Holm-Nielsen et al.* 5081, S. **C – D.** *Beck* 7784, K)

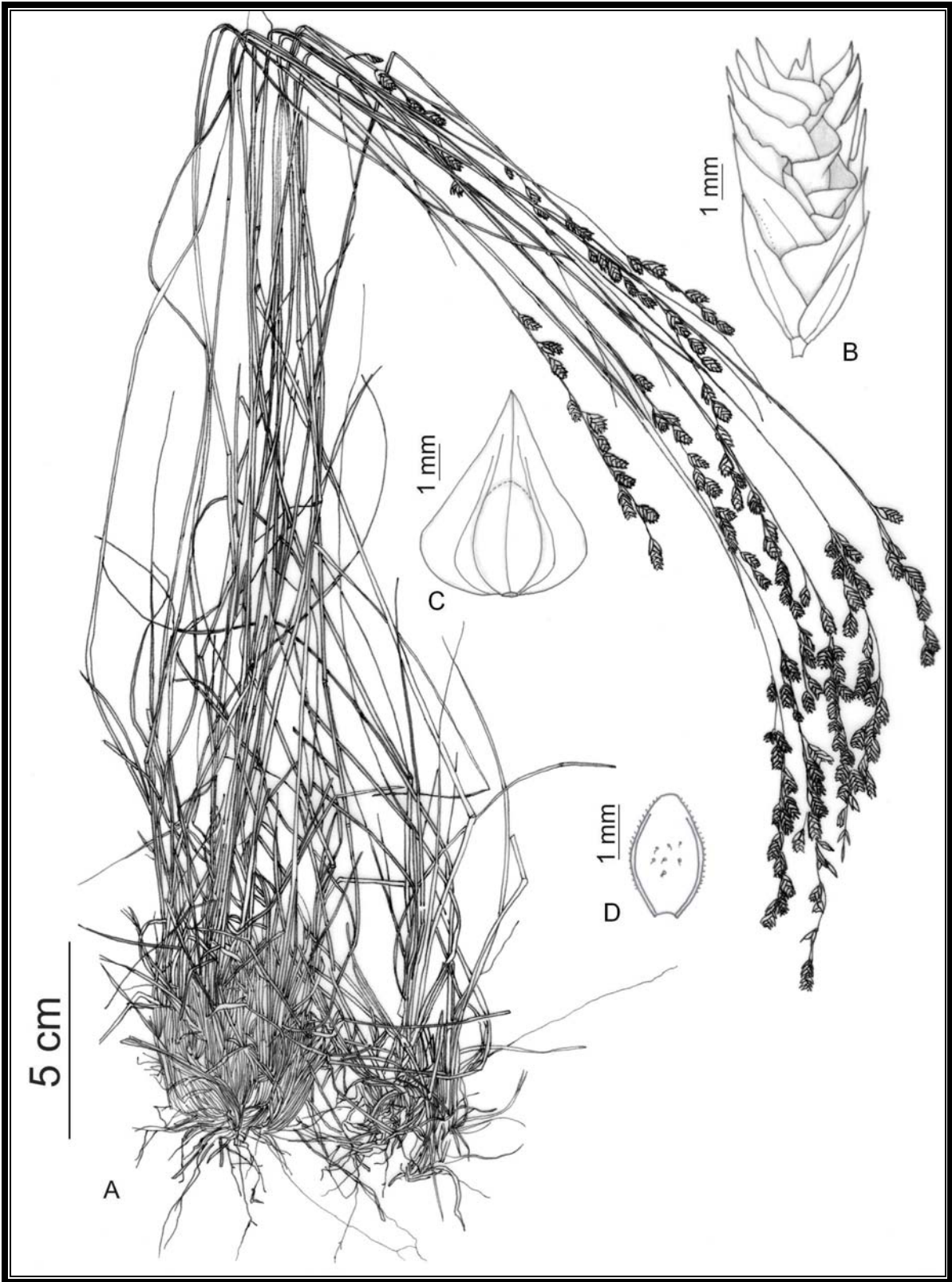


Fig. 19. *Chascolytrum paleopiliferum*. **A.** Habit. **B.** Spikelet. **C.** Lemma (dorsal view). **D.** Palea. (A - D. Renvoize 3775, K)

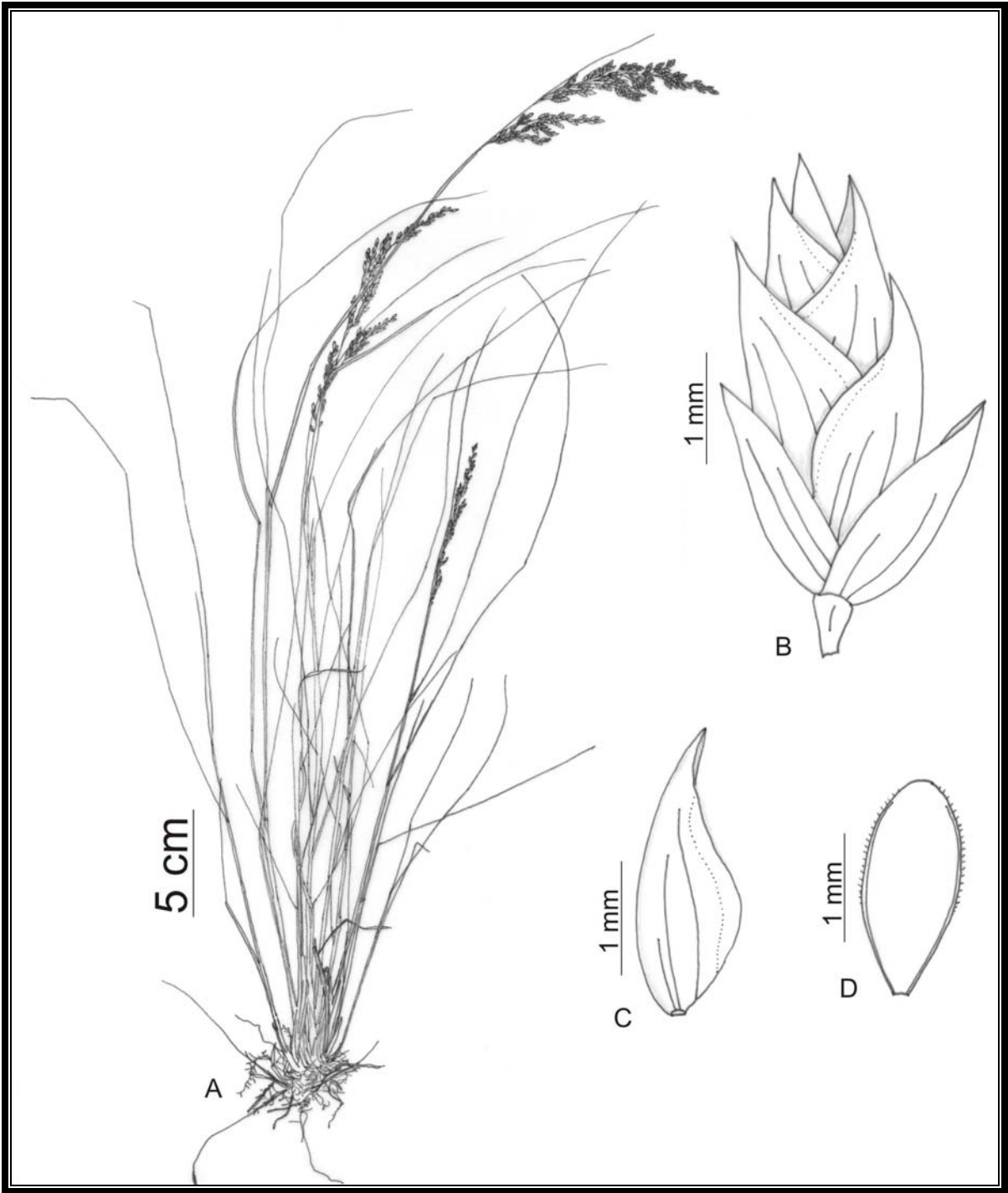


Fig. 20. *Chascolytrum parodianum*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (**A.** *Essi et al.* 292, ICN. **B – D.** *Essi et al.* 290, ICN)

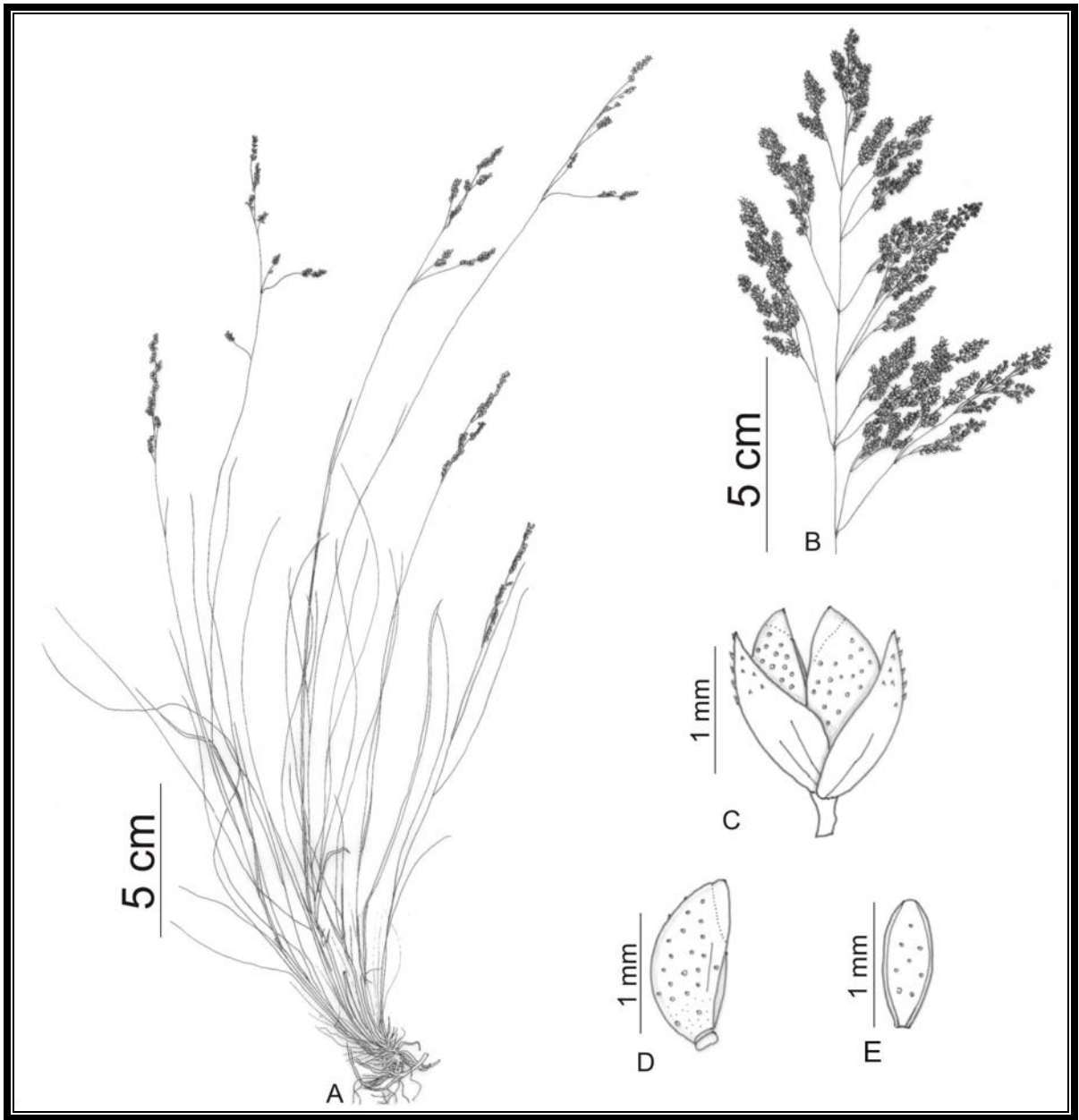


Fig. 21. *Chascolytrum poaemorphum*. **A.** Habit. **B.** Detail of a contracted panicle. **C.** Spikelet. **D.** Lemma (lateral view). **E.** Palea. (**A.** *Essi et al.* 102, ICN. **B.** *Essi et al.* 65, ICN. **C - D.** *Essi et al.* 208, ICN).

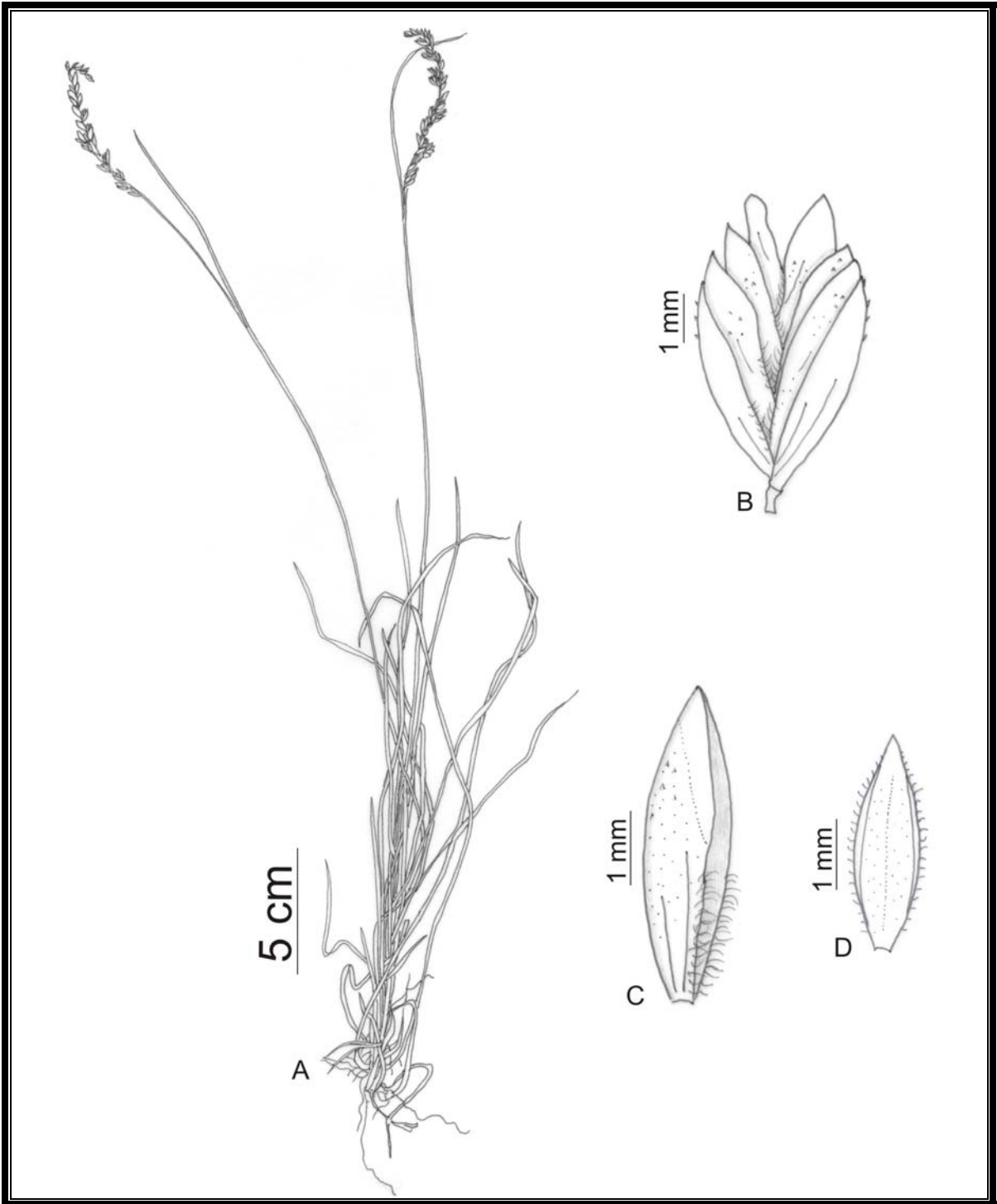


Fig. 22. *Chascolytrum rhomboideum*. **A.** Habit. **B.** Spikelet. **C.** Lemma (lateral view). **D.** Palea. (A – D. *Montero 488, K*).

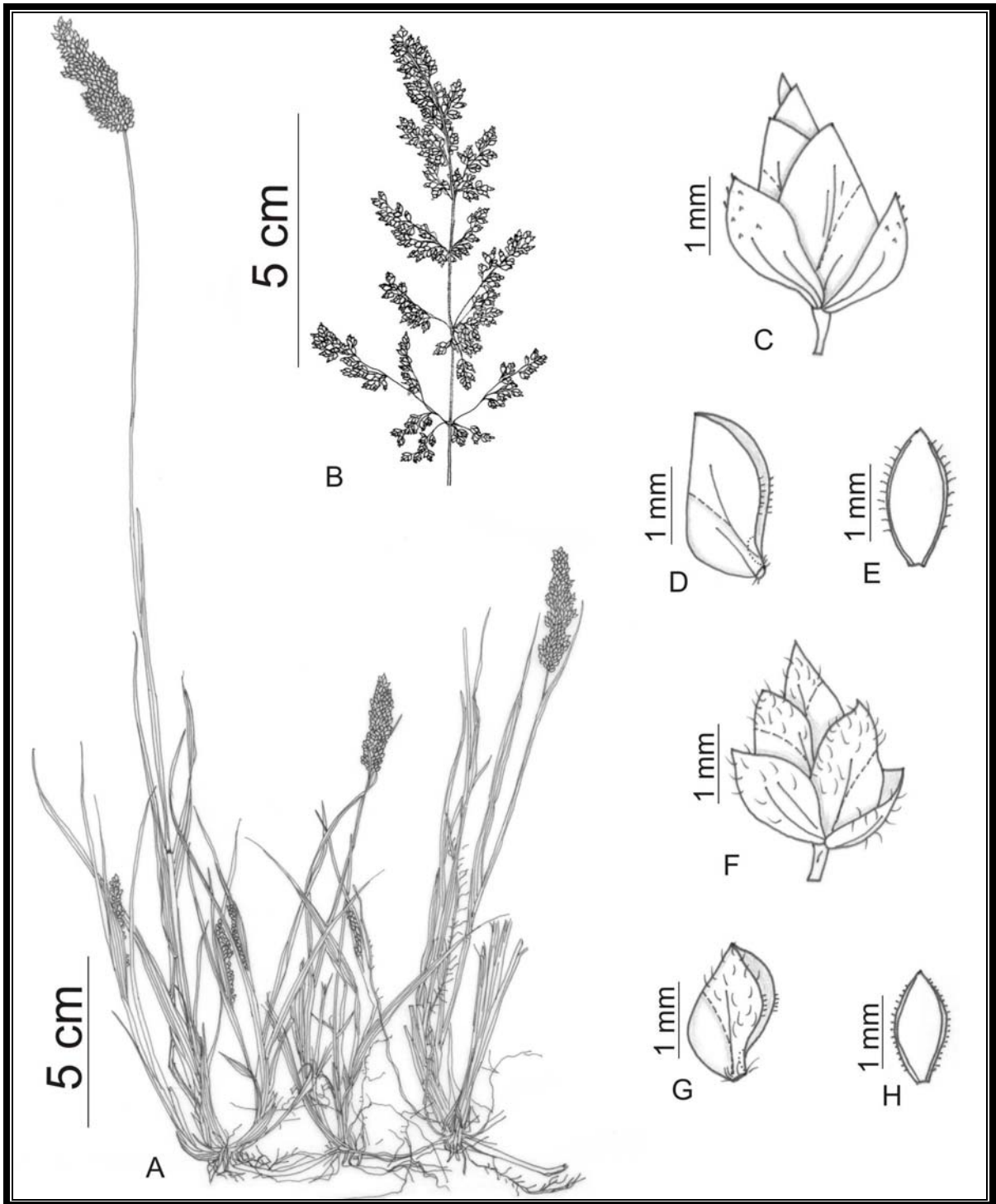


Fig. 23. *Chascolytrum rufum*. **A.** Habit. **B.** Detail of an open panicle. **C – E.** *Chascolytrum rufum* var. *rufum*: **C.** Spikelet; **D.** Lemma (lateral view); **E.** Palea. **F – H.** *Chascolytrum rufum* var. *sparsipilosum*: **F.** Spikelet; **G.** Lemma; **H.** Palea. (**A.** Essi et al. 120, ICN. **B.** Essi et al. 281, ICN. **C – E.** Longhi-Wagner et al. 10146, ICN. **F – G.** Essi et al. 96, ICN).

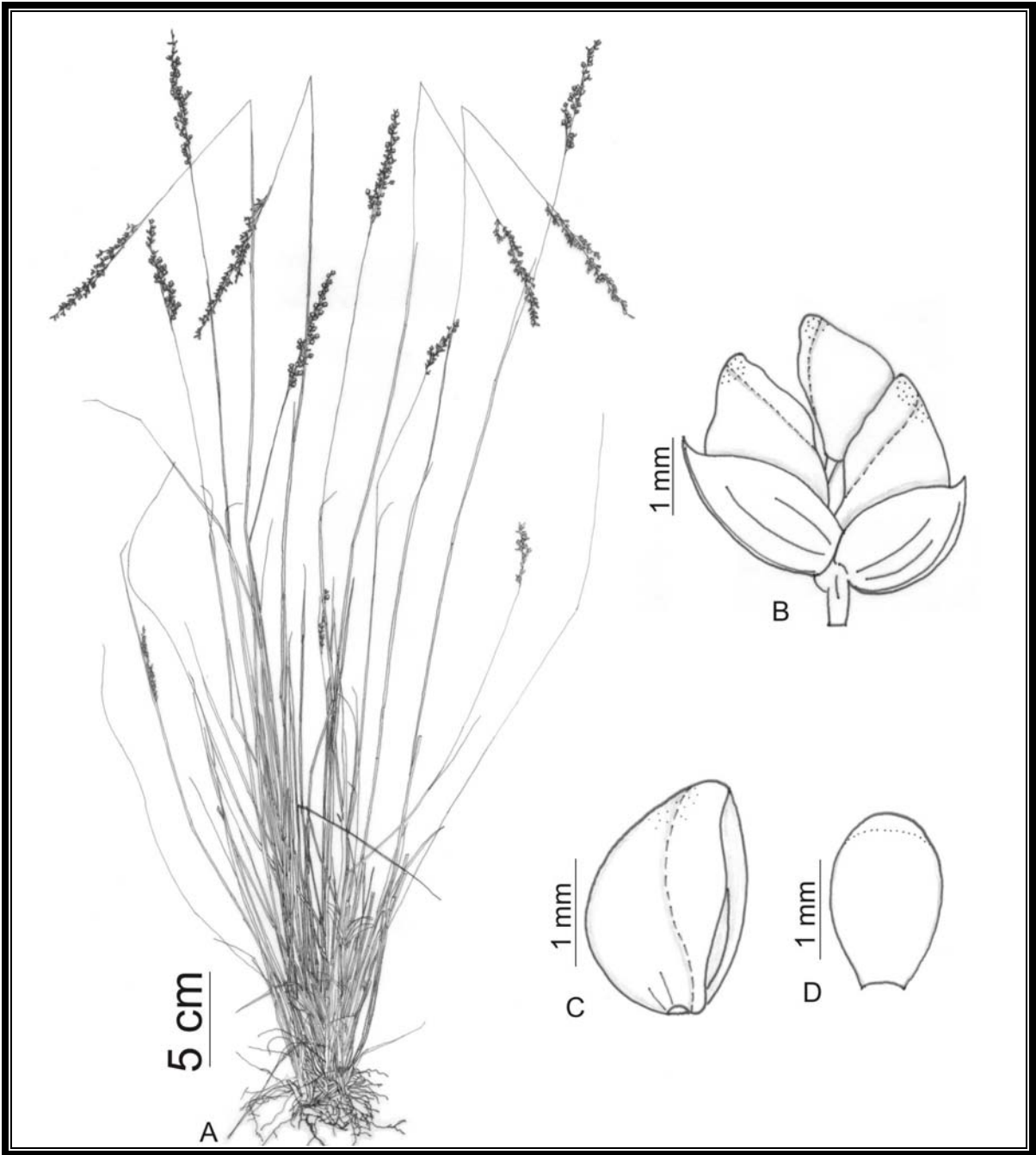


Fig. 24. *Chascolytrum scabrum*. **A.** Habit. **B.** Spikelet. **C.** Lemma. **D.** Palea. (A – D. Essi et al. 101, ICN)

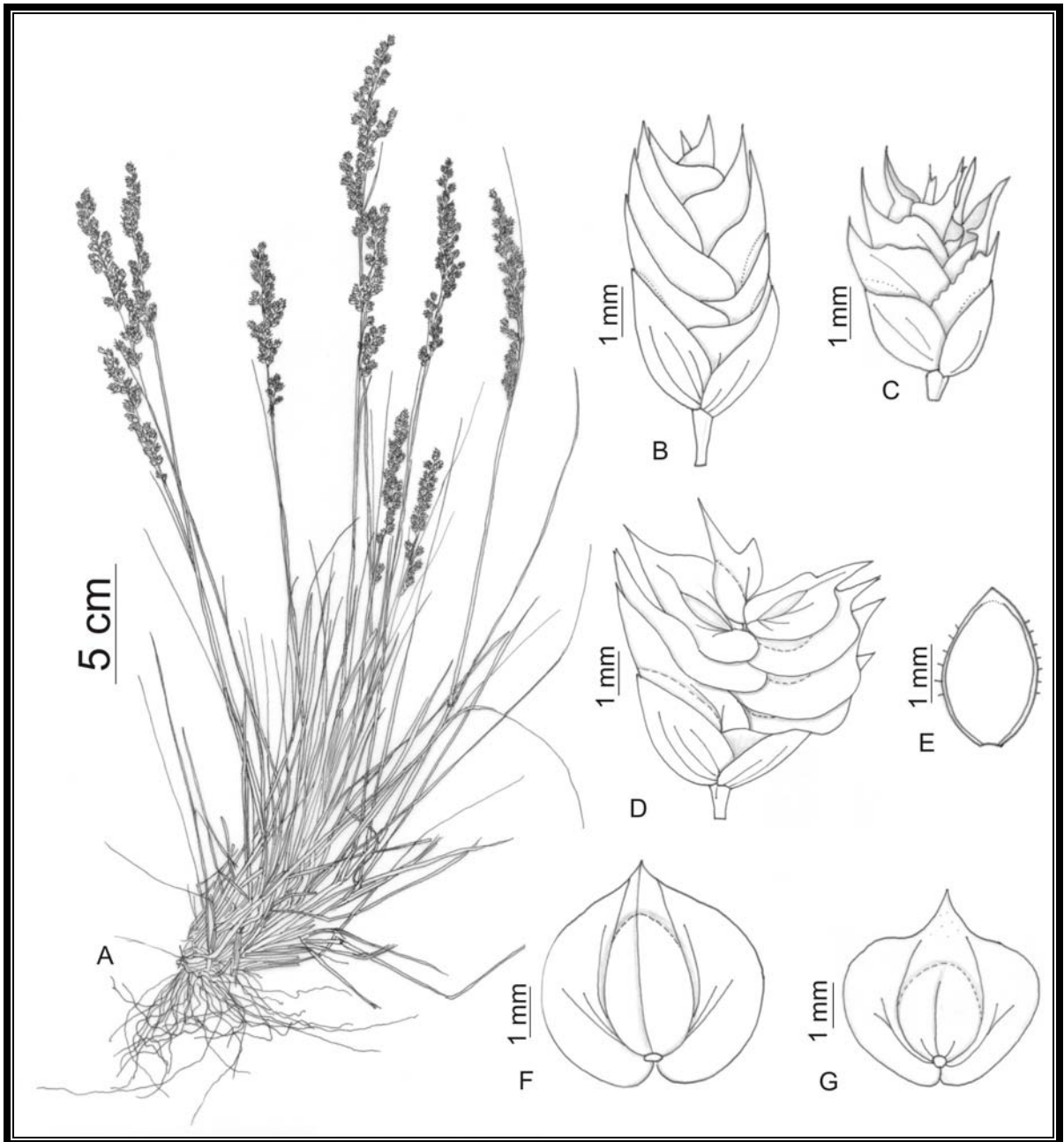


Fig. 25. *Chascolytrum subaristatum*. **A.** Habit. **B - D.** Spikelet variation. **E.** Palea. **F - G.** Lemma. (**A.** *Essi et al.* 19, ICN. **B, G.** *Essi et al.* 94, ICN. **C.** *Essi et al.* 177, ICN. **D - F.** *Essi et al.* 181, ICN).

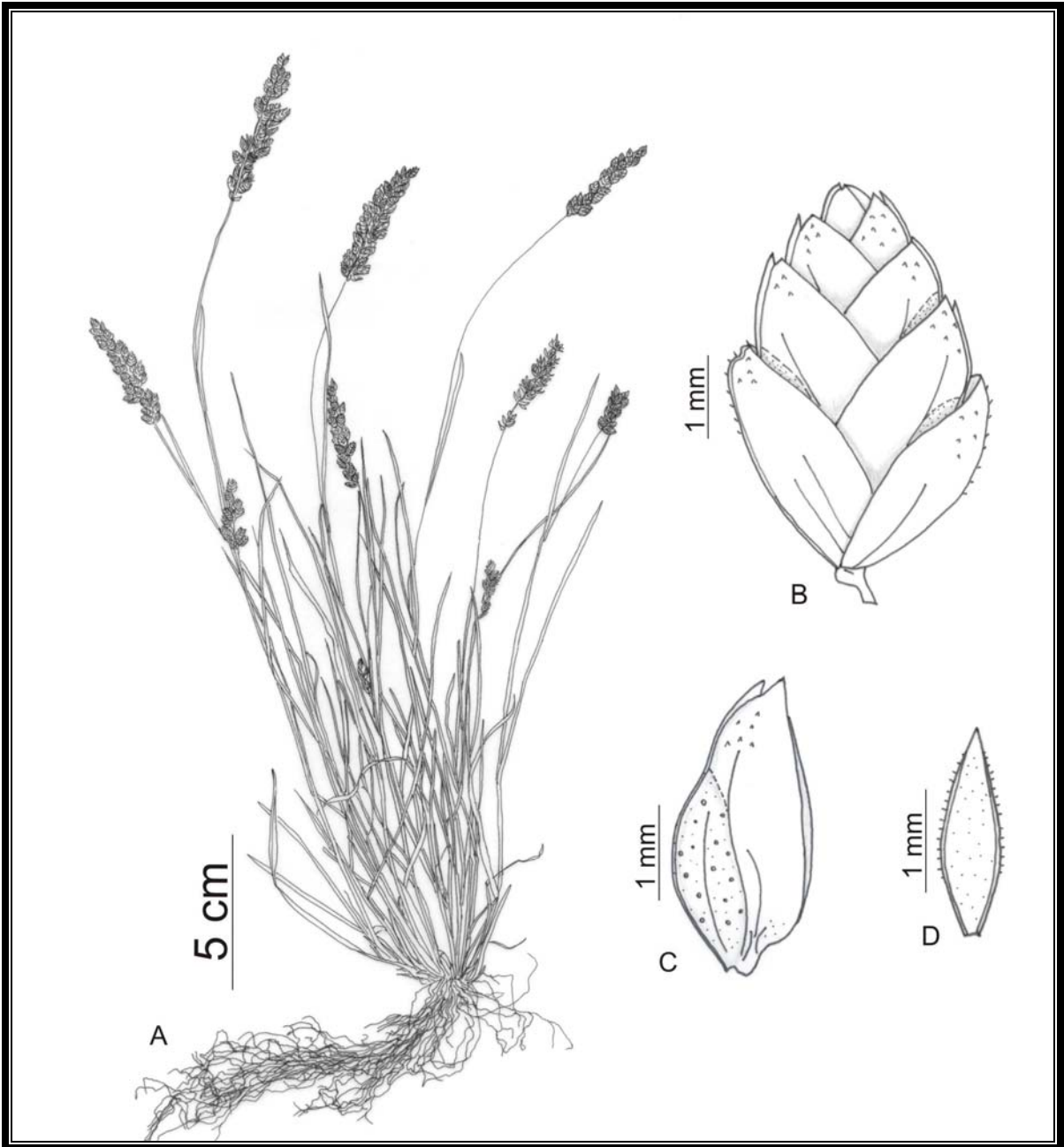


Fig. 26. *Chascolytrum uniolae*. **A.** Habit. **B.** Spikelet. **C.** Lemma. **D.** Palea. (**A.** Lima 200, ICN. **B - D.** Essi et al. 164, ICN)

References

- Arechavaleta, J. 1897. Las Gramineas uruguayas. *Anales Mus. Nac. Montevideo* 1: 469.
- Bayón, N. D. 1998. Cladistic analysis of the Briza Complex (Poaceae, Poaeae). *Cladistics* 14: 287 - 296.
- Beauvois, P. 1812. *Essai d'une nouvelle Agrostographie*. Paris.
- Bentham, G. & Hooker, J. D. 1883. *Genera Plantarum*. Bd. 3. London.
- Bieberstein, M. 1808. *Flora Taurico-Caucasica* 1: 66.
- Brummit, B.K. & Powell, C. E. 1992. *Authors of plant names*. Royal Botanic Gardens, London.
- Burman, N. L. 1768. *Fl. Indica* 3.
- Cavanilles, A. J. 1803. *Elenchus Plantarum Horti Regni Botanici Matritensis* 28. 1803.
- Clayton, W.D., Renvoize, S.A. 1986. *Genera graminum: grasses of the world*. Royal Botanic Gardens, London.
- Cvelev, N.N. 1976. *Zlaki SSSR*. Nauka, Leningrad.
- Chrtek, J. & Hadač, E. 1969. *Candollea* 24: 170.
- Desvaux, A. N. 1810. Extrait d'un Mémoire sur quelques nouveaux genres de la famille des Graminées. *Nouv. Bull. Sci. Soc. Philom. Paris* 2: 187 - 190.
- Desvaux, A. N. 1813. Mémoire sur les Graminées, et sur quelques genres et espèces nouvelles de cette famille. *J. Bot.* 1: 63 - 77.
- Desvaux, E. 1853. Gramineas. In Gay (ed.): *Historia física y política de Chile*. Bd. 6: 233 - 469. Paris.
- Doell, J. C. 1878. Gramineae. In: Martius (ed.): *Flora Brasiliensis*. Monachii, Fleischer. T. 2 pt. 3.

- Doell, J. C. 1883. Gramineae II. In Martius (ed.): Flora Brasiliensis. Bd. 2, 3. Teil. Leipzig.
- Ekman, E. L. 1913. Die Gräser des brasilianischen Staates Paraná. Ark. Bot. 13, 10: 1 - 83.
- Gouan, A. 1762. Hortus Regius Monspeliensis 45. 1762.
- Grisebach, A., 1879. Symbolae ad Floram argentinam, Zweite Bearbeitung argentinischer Pflanzen. Gramineae. Abh. Königl. Ges. Wiss. Göttingen 24: 284 - 300.
- Hackel, E., 1902. Neue Gräser. Österr. Bot. Z. 52, 8: 303 - 310.
- Hackel, E., 1915: Neue Gräser aus Brasilien. Verh. K. K. Zool. Bot. Ges. Wien 65: 70 - 77.
- Heister, L. 1759. Enumeratio Methodica Plantarum 207.
- Henrard, J. T. H. 1921. Die von Dr. Th. Herzog auf seiner zweiten Reise durch Bolivien in den Jahren 1910 und 1911 gesammelten Pflanzen. Gramineae. Meded. Rijks-Herb. 40: 39 - 77.
- Hitchcock, A. S. 1923. Type species of the first 100 genera of Linnaeus' Species plantarum. Am. J. Botany 10: 510-516.
- Hubbard, C. E. 1970. *Briza*. Pp. 51-53. In Milne –Redhead, E. & Polhill, R. M. (ed.): Flora od tropical East Africa, Gramineae. Crown Agents, London.
- Jirásek, V. & Chrtek, J. 1966. *Brizochloa*, eine neue Graspattung. Bot. Delect. Seminum Horti Bot. Carol. Prag. 39 – 41.
- Jirsek, V. & Chrtek. 1966 [1967]. Novit. Bot. & Del. Sem. Hort. Bot. Univ. Carol. Prag. 1966: 40. 1966 [1967].
- Kergélen, M. 1975. Les Gramineae (Poaceae) de la flore française. Essai de mise au point taxonomique et nomenclaturale. Lejeunia, ser. 2, 75: 1-343.
- Knapp, J. L. 1804. Gramina Britannica pl. 61.
- Kneucker, A. 1902. Bemerkungen zu den Gramineae exsicatae. Allgemeine Botanische Zeitschrift. P. 97.

- Kunth, C. S. 1815 [1816]. *Nova Genera et Species Plantarum* (quarto ed.) 1: 152-153.
- Kunth, C. S. 1833. *Enumeratio Plantarum*. Bd. 1. Stuttgart und Tübingen.
- Kuntze, O. 1898. *Revisio Generum Plantarum*. 3, 2: 1 –576. Würzburg.
- Lamarck, J. B. 1778. *Flore française* 3: 586
- Lamarck, J. B. 1791. *Tabl. Encycl.* Bd. 1. Paris.
- Linnaeus, C. 1753. *Species Plantarum*. Bd. 1. Stockholm.
- Linnaeus, C. 1762. *Species Plantarum, Editio Secunda* 1: 103.
- Link, J.H.F. 1833. *Hort. Berol.* 2: 296.
- Longhi-Wagner, H. M. 1987. *Flora Ilustrada do Rio Grande do Sul*, Fasc. 17. Gramineae. Tribo Poeae. *B. Inst. Bioc.* 41, 1 - 191.
- Longhi-Wagner, H. M., Wanderley, M. G. L. & Scheferd, J. 2001. *Flora Fanerogâmica do Estado de São Paulo- Vol. 1: Poaceae*. Hucitec, São Paulo.
- Matthei, O. 1975. Der Briza-Komplex in Südamerika: *Briza*, *Calotheca*, *Chascolytrum*, *Poidum* (Gramineae). *Willdenowia*, 8: 7 – 168.
- Meikle, R. D. 1985. *Flora of Cyprus*, 2. Kew.
- Nash, G. V. 1912. Poaceae [part]. Pp 99-196 in Britton, N. L. & Underwood, L. M. (ed.), *North American Flora* 17. New York Botanical Garden, New York.
- Nees, C. G. 1829. *Agrostologia Brasiliensis*. Stuttgart & Tübingen.
- Nees, C. G. 1836. In Lindley (ed.): *A Natural System of Botany*. 2. Aufl. London.
- Nicolson, D. H. Seventy –two proposals for the conservation of types of selected Linnaean generic names, the report of Subcommittee 3C on the lectotypification of Linnaean generic names. *Taxon*, 41: 552-583.
- Nicora, E. G. & Rógulo de Agrasar, Z. E. 1981. Los géneros sudamericanos afines a *Briza* L. (Gramineae). *Darwiniana*, 23 (1): 279 – 309.

- Parodi, L. R. 1920. Notas sobre las especies de Briza de la Flora Argentina. Rev. Fac. Agr. Y Vet. B. Aires, 3: 113 – 137.
- Pilger, R. in Werdermann 1929. Plantae Raimondianae VI. Notizbl. Bot. Gart. Berlin-Dahlem 10: 725.
- Presl, J.S., 1830. Reliq. Haenk. Bd. 1. Prag.
- Prokudin, J. N. 1954. Bot. Mater. Gerb. Inst. Bot. Akad. Nauk Kazahsk. SSR 16: 42, 43. 1954.
- Rosengurt, B. & Arrillaga de Maffei, B. 1979. Lombardochloa, nuevo genero de Gramineae. Anales Fac. Quím., 9: 255 – 288.
- Rosengurt, B., Arrillaga de Maffei, B. R., et Izaguirre de Artucio, P., 1968. Sinopsis de *Briza* (Gramineae) del Uruguay y notas taxonómicas sobre otras especies de este género. Bol. Univ. Republ. Fac. Agron. Montevideo 105: 1 – 35.
- Rosengurt, B., Arrillaga de Maffei, B., Izaguirre de Artucio, P. 1970. Gramíneas Uruguaias. Universidad de la Republica, Departamento de Publicaciones. Montevideo.
- Sherif, A. S. & Siddiqi, M. A. 1988. Poaceae. In: El-Gadi, A. A. (ed.), Flora of Libya, 145. Al Faateh University, Tripoli.
- Sibthorp, J. & Smith, J. E. 1806. Flora Graeca 1: 60, t. 77.
- Stearn, W. T. 1992. Botanical Latin. 4.ed. Timber Press, Portland Oregon.
- Studel, E. G. 1840. Nomenclatur Botanicus. 2. Aufl. Bd. 2. Stuttgart & Tübingen.
- Studel, E. G. 1854. Synopsis Plantarum Glumacearum. Bd. 1. Stuttgart.
- Townsend & Guest (eds.), 1968. Flora of Iraq. Vol. 9. Ministry of Agriculture of the Republic of Iraq.
- Trinius, C. B. 1831. Graminum Genera quaedam speciesque complures definitionibus novis. Mém. Acad. Imp. Sci. St. Petersburg, Sér. 6, Sci. Math. 4, 1: 353 – 416.

- Trinius, C. B. 1836. Mém. Acad. Imp. Sci. St. Petersbourg, Sér. 6, Sci. Math. 4,2(1): 62.
- Tzvelev, N. N. 1970. Novosti Sistematiki Vysshchikh Rastenii 21.
- Tzvelev, N. N. 1983. Grasses of the Soviet Union. Part II. Oxonian Press. New Dehli.
- Tzvelev, N. N. 1987. Komarovskie Čtenija (Moscow & Leningrad) 37: 32.
- Tzvelev, N. N. 1993. Botaničeskij Žurnal (Moscow & Leningrad) 78(10): 91.
- Woronow, G. ex Fedtschenko, B. A.. 1915. Izv. Imp. Bot. Sada Petra Velikago 14(Suppl. 2): 74.

Discussão geral

Como resultado deste trabalho, estão sendo aceitas 26 espécies, das quais duas com duas variedades cada. Duas espécies e uma variedade são novas para a Ciência. Cerca de 115 nomes foram incluídos ou aceitos em sinonímia. Novas combinações foram realizadas para 14 espécies aceitas, como consequência da nova circunscrição aqui proposta para o grupo, que reconhece apenas dois gêneros para o Complexo *Briza*: *Briza* L. *stricto sensu* e *Chascolytrum* Desv. *lato sensu*.

De acordo com a nova circunscrição aqui proposta, *Briza* inclui quatro espécies e *Chascolytrum* 22 espécies, das quais duas aceitas com duas variedades cada. O grupo todo se distribui principalmente em regiões temperadas ou subtropicais, e as espécies são microtêrmicas, de ciclo hibernal, e perenes, em sua maioria. Entretanto, as espécies de *Briza* são euro-asiáticas, enquanto as de *Chascolytrum* ocorrem principalmente na América do Sul, com maior riqueza específica na Região Sul do Brasil.

Todas as espécies apresentam registros de coletas nos últimos 50 anos. Porém, algumas apresentaram um número muito pequeno de registros no mesmo período, indicando sua raridade ou um importante declínio no número de populações. Foram consideradas raras as espécies: *Chascolytrum scabrum* (Nees ex Steud.) Matthei, *Chascolytrum bulbosum* (Parodi) Essi, Longhi-Wagner & Souza-Chies, *Chascolytrum parodianum* (Roseng., Arrill. & Izag.) Matthei e *Chascolytrum ambiguum* (Hack.) Essi, Longhi-Wagner & Souza-Chies, todas espécies sul-americanas. *Chascolytrum bulbosum* consta na lista de espécies ameaçadas do Rio Grande do Sul, devido à degradação de seu hábitat no Estado. Entretanto, deve ser levado em conta que esta espécie ocorre no Uruguai. Além disso, deve-se ressaltar que a inclusão de espécies com distribuição conhecida considerada muito restrita em listas de plantas ameaçadas pode ser prematura, pois

excursões de coleta mais exaustivas, por um período mais prolongado e em áreas pouco percorridas, podem revelar a ocorrência de mais populações na natureza.

Considerando o grupo euro-asiático, que se manteve em *Briza*, a espécie menos abundante é *B. humilis* M.Bieb., a qual apresenta distribuição restrita a habitats de altitude na Península Balcânica e da região do Cáucaso, limite sudoeste entre Europa e Ásia. Esta espécie foi bastante coletada do início do século passado até a década de 80, porém os registros mais recentes são escassos nos herbários.

A seguir, serão discutidos os principais resultados obtidos no que concerne à circunscrição dos táxons estudados neste trabalho.

Diferenças interespecíficas

Além da problemática referente à circunscrição em nível genérico, alguns complexos de espécies morfológicamente afins merecem ser discutidos.

Diversos complexos já foram abordados por Torres de Lemos (1983), Hickenbick (1983), Schifino-Sampaio (1979), Schifino & Winge (1979) e Leyser & Winge (1979), utilizando abordagens não macro-morfológicas. Nem todas as abordagens produziram resultados concordantes, sendo os resultados mais marcantes sumariados em Winge *et al.* (1984) e Longhi-Wagner *et al.* (1987).

Entre tais complexos, três foram reconsiderados no presente trabalho. O primeiro consiste do Complexo *Briza subaristata* Lam. – *B. subaristata* var. *interrupta* (Hack. ex Stuck.) Roseng., Arrill. & Izag.– *B. erecta* Lam.– *B. macrostachya* (J. Presl) Steud., tratado no Grupo 1 da análise de ISSRs (Cap. II) – Todas aceitas na sinonímia de *Chascolytrum subaristatum* (Lam.) Desv., no capítulo IV desta tese. Para esse complexo, os resultados obtidos pelos autores supracitados permitiriam separar com segurança apenas os dois

últimos táxons como elementos distintos. Entretanto, em nível macro-morfológico, as diferenças entre os mesmos e os demais elementos são discretas, e a ocorrência de diversos intermediários na natureza, com difícil categorização, levaram à necessidade de reconsideração dos limites entre os mesmos.

Rosengurtt *et al.* (1968) diferenciaram *Briza subaristata* var. *subaristata* de *B. subaristata* var. *interrupta* principalmente pelo maior tamanho das espiguetas da primeira. Esses autores, no mesmo artigo, referiram-se à *B. stricta* (Hook.) Steud. como intermediária entre *B. subaristata* e *B. macrostachya*. Os mesmos autores, no entanto, puseram em dúvida a aceitação de *B. stricta* e, em trabalho posterior (1970), não fizeram referência à mesma.

Matthei (1975) reuniu em sinonímia todos estes táxons inicialmente tratados por Rosengurtt *et al.* (1968) como distintos. Entretanto, Matthei (1975) incluiu *B. macrostachya* na sinonímia de *Chascolytrum erectum*, e *B. stricta* e *B. subaristata* var. *interrupta* na sinonímia de *C. subaristatum*. O referido autor apenas comentou que *C. subaristatum* constitui uma espécie muito variável.

Na análise por ISSRs, além dessas espécies, foi incluído um exemplar de cada uma das espécies apresentadas como novas no presente trabalho. O resultado foi a separação clara das espécies novas dos demais acessos. Não houve separação de acessos típicos de *Briza subaristata*, nem da variedade *interrupta*, nem de *B. macrostachya* e *B. erecta*.

A análise dos exemplares-tipo de *Briza subaristata*, *B. erecta* e *B. macrostachya* demonstrou que a primeira difere das demais basicamente pela cor e tamanho das espiguetas, enquanto as duas últimas são similares em todos os aspectos. A não-separação de grupos correspondentes aos limites destes táxons, somada à observação de intermediários na natureza, apoiou a inclusão de *B. erecta* e *B. macrostachya* na sinonímia de *B. subaristata* (leia-se *Chascolytrum subaristatum*).

Briza erecta vinha sendo tratada como espécie distinta em todas as floras do Uruguai, Argentina e Brasil, mas muito mais por tradição do que por efetividade dos limites taxonômicos. Na prática, costumeiramente eram classificadas em *B. erecta* plantas de espiguetas grandes, com poucos antécios, estes amarelados, apresentando rizomas finos e longos e lâminas foliares eretas, limites esses artificialmente estabelecidos. Todas as demais plantas que agrupavam muitas dessas características, mas não todas, ou eram identificadas como *B. macrostachya* ou como *B. subaristata*. Tais caracteres diagnósticos mostraram-se, no entanto, inadequados. A presença de rizomas longos e finos está associada, também em outras espécies do complexo, a solos arenosos. Tais rizomas já foram encontrados em *B. rufa* (= *Chascolytrum rufum*) e *B. bidentata* (= *Chascolytrum bidentatum*), por exemplo, quando crescendo neste tipo de solo. Já a cor das espiguetas atribuída à *B. erecta* também ocorre em plantas com espiguetas de tamanho aceito para *B. subaristata*, especialmente quando a mesma é coletada em dunas litorâneas. Não por acaso, a distribuição atribuída à *B. erecta* era litorânea, em especial em dunas do Uruguai, o que reforça a idéia de que tais caracteres, utilizados como diagnósticos por diferentes autores, são influenciados pelas condições ambientais.

O segundo complexo abordado neste trabalho incluiu as duas variedades aceitas para *Briza rufa* (= *Chascolytrum rufum*): *B. rufa* (J. Presl) Steud. var. *rufa* e *B. rufa* var. *sparsipilosa* Roseng., Arrill. & Izag. Pelo fato de o caráter diagnóstico da variedade *sparsipilosa* ser unicamente a presença de tricomas nos lemas, a mesma era pouco citada nas floras, ou era considerada em sinonímia. De fato, a presença de tricomas como único caráter diagnóstico desencoraja um pouco o pesquisador a aceitar esta variedade, especialmente se for considerado que parte da pilosidade pode se desprender da exsicata, com o passar dos anos.

Entretanto, análises de flavonóides (Hickenbick, 1983), de grãos de pólen (Leyser & Winge, 1979) e citológica (Shifino-Sampaio, 1979; Schifino & Winge, 1983) apóiam claramente o reconhecimento das duas variedades.

As seqüências de DNA das duas variedades apresentaram-se praticamente idênticas, de forma que foram tratadas como consenso nas matrizes para análise filogenética (Cap. I). Porém, na análise por ISSR, as duas variedades apresentaram-se em dois grupos distintos, apoiando a decisão de aceitá-las. A categorização em nível varietal parece bastante apropriada, pelo baixo grau de diferenciação morfológica. Além disto, o reconhecimento das duas variedades é importante para enfatizar que os dois grupos são geneticamente distintos.

O terceiro complexo tratado envolveu *Briza calotheca* (= *Chascolytrum calotheca*), *B. juergensii* (= *Chascolytrum juergensii*), *B. ambigua* (= *Chascolytrum ambiguum*) e *Briza* aff. *juergensii* (= *Chascolytrum juergensii* var. *angustilemma*). A última foi incluída unicamente na análise filogenética, e, apesar de compartilhar diversas características macromorfológicas com *B. juergensii*, apareceu sempre como um elemento distinto nas árvores obtidas, o que fortaleceu a idéia de considerá-la um novo táxon no nível varietal (Cap. III). A aceitação no nível varietal, e não no nível específico, deve-se ao fato de sua separação com relação à variedade típica estar baseada, em grande parte, na largura dos lemas. Diferenças na estatura da planta são bastante marcantes a campo, porém são difíceis de delimitar. Seu reconhecimento como nova variedade já havia sido sugerido com base em taxonomia numérica e em estudo de flavonóides (Hickenbick, 1983), tendo sido tratada como *Briza* aff. *juergensii* por Longhi-Wagner (1987).

As demais espécies envolvidas no terceiro complexo foram tanto estudadas utilizando ISSRs, quanto mediante inclusão na análise filogenética do grupo. No estudo envolvendo ISSRs, poucas conclusões puderam ser tiradas, pois o tratamento de todos esses

táxons simultaneamente gerou índices de similaridade muito baixos, até mesmo entre espécimes considerados pertencentes a uma mesma espécie.

Hickenbick (1983) já havia referido *Briza juergensii* como bastante complexa. No caso do Grupo 3 (Cap. II), possivelmente outro estudo com uma maior amostragem por espécie - porém analisando-as aos pares, e não num bloco único, como no presente trabalho -, possa contribuir para o melhor entendimento do mesmo.

No que diz respeito à circunscrição de *Briza monandra* (Hack.) Pilg. (= *Chascolytrum monandrum*), incluída tanto na análise por ISSRs quanto na análise filogenética, é interessante salientar que os indivíduos que ocorrem no sul do Brasil apresentam espiguetas menores e lemas bem mais estreitos do que aqueles que ocorrem em outros países. Apesar dessas diferenças, o acesso, representando a variante morfológica encontrada no nosso País agrupou-se com o acesso coletado na Bolívia, representante da variante morfológica extrabrasileira, tanto no estudo baseado em ISSRs (Grupo 4, Cap. II), quanto nas árvores baseadas em seqüências de cloroplasto (Cap. I). Todavia, o índice de similaridade entre os acessos foi baixo, e a diferença no comprimento dos ramos das árvores entre os mesmos foi grande. Essa diferenciação genética pode ser devida ao isolamento das populações brasileiras, que podem estar experimentando o efeito gargalo-de-garrafa (*bottle neck*). Exemplos de espiguetas pequenas e lemas estreitos também são encontrados em outros países, como na Bolívia (a título de ilustração, vide Figura 7 da Introdução). Por outro lado, os exemplares de espiguetas maiores não ocorrem no Brasil. Possivelmente, esta espécie já foi mais amplamente distribuída nas Américas, havendo uma posterior retração na distribuição das populações da mesma. O isolamento dessas populações deve estar levando à divergência morfológica que hoje é observada nos exemplares do sul do Brasil.

Como esse não é o único caso de distribuição disjunta no complexo (ver também mapas 9 e 12 do Cap. IV, para *C. brizoides* e *C. juergensii*), é possível que diversas outras espécies já tenham sido mais amplamente distribuídas, possivelmente em um clima pretérito mais ameno em boa parte das Américas.

A circunscrição em nível genérico

Embora pareça radical, à primeira vista, a decisão de agrupar todas as espécies sul-americanas em um único gênero, incluindo *Erianthecium bulbosum* (= *Chascolytrum bulbosum*) e *Briza brizoides* (= *Chascolytrum brizoides*), que são morfológicamente muito distintas das demais, a aceitação de espécies muito variáveis em um único gênero não é nenhuma novidade. O gênero *Briza* já foi tratado diversas vezes num sentido amplo, aceitando-se ou não seções ou subgêneros (Parodi, 1920; Rosengurtt et al., 1968, Longhi-Wagner, 1987). Como salientado por Rosengurtt et al. (1968), o polimorfismo de *Briza lato sensu* e a imprecisão de seus limites é desconcertante para um gênero tão pequeno.

Mesmo Matthei (1975), que separou as espécies usualmente incluídas no gênero *Briza lato sensu* em quatro gêneros distintos, admitiu as afinidades morfológicas entre as mesmas, denominando o grupo de “*Briza-Komplex*”.

As justificativas para a inclusão ou a exclusão de espécies do complexo foram as mais diversas. Nicora & Rúgolo de Agrasar (1981) excluíram *Briza bidentata* do complexo, transferindo-a para o gênero *Rhombolytrum*, por suas claras similaridades com *R. rhomboideum*. Na mesma obra, estas autoras comentaram, no entanto, que *Rhombolytrum* relaciona-se à *Briza* principalmente pela afinidade com *B. uniolae* (= *Chascolytrum uniolae*), possivelmente referindo-se às inflorescências contraídas de *R. rhomboideum* e *B.*

uniolae. As autoras também excluíram *B. ambigua*, transferindo-a para o gênero *Poa* L., pelos lemas muito estreitos. Porém, mantiveram no complexo, sob o gênero *Poidium*, *B. brasiliensis* (= *Chascolytrum brasiliense*) e *B. itatiaiae* (= *Chascolytrum itatiaiae*), que também apresentam lemas estreitos.

Nicora & Rúgolo de Agrasar (1981) aceitaram parte das espécies sul-americanas do complexo como pertencentes ao gênero *Briza*, agrupando-as com as espécies euro-asiáticas. As autoras consideraram que estas últimas são muito heterogêneas entre si para formar um gênero independente, como proposto por Matthei (1975), e se relacionam às espécies sul-americanas incluídas pelas autoras em *Briza*, pela compressão lateral dos lemas. Deste modo, Nicora & Rúgolo de Agrasar (1981) aceitaram os seguintes gêneros, para o Complexo: *Briza*, incluindo as espécies euro-asiáticas e parte das sul-americanas; *Poidium*, incluindo *P. itatiaiae* e *P. brasiliense*, e *Microbriza*, incluindo *M. brachychaete* e *M. poeamorpha*.

Clayton & Renvoize (1986) adotaram circunscrição semelhante a Nicora & Rúgolo de Agrasar (1981). Contudo, destacaram os lemas largos como caráter comum entre as euro-asiáticas e diversas espécies sul-americanas. Salientaram, também, a proximidade do gênero *Briza* com *Poa*, citando *Briza monandra* como intermediária, e com o gênero *Microbriza* Parodi ex Nicora & Rúgolo, citando que *B. rufa* pode apresentar lemas enrolados na base, na maturidade, como ocorre em *Microbriza poeamorpha* (J. Presl) Parodi ex Nicora & Rúgolo.

Rosengurtt & Arrillaga de Maffei (1979) descreveram o novo gênero *Lombardochloa*, baseando-se em *Briza rufa* (= *Chascolytrum rufum*), pela ocorrência peculiar de glândulas de óleo na base do lema, mas a separação deste gênero não foi aceita por autores posteriores. Longhi-Wagner (1987), entretanto, reconheceu tal característica

como de grande importância, propondo o reconhecimento da seção *Lombardochloa* dentro do gênero *Briza* tratado *lato sensu*.

Estudos de flavonóides (Williams & Murray, 1972), análise de proteínas de sementes (King, 1986), de fitoecdisteróides (Savchenko et al., 1998) e contagens cromossômicas (Murray, 1975, 1976a, 1976b; Matthei, 1975; Sampaio et al., 1979) apontaram para a distinção do grupo euro-asiático, mas não apresentaram qualquer separação entre as espécies sul-americanas. Observando-se a lista de sinônimos de *Chascolytrum subaristatum* (Cap. IV), por exemplo, verifica-se que esta espécie (incluindo sinônimos homotípicos e heterotípicos) já foi tratada como *Briza*, *Calotheca* e *Chascolytrum*, do mesmo modo que *C. uniolae* já foi tratada em *Briza*, *Chascolytrum* e *Poidium*, o que ilustra as enormes dúvidas quanto à circunscrição interna do grupo. O próprio gênero *Briza stricto sensu*, com apenas quatro espécies, reconhecido como gênero independente, seção ou subgênero por diferentes autores (subgênero *Eubriza*, Parodi, 1920; gênero *Briza*, Matthei, 1975; seção *Briza*, Longhi-Wagner, 1987) é morfologicamente diverso. Nem por isso, o agrupamento de suas espécies causa estranheza aos pesquisadores. *Briza maxima* já foi separada no gênero *Macrobriza* (Tzvelev) Tzvelev, e *Briza humilis* já foi transferida para um gênero monotípico, *Brizochloa* Jirásek & Chrtek.

Os resultados obtidos na análise filogenética do complexo indicam a separação do grupo euro-asiático no gênero *Briza stricto sensu*. Como as espécies sul-americanas não se agruparam conforme nenhuma circunscrição já proposta, e a resolução interna do agrupamento é baixa, a consequência é a proposição de uma nova circunscrição para o grupo. Naturalmente, esta nova circunscrição para o grupo sul-americano, tratado no Cap. IV como gênero *Chascolytrum lato sensu*, necessita de uma diagnose mediante a combinação de vários caracteres. A escolha deste nome deve-se ao fato de o mesmo ser mais antigo, e, portanto, prioritário em nível genérico. Igualmente adequado, seria aceitar o

gênero *Calothea*, por ter sido publicado na mesma obra. Entretanto, *Chascolytrum* foi o nome escolhido, pois é aquele que requer um número menor de combinações novas.

O fato do grupo sul-americano não apresentar resolução interna suficiente para ser desmembrado em outros gêneros menores ou mesmo seções ou subgêneros poderia suscitar a seguinte questão: acrescentando mais fragmentos de DNA a uma nova análise filogenética do grupo, seria possível o incremento da resolução dos grupos e, conseqüentemente, a separação de outras categorias?

É possível, mas pouco provável. Um conjunto de quase 3.000 pares de bases é considerado bastante satisfatório para as análises filogenéticas realizadas atualmente. Ainda que o número de caracteres informativos para parcimônia seja menor, ele ainda é muito superior ao número de caracteres utilizados, por exemplo, por Bayón (1998), em sua análise cladística. Também não se pode ignorar o fato de as análises probabilísticas considerarem as seqüências como um todo, ampliando o número de caracteres informativos para a construção da filogenia.

As politomias são comuns em análises com grande número de indivíduos, e nem sempre são resolvidas com a adição de mais fragmentos. Há grupos em que a especiação se deu de forma explosiva, com uma radiação adaptativa muito rápida, não necessariamente recente. Nestes casos, é natural que muitas seqüências de DNA sejam ainda conservadas em grupos morfologicamente distantes.

A hipótese de diversificação recente deve ser considerada com cautela, já que os padrões disjuntos de distribuição geográfica de algumas espécies sugerem que as mesmas foram mais amplamente distribuídas, e, portanto, sua diversificação pode não ter sido tão recente. Mas se pode perfeitamente considerar que a diversificação tenha sido rápida, de modo que adicionar fragmentos de DNA à análise não seria garantia de árvores com melhor resolução.

Como o monofiletismo do grupo sul-americano ficou evidente através da análise filogenética realizada, aceitá-lo como gênero único parece bastante adequado.

Naturalmente, ainda há diversas questões a serem respondidas. As principais dizem respeito às incongruências encontradas entre dados de seqüências nucleares e plastidiais, conforme discutido no Cap. I.

Os resultados mais significativos do conjunto deste trabalho são os seguintes:

- aceitação do grupo euro-asiático no gênero *Briza stricto sensu*, com quatro espécies;

- aceitação de um gênero sul-americano único, sem divisões infra-genéricas, com 22 espécies, destacando-se a inclusão de *B. bidentata* (= *Chascolytrum bidentatum*), *Rhombolytrum rhomboideum* (= *C. rhomboideum*), *Gymnachne koelerioides* (= *C. koelerioides*), *B. ambigua* (= *C. ambiguum*), *B. brasiliensis* (*C. brasiliense*) e *Erianthecium bulbosum* (= *C. bulbosum*);

- inclusão de *B. erecta* e *B. macrostachya* como sinônimos de *B. subaristata* (= *Chascolytrum subaristatum*);

- aceitação de *B. brasiliensis* (= *C. brasiliense*) e *B. itatiaiae* (= *C. itatiaiae*) como espécies distintas, baseando-se em morfologia, e o reconhecimento de sua estreita afinidade, baseado nos resultados da análise filogenética;

- descrição de três táxons novos para a ciência, baseando-se na análise morfológica respaldada por análises moleculares;

- a apresentação de uma monografia para o grupo, com chaves, descrições e ilustrações preparadas no intuito de auxiliar a identificação de espécies, seja qual for a região em questão.

Finalmente, é importante salientar que o trabalho aqui apresentado deixa margens a novos estudos com o grupo. Uma análise mais profunda dos padrões de distribuição

geográfica deve ser iniciada em breve, e um estudo de anatomia da lâmina foliar e de micromorfologia de lema e pálea encontra-se em andamento em nosso Programa de Pós-Graduação. O estudo evolutivo do grupo deverá incluir, no futuro, uma análise mais acurada das incongruências entre filogenias, possíveis eventos de hibridação, e o estudo de sua evolução numa escala temporal.

REFERÊNCIAS BIBLIOGRÁFICAS

- Bayón, N.D. 1998. Cladistic analysis of the *Briza* Complex (Poaceae, Poeae). *Cladistics* 14, 287 - 296.
- Clayton, W.D., Renvoize, S.A. 1986. *Genera graminum: grasses of the world*. Royal Botanic Gardens, London.
- Hickenbick, M.C.M. 1983. *Relações Fenéticas e Cladísticas entre as Espécies do Complexo Briza (Gramineae): Flavonóides*. Tese de Doutorado. Curso de Pós-Graduação em Genética. Universidade Federal do Rio Grande do Sul. Porto Alegre.
- King, G.J. 1986. A taxometric analysis of seed proteins in the genus *Briza* s.l. (Poaceae). *Pl. Syst. Evol.* 151, 163 - 174.
- Leyser, V.E., Winge, H. 1979. Análise palinológica de espécies do complexo *Briza* (Gramineae). *Ciência e Cultura* 31, 577.
- Longhi-Wagner, H.M. 1987. *Flora Ilustrada do Rio Grande do Sul, Fasc. 17. Gramineae. Tribo Poeae*. B. Inst. Bioc. 41, 1 - 191.
- Longhi-Wagner, H.M., Winge, H., Hickenbick, M.C.M., Schifino, M.T., Torres de Lemos, C., Fonseca, I.A., Freitas-Sacchet, A.M.O., Passaglia, L.M.P. 1987. Sistemática e origem das espécies neotropicais de *Briza* L. (Gramineae). In: *Anales del IV Congreso Latinoamericano de Botánica, III. Simposio: Citología y citotaxonomia*. Bogotá.
- Matthei, O. 1975. Der *Briza*-Komplex in Südamerika: *Briza*, *Calotheca*, *Chascolytrum*, *Poidium* (Gramineae). *Willdenowia* 8, 7 - 168.
- Murray, B.G. 1975. The cytology of the genus *Briza* L. (Gramineae). I. Chromosome numbers, karyotypes and nuclear DNA variation. *Chromosoma (Berl.)* 49, 299 - 308.

- Murray, B.G. 1976a. The cytology of the genus *Briza* L. (Gramineae). II. Chiasma frequency, polyploidy and interchange heterozygosity. *Chromosoma* (Berl.) 57, 81 - 93.
- Murray, B.G. 1976b. The cytology of the genus *Briza* L. (Gramineae). III. B Chromosomes. *Chromosoma* (Berl.) 59, 73 - 81.
- Nicora, E.G., Rúgolo de Agrasar, Z.E. 1981. Los géneros sudamericanos afines a *Briza* L. (Gramineae). *Darwiniana* 23(1), 279 - 309.
- Parodi, L.R. 1920. Notas sobre las especies de *Briza* de la Flora Argentina. *Rev. Fac. Arg. Agr. y Vet. B.* Aires 3, 113 - 137.
- Rosengurt, B., Arrillaga de Maffei, B., Izaguirre de Artucio, P. 1968. Sinopsis de *Briza* (Gramineae) del Uruguay y notas taxonomicas sobre otras especies de este genero. *Bol. Fac. Agr. Montevideo* 105, 1 - 35.
- Rosengurt, B., Arrillaga de Maffei, B., Izaguirre de Artucio, P. 1970. Gramíneas Uruguaias. Universidad de la Republica, Departamento de Publicaciones. Montevideo.
- Rosengurt, B., Arrillaga de Maffei, B. 1979. *Lombardochloa*, nuevo genero de Gramineae. *Anales Fac. Quím.* 9, 255 - 288.
- Sampaio, M.T.S., Hickenbick, M.C.M., Winge, H. 1979. Chromosome numbers and meiotic behavior of South American species of the *Briza* complex (Gramineae). *Rev. Bras. Genet.* 2(2), 125 - 134.
- Savchenko, T., Whiting, P., Šik, V., Underwood, E., Sarker, S.D., Dinan, L. 1998. Distribution and identities of phytoecdysteroids in the genus *Briza* (Gramineae). *Biochem. Syst. & Ecol.* 26, 781 - 791.
- Schifino-Sampaio, M. T. 1979. Citotaxonomia do complexo *Briza* (Gramineae): número cromossômico, cariótipo, quantidade de DNA nuclear, comportamento meiótico. Dissertação de Mestrado. Curso de Pós-Graduação em Genética. Universidade Federal do Rio Grande do Sul. Porto Alegre.

- Schifino, M.T., Winge, H. 1983. Karyotypes and nuclear DNA content of species of the *Briza* complex and some other genera of Poeae (Gramineae). Rev. Bras. Genet. (Brazil. J. Genetics) 4(2), 245 - 259.
- Torres de Lemos, C. 1983. Relações fenéticas entre as espécies do complexo *Briza* (Gramineae): isoesterases. Dissertação de Mestrado. Curso de Pós-Graduação em Genética, Universidade Federal do Rio Grande do Sul, Porto Alegre.
- Williams, C.A., Murray, B.G. 1972. Flavonoid variation in the genus *Briza*. Phytochemistry 11, 2507 - 2512.
- Winge, H., Hickenbick, M.C.M, Longhi-Wagner, H.M., Torres de Lemos, C., Schifino, M.T., Fonseca-Born, I., Moure, J.S., Leyser da Rosa, V. 1984. Sistemática e evolução das espécies sul-americanas do Complexo *Briza* (Gramineae). In: Colóquio Sobre Citogenética e Evolução de Plantas, 1. Sociedade Brasileira de Genética, Piracicaba, pp. 185 - 203.

Anexos

1. Alinhamentos

1a. Alinhamento dos fragmentos nucleares (ITS + GBSSI)

CLUSTAL X (1.81) multiple sequence alignment

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AFFJU      TCGTGACCCTTAAACAAAATAGACCGTGAANGTGTNATCC--ATCCGCATGGGCTTCTN-
AFFLA      TNGTGACCCTG-ACCAAAACAGACCGCGCACGCGTTATCC--AGCATGCCGG-CGGCGGC
AFMON      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTTATCC--AGCCTGCCGG-CGGCGGC
AMPHI      TCGTGACCCTG-ACCAAAANAGACCGCGCACGCGTCATCC--ATCCTGCCGG-CGGCGAC
ASALA      NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNGG-CGGCAC
BIDEN      TCGTGACCCTKAAMCAAAACAGACCGYGAACRYGTCATCC-AWGYCTGCYSG-CGSYSSG
BRACH      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTCATCT--AGCCTGCCGG-CGGCGGC
BRASI      TCGTGACCCTTAAACAAAACAGACCGTGAATTTGTCA-CC--TA--TGTCGC-TGG----
BROMU      TCGTGACCCTG-ACCAAAACAACACCGCGCACGCGTCATCC-AATCCGTCGATGATG-GGC
CALO2      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTTATCC--AGCCTGCCGG-CGGCGGC
CALOT      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTTATCC--AGCCTGCCGG-CGGCGGC
CBRIZ      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTCATCC--AGCCTGCCGG-CGGCGGC
ERECT      NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNGCCTGCCGG-CGGCGGC
ERIAN      TCGTGACCCTTAAANCANAACAGNCANCAACATGTCATCCCATGCCNGTCGG-CGT-GGC
GYMNA      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTCATCC--TGCCTGCCNG-CNGCG--
ITATI      TCGTGACCCTTAAACAAAACAGNCCGTGAATTTGTCA-CN--TA--TGTCGC-TGG----
JUERG      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTGTATCC--AGCCTGCCGG-CGGCGGC
LAMAR      ACGTGACCCTG-ACCAANN-ATACCGTGAACGTGTTATCT--ACCCTGCCGG-CGGCGGC
LAMSC      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTTATCC--AGCCTGYCGG-CGRCGGC
MACRO      TCGTGACCCTG-ACCAAAACAGACCGYGCACGCGTTATCC--AGCCTGCCGG-CGGCGGC
MAXIM      TCGTGACCCTG-ACCAAAACAGACCGTGCACGCGTTATCT--AATCTGCCGAGCTGCCGC
MEDIA      NNNNNNCCCTG-ACCANNNNNNACCGCGCACGAGTCATCC--ATCCTGCCGG-CGGCGGC
MINOR      TCGTGACCCTG-ACCAAAACAGATTGTGCACGCGTTATCC--ATCCCCTGG-TGACGGC
MONAN      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTCATCC--AGCCTGCCGG-CGGCGGC
PALEA      TCGTGACCCTT-AACAAANNNNNNNNNNNNNNNNNNNNNNTTATCC--AGCCTGNNNN-CGGCGGC
PAROD      TCGTGACCCTG-ACCAAAACMGACCGCGCACGYGTTATCC--AGCCTGTCCG-CGGCGGC
POAAN      TCGTGACCCTG-ACCAAAATAGACCGTGAACCGTCATCT--AGCCCGCCAG-CAGCAG-
POABR      TCGTGACCCTG-ACCAAGACAGACCGTGAACGCGTCATCT--TGCCTGCCAG-CGGCGG-
POAEM      TCGTGACCCTTAAACAAAATAGACCGTGAACGTCATCC--A--TGCCG--CGTGGGC
POOID      NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN
RUFU      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTCATCC--AGCCTGCCGG-CGGCGGC
SCABR      NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNTTATCC--AGCCTGCCGGCGCGCGGC
SUBAR      NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNG-C
UNIOL      TCGTGACCCTG-ACCAAAACAGACCGCGCACGCGTTATCC--AGCCTGCCGG-CGGCGGC

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AFFJU      -CCTGTGC-----AAGG-CCCCGA--CCTTCCTTTCT--GAGGG-AGAGG
AFFLA      NCC-GTTCGTGCTC-GGC-CAA-G-TCCTCNACAACCTCCNNTCCTNGGAGNN--GGGG
AFMON      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
AMPHI      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
ASALA      ---GTTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GAGG
BIDEN      SACCCTTTCGTGCTC-GGM-CAAGGCTCCYCGACAACYTCTYTCCTCGGAGTG-AGRGG
BRACH      ACC-GTTCGTGCTC-GGT-CAA-G-TCCTCGACAACCTCCTCTCCTGGGAGAG--GGGG
BRASI      ACC-----TCGGTCCGGC-TTTGG-CCCCGA--CCTTCG-TCTT-GGAGGG--GAGG
BROMU      ATC-GTCCATGCTC-GGC-CAT---CCTCGGTCACCTACACTCCTCGGAGTG-GGTTG
CALO2      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
CALOT      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
CBRIZ      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
ERECT      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
ERIAN      ACC-GTTCCTTAGGTC-GGC-CAA-G-TCC-CGACA-CCTCCTCGTNTCGGAGGGGAGTG
GYMNA      ACT-GTTCGTGCTC-GGC-CAA-G-TNNTCGCCANCCTCCTCTCCTCGGANNNT-GGGGG
ITATI      ACC-----TCGGTCCGGC-TTTGG-CCCCGA--CCTTCG-TCTT-GGAGGG--GAGG
JUERG      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
LAMAR      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
LAMSC      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GRGG
MACRO      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
MAXIM      ACC-GTCCGTAGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGCG--GGGG
MEDIA      ACC-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
MINOR      ATC-GTCCGTGCTC-GGC-CAA-G-TCCTTGAGAACCTCCTCCTTTGGAGTT--GGTA
MONAN      ACA-GTTCGTGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG

```

PALEA ACC-GTTCGTC-CTC-GGC-CAAAG-TCCTCGACAACCTCCTCTTCTCGGAGTG--GGGG
PAROD ACC-GTTCGTCGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
POAAN -CCATGCCGTTGCTT-GGC-CAAAT-TCCTCGAGAACCTCCCCTCCTTGGAGTG--GTGG
POABR -CCCTGCCGTCGCTT-GGC-CAAAG-TCCTCGATAGCCTCCTCTCCTCGGAGCG--GGGG
POAEM -----TTCTGC-CTG-TG--CAAGG-CCCCGA---CCTTCTTTCT---GAGGG-AGAGG
POOID NNNCTCNNGCGNGNGGGGGGNN
RUF A ACC-GTTCGTCGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
SCABR ACC-GTTCGTCGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG
SUBAR NNNNNNNNNNNNNNNNNNGCCNNNNNNCCCTGATCACCTMMTCTYYT-GGAGTG-AGRGG
UNIOL ACC-GTTCGTCGCTC-GGC-CAA-G-TCCTCGACAACCTCCTCTCCTCGGAGTG--GGGG

*

AFFJU G-GCCGC--AAAAGAACCCTACGGNGCCGAAGNGTCAAGGAACACTTGATATTGCC-TT
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AFMON CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TA
AMPHI CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TA
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BRACH CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TTA
BRASI G-GCCAC--AACAGAACC-ACGGCGCATAGGCGTCAAGGAACACT-GATATTGCC-TT
BROMU CTCGGGG-TAAAAGAACC-ACGGCGCCGAAGGCGTCAAGGAACACT-G----TGTC-TA
CALO2 CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TA
CALOT CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TA
CBRIZ CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TA
ERECT CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TA
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GYMNA CTCGGGG-TAAAAGAACC-ACGACGCCTANGNNNNNNNNNGAACANT-G----TGNC-TA
ITATI G-GCCAC--AACAGAACC-ACGGCGCCANNNNCGTCAAGGAACACT-GATATTGCC-TT
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MAXIM CTCGGGG-TAAAAGAACC-ACGGCGCCGAAGGCGTCAAGGAACACT-G----TGCC-TA
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POAEM G-GCCGC--AAAAGAACC-ACGGCGCCGAAGGCGTCAAGGAACACT-GATATTGCC-TT
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RUF A CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TA
SCABR CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TA
SUBAR S-GCCTC--AAMAGAACY-ACGRGCGCNAAGGCGTCAAGGAACACT-GNTNTTGCC-TW
UNIOL CTCGGGG-TAAAAGAACC-ACGACGCCTAAGGCGTCAAGGAACACT-G----TGCC-TA

** *

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AFMON GTCC-GGGGACGCGGACGGCTTG-CTGGCCG-CTCCCCNNNNNNNNNNNNNNNNNNNNNN
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BROMU ACCC-GAGGGCATGGCTAGCTTG-CTGGTCA-TCTCTGTGTGCAATGCTAT-TTAATC
CALO2 GTCC-GGGGACGCGGACGGCTTG-CTGGCCG-CCCCCGTGCCTGCAATGCTAT-TTAATC
CALOT GTCC-GGGGACGCGGACGGCTTG-CTGGCCG-CCCCCGTGCCTGCAATGCTAT-TTAATC
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ERECT GTCCGGGGGAAGCGGACGGCTTG-CTGGCCG-CCCCCGTGCCTGCAATGCTAT-TTAATC
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MEDIA GTCC-GGGGACGCGGACGGCTTGCTTGCCG-CCCCCGTGCCTGCAATGCTAT-TTAATC
MINOR GCAA-GGGGT-GCGGACAGCTTG-CTGGCTG-CACCTCGTGCCTGCAATGCTAT-TTAATC

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RUF A GTTC-GGGGACGCGGACGGCTTG-CTGGCCG-CCCCCGTGCTGCAATGCTAT-TTAATC
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SUBAR GCGA-GGGGWYCGGWYRGCTTG-CTGGCCG-MCCCYTKWGCWGCRAATGMTATGTYAATC
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*

AFFJU NNN
AFFLA NNN
AFMON NNN
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AFMON NNN
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UNIOL GCCCGAGGCCATTTCGGCCGAGGGCAGCGCTGCCTGGGCGTCACGC-AAAACACGCTCCCA

AFFJU --ACCCATCCTTGGGGA--N-GGACGTGGTGTGTTGACTCTTCTCGTGNATGGNGC--GG
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SUBAR MAACCCCTCATRKGKRA-GMRGGACGYGGRYRKTGGCCCCCGTCGSSAAGGGSM T--GG
UNIOL C-ACCC-TCATCGGGGA-GCGGGACGCGGCATGTGGCCCCCGTCGCGCAAGGGGC--GG
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AFFJU NNGGNCNAAGTT-GGGGCTGCT-GGCA--TAAC-TTGCCNGCACCCGACCGTGGNNGGGCG
AFFLA NNGGCCNAANATAT-GGNTG-CCGGCG--TNTC-NTGCCGANNNNNCGCGNNGGNGCN
AFMON TGGGCCGAAGATAT-GGCTG-CCGGCG--TATC-GTGCCGGACACAGCGCGTGGTGGGCG
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ERECT TGGGCCGAAGATAT-GGCTG-CCGGCG--TATC-GCGCCGGACACAGCGCGTGGTGGGCG
ERIAN TGGGCCGAANATAT-GGCTGCT-GGCG--TATC-GNGCCGGACACAGCGCGTGGTGGGCG
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MONAN TGGGCCGAAGATAT-GGCTG-CCGGCG--TATC-GTGCCGGACACAGCGCGTGGTGGGCG
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SUBAR TGGGCCGAAGWTGKGBTGSYCGGYG--TATC-RTRCCRKRACMGRCRGTGGTGGGCG
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* * **

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 CBRIZ NNN
 ERECT GAAGTTGAGCGGATGCTCAAGAGCGCTGAGGAGAAGTTCCTCAGACAAGGTGAGAGCCGT
 ERIAN GAAGTTTGAAGCGGATGCTCAAGAGCGCTGAGGAGAAGTTCCTCAGACAAGGTGAGAGCCGT
 GYMNA NNN

ERIAN -ATC-TTAGACGAAATCCATGGCCA----TGATCAAAATTTCTTCCTAAATTTTCAGCCC
GYMNA NNN
ITATI NNN
JUERG NNN
LAMAR NNN
LAMSC -ATC-TTAGACGAAATCCATGGCCA----TGATCAAAATTTCTTCCTAAATTTTCAGCCC
MACRO -ATC-TTAGACGAAATCCATGGCCA----TGATCAAAATTTCTTCCTAAATTTTCAGCCC
MAXIM -ATCCTGATACG-ATTCATGGCCATGCATGACCGAAACTTCTTGCT-AATTC-CAGCCC
MEDIA -ATC-TTAGACGAAATCCATGGCCA----TGATCAAAATTTCTTCCTAAATTTTCAGCCC
MINOR GATCCTGTAACGAAATCCATGGCCA----CGACCGATATTTCTTCCTCAATTT-CAGCCA
MONAN --TC-TTAGAC-AAATCCATGGCC-----TGA-CAAAATTTCTTCCTAAATTTT-AAACC
PALEA -ATC-TTAGACGAAATCCATGGCCA----TGATCAAAATTTCTTCCTAAATTTTCAGCCC
PAROD NNN
POAN NNN
POABR NNN
POAEM NNN
POOID NNN
RUFFA NNN
SCABR -ATC-TTAGACGAAATCCATGGCCA----TGATCAAAATTTCTTCCTAAATTTTCAGCCC
SUBAR NNN
UNIOL NNN

AFFJU NNN
AFFLA NNN
AFMON TGTCG- GTGCGCG--TCCACC-GGCGGGCTCGTCGACACGATCGTGG-AAGGCAAGACTG
AMPHI TGNGC- GTGCGCG--TCCACC-GGCGGACTCGTCGACACGATCGTGG-AGGGCAAGACTG
ASALA NNN
BIDEN TGCGC- GTGCGCG--TCCACC-GGCGGGCTCGTAGACACGATCGTGG-AGGGCAAGACTG
BRACH TGCGC- GTGCGCG--TCCACC-GGCGGGCTCGTCGACACGATCGTGG-AGGGCAAGACTG
BRASI NNN
BROMU TGCGC- GTGCGCG--TCCACC-GGAGGGCTCGTCGACACCGTCTGTTGG-AGGGCAGGACCG
CALO2 NNN
CALOT TGTCG- GTGCGCG--TCCACC-GGCGGGCTCGTCGACACGATCGTGG-AGGGCAAGACTG
CBRIZ NNN
ERECT TGCGC- GTGCGCG--TCCACC-GGCGGGCTNGTCGACACGATCGTGG-AGGGCAAGACTG
ERIAN TGCGC- GTGCGCG--TCCACC-GGCGGGCTCGTCGACACGATCGTGG-AGGGCAAGACTG
GYMNA NNN
ITATI NNN
JUERG TGTCGCGTGC GGCGTTCCACC-GGCGGGCTNGTNGACACGATCGTGGGAGGGCAAGANTG
LAMAR NNN
LAMSC TGCGC- GTGCGCG--TCCACC-GGCGGGCTCGTCGACACGATCGTGG-AGGGCAAGACTG
MACRO TGCGC- GTGCGCG--TCCACC-GGCGGGCTCGTCGACACGATCGTGG-AGGGCAAGACTG
MAXIM TGCGC- GTGCGCG--TCCACC-GGCGGGCTCGTCGATACGATCGTGG-AGGGCAAGACTG
MEDIA TGCGC- GTGCGCG--TCCACC-GGCGGGCTCGTAGACACGATCGTGG-AGGGCAAGACTG
MINOR TGCGG- GTGCGCG--TCCACC-GGCGGACTCGTCGACACGATCGTGG-AGGGCAAGACTG
MONAN TGCGC- GTGGG-G--TCCACC-GGCGGGTT-GTCGAC-CAATC-TGG-AGG-CAAGACTG
PALEA TGCGG- GTGCGCG--TCCACC-GGCGGGCTCGTCGACACGATCGTGG-AGGGCAAGACTG
PAROD NNN
POAN NNN
POABR NNN
POAEM NNN
POOID NNN
RUFFA NNN
SCABR TGCGC- GTGCGCG--TCCACC-GGCGGGCTCGTCGACACGATCATGG-AGGGCAAGACTG
SUBAR TGCGG- GTGCGCG--TCCACC-GGCGGGCTCGTGNNNNNNN-ATCGTGG-AGGGCAAGACTG
UNIOL NNNNNNNNNNNNGCG--TCCACC-GGCGGGCTCGTCGACACGATCGTGG-AGGGCAAGACTG

AFFJU NNN
AFFLA NNN
AFMON GATTCACATGGCCGCTCAGCGTCGACGATAGGCCCT----CGTTCTCTTGTCTAAAT
AMPHI GATTCACATGGCCGCTCAGCGT- GACGTAGGCTCA----TTGATCTCTTGT-TAAAT
ASALA NNT
BIDEN GATTCACATGGCCGCTCAGCGTCGACGATAGGCCCT----CGTTCTCTTGTCTAAAT
BRACH GATTCACATGGCCGCTCAGCGTCGACGATAGGCCCT----CGTTCTCTTGTCTAAAT
BRASI NNN
BROMU GGTTCACATGGCCGCTCAGCGTCGACGATAGGCAGCTCACCAGTCTATTGTGTACAT
CALO2 NNN
CALOT GATTCACATGGCCGCTCAGCGTCGACGATAGGCCCT----CGTTCTCTTGTCTAAAT
CBRIZ NNN

ERECT GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
ERIAN GATTCCACATGGGCCGT-TCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
GYMNA NNN
ITATI NNN
JUERG GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
LAMAR NNN
LAMSC GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
MACRO GATTCCACATGGGCCGTCTCAGCGTCGACGTANNNNNNNNNNNNNNNNNNNNNNNNNNAAT
MAXIM GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
MEDIA GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
MINOR GATTCCACATGGGCCGT-TC-GCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
MONAN GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
PALEA GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
PAROD NNN
POAN NNN
POABR GATTCCACATGGGCCGCTTAGCGTCGACGTAAGCTT-----CGTG-TCTTGTCTGAAC
POAEM NNN
POOID NNN
RUF A NNN
SCABR GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
SUBAR GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT
UNIOL GATTCCACATGGGCCGTCTCAGCGTCGACGTAGGCCCT-----CGTTCTCTTGTCTAAAT

AFFJU NNN
AFFLA NNN
AFMON CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
AMPHI C-ATTGATCT-GTCGATCCTT---GCAGCTCATATCAA--TGATTCGTTTT---
ASALA CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
BIDEN CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
BRACH CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
BRASI NNN
BROMU TCATCCATCTTGTCCATCATGGCAGCTCTGACGGATCATGAAA-TGATTCCTTTT---
CALO2 NNN
CALOT CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
CBRIZ NNN
ERECT CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCACC AAC-TGATTCGTTTT---
ERIAN CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
GYMNA NNN
ITATI NNN
JUERG CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
LAMAR NNN
LAMSC CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
MACRO CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
MAXIM NCATTGATGTTNNNGTTTT---
MEDIA CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
MINOR CCATTGATGTCGTCGATCCTTGCAGCCAGCTCGGATCATCAGC-TGATTCGTTTT---
MONAN CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
PALEA CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
PAROD NNN
POAN NNN
POABR ---TGATCTATC-ATCAATATTGACGCTCAGATCATCAAATGATTGGATCTATAA
POAEM NNN
POOID NNN
RUF A CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
SCABR CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
SUBAR CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---
UNIOL CCATTGATGTCGTTGATCCTT---GCAGCTCGGATCATCAAC-TGATTCGTTTT---

AFFJU NNN
AFFLA NNN
AFMON -----CTTGGTGGCCA-----GTGCGACGTGGTGGAGCCTGCCGACGTGAAGA
AMPHI -----TTCTGTGACCTGCATGACGTCGAGTGCACGTCGTTGGAGCCTGCAGACGTGAAGA
ASALA -----CTTGGTGGCCA-----GTGCGACGTGGTGGAGCCTGCCGACGTGAAGA
BIDEN -----CTTGGTGGCCA-----GTGCGACGTGGTGGAGCCTGCCGACGTGAAGA
BRACH -----CTTGGTGGCCA-----GTGCGACGTGGTGGAGCCTGCCGACGTGAAGA
BRASI NNN
BROMU ---TTCTTGTGGTGGCCA-----GTGCAACGTGGTGGAGCCTGCCGACGTGAAGA
CALO2 NNN
CALOT -----CTTGGTGGCCA-----GTGCGACGTGGTGGAGCCTGCCGACGTGAAGA

CALOT A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 CBRIZ NNN
 ERECT A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 ERIAN A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 GYMNA NNN
 ITATI NNN
 JUERG A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 LAMAR ASATGGTCAAGAWCWCATGGCCCTGGATCTGTCTCKGAMGKKAYGGRWTCRAGMTAGCA
 LAMSC A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 MACRO A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 MAXIM A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACTCTAAGA-----
 MEDIA A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 MINOR A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCG
 MONAN A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 PALEA A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 PAROD NNN
 POAN NNN
 POABR A--TGGTCAAGAAGTGCATTGGTCCAGGATCTCTCCTGGAAGGTACGTAACCAAGCTAGCA
 POAEM NNN
 POOID A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 RUF A--NGGTCAAGANCTGCANGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 SCABR A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 SUBAR A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA
 UNIO L A--TGGTCAAGAAGTGCATGGCCAGGATCTCTCCTGGAAGGTACGTAATCAAGCTAGCA

AFFJU ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 AFFLA NNN
 AFMON ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 AMPHI ACACCTT-CACTGCACAACCTAAGAGTAAAGCTCTAGC---TAAATT-----
 ASALA ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 BIDEN ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 BRACH ACACCTT-CACAGCACAACTAAGAGTAAATTGCAGC---TAAATTATAGGCTAGTGCCTA
 BRASI NNN
 BROMU ATAGTG-GTTTGCAGAAC-AA---TTTAAAG-----ACTAT-GGCT-----
 CALO2 NNN
 CALOT ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 CBRIZ NNN
 ERECT ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 ERIAN ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 GYMNA NNN
 ITATI NNN
 JUERG ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 LAMAR ATASYW-CCCAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 LAMSC ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 MACRO ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 MAXIM -----GTAAGTGTAGCTAGCTAAACTATTTGTTACCGCCTA
 MEDIA ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 MINOR ATA--T-CACCGCACAACTAAGTGTAACTGCAGC---TAAATC-----
 MONAN ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 PALEA ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 PAROD ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 POAN NNN
 POABR GCACCTTTTATAGCACAACTAGCAGTAAACTGTAGC---TGAATTAATGGCTAGTTCGTA
 POAEM AACTA-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATYATAGGCTAGAGCGTA
 POOID ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 RUF NNN
 SCABR ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 SUBAR ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA
 UNIO L ACACCTT-CACAGCACAACTAAGAGTAAATTGTAGC---TAAATTATAGGCTAGTGCCTA

AFFJU MGATGCATT---GCTC---GTGTTGGTGATCAAATC---GATCAANNNNNNNNNNNNNN
 AFFLA NNN
 AFMON CGATGCATT---GCTC---GTGTTGGTGATCAGATC---GATCAATGGTGCT-TT-GT
 AMPHI -----CA-ATC---GATCAATGGTGCT-TT-GC
 ASALA CGATGCATT---GCTC---GTGTTGGTGATCAGATC---GATCAATGGTGCT-NN-GC
 BIDEN CGATGCATT---GCTC---GTGTTGGTGATCAGATC---GANNNNNNNNNNNNNNNNNNN
 BRACH MGATGCATT---GCTC---GTGTTGGTGATCAGATC---GATCAACGGTNNNNNNNNNN
 BRASI NNN
 BROMU -----CAA-----TGGTGGT-TTGCG-----TGTGCTGTATGC

1b. Alinhamento dos fragmentos plastidial (*trnL-trnF-trnF*)

CLUSTAL W(1.60) multiple sequence alignment

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AFFJU      GAA-CCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
AFFLA      NNNNNNNNNNNNNNNNNNNCTTCAA-TTCAGAGNNNNCTGGAATTA AAAAAGGGCAAT
AFMON      NNNNNCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
AMPHI      GAAACCTGCTA-GTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
ASALA      GAAACNNNGNTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
BIDEN      GAAACCTACTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
BRACH      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
BRASI      GAAACCTNNTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
BROMU      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
CALO2      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
CALOT      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
CBRIZ      GAAACCTGNNNNGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
ERECT      NNNNNNNNNNNNGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
ERIAN      GAAACCTNCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
GYMNA      NNNNNNNNNNTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
ITATI      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
JUERG      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
LAMAR      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
LAMSC      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
MACRO      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
MAXIM      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
MEDIA      GAAACCTGCTA-GTGGTA-CTTCCAAATTCAGAGAAA-CCTGGAATTA AAAAAGGGCAAT
MINOR      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
MONAN      GA-CCCTGCTA-GTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA WKAAGGGCAAT
PALEA      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
PAROD      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
POAAN      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
POABR      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
POAEM      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
POOID      GG-CCCTG-TGAGTGGTTCCTKCCMAWTTTCAGAGAAACCTGGAATTA WMMWAAGGGCAAT
RUF      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
SCABR      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
SPNOV      NNNACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
SUBAR      NNNNNNNNNNTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
UNIOL      GAAACCTGCTAAGTGGTAACTTCCAAATTCAGAGAAACCTGGAATTA AAAAAGGGCAAT
```

```
AFFJU      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
AFFLA      CCTGAGCCAAATCCG----TGTTTTGAGAAAAGCAGGGGTTCTCGAATCGAACTAGAAAT
AFMON      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
AMPHI      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAA-----CTAGAAAT
ASALA      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
BIDEN      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
BRACH      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
BRASI      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
BROMU      CCTGAGCCAAATCCC----TTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
CALO2      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
CALOT      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
CBRIZ      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
ERECT      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
ERIAN      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
GYMNA      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
ITATI      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
JUERG      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
LAMAR      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
LAMSC      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
MACRO      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
MAXIM      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAA-----CTAGAAAT
MEDIA      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAA-----CTAGAAAT
MINOR      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAA-----CTAGAAAT
MONAN      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCRAACTAGAAAT
PALEA      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
PAROD      CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAAT
POAAN      CCTGAGCCAAATCCA----TGTTTTGAGAAAACAAGGGGTTCTCGAA-----CTAGAAAT
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POABR CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAA-----CTAGAAT
POAEM CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAT
POOID CCTGRGCCWAATCCG----TGTTTTGAGAAAACWAWGGGGTTCTCGAATCKRACKTAKRRT
RUF A CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAT
SCABR CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAT
SPNOV CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAT
SUBAR CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAT
UNIOL CCTGAGCCAAATCCG----TGTTTTGAGAAAACAAGGGGTTCTCGAATCGAACTAGAAT

AFFJU ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
AFFLA ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
AFMON ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
AMPHI ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
ASALA ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
BIDEN ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
BRACH ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
BRASI ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
BROMU CCAAATGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAGGTTA
CALO2 ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
CALOT ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
CBRIZ ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
ERECT ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
ERIAN ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
GYMNA ACNNGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
ITATI ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
JUERG ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
LAMAR ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
LAMSC ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
MACRO ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
MAXIM ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
MEDIA ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
MINOR ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
MONAN ACRRRGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
PALEA ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
PAROD ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
POAAN ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
POABR ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
POAEM ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
POOID ASRRRGGAWMMGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACRAATCRRG--KTA
RUF A ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
SCABR ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
SPNOV ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
SUBAR ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA
UNIOL ACAAAGGAAAAGGATAGGTGCAGAGACTCAATGGAAGCTGTTCTAACGAATCGAG--TTA

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AFMON ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAGGGGCTTTATACATC
AMPHI ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
ASALA ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAGGGGCTTTATACATC
BIDEN ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
BRACH ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAGGGGCTTTATACATC
BRASI ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAGGGGCTTTATACATC
BROMU ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAGGGGCTTTATACATC
CALO2 ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAGGGGCTTTATACATC
CALOT ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAGGGGCTTTATACATC
CBRIZ ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
ERECT ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
ERIAN ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
GYMNA ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
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JUERG ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAGGGGCTTTATACATC
LAMAR ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
LAMSC ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
MACRO ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
MAXIM ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
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MINOR ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC
MONAN ATTACGTTGTGTGTTAGTGGAACCTCTTCTAAATTTGAGAAAGAAGGGGCTTTATACATC

PALEA ATTACGTTGTGTGTTAGTGGAACTCCTTCTAAATTTGAGAAAGAGGGGCTTTATACATC
PAROD ATTACGTTGTGTGTTAGTGGAACTCCTTCTAAATTTGAGAAAGAAGGGCTTTATACATC
POAAN ATTACGTTGTGTGTTAGTATAAATTCCTTCTAAATTCGAAAAAGAAGGGCTTTATACATC
POABR ATTACGTTGTGTGTTAGTGGAAATTCCTTCGAAATTCGAAAAAGAAGGGCTTTATACAGC
POAEM ATTACGTTGTGTGTTAGTGGAACTCCTTCTAAATTTGAGAAAGAAGGGCTTTATACATC
POOID WTTACGKKGTGYGYTMSKGGAACTCCTTCTAAATTTGABAWAKAMGGGSWTWATAYMTC
RUF A ATTACGTTGTGTGTTAGTGGAACTCCTTCTAAATTTGAGAAAGAAGGGCTTTATACATC
SCABR ATTACGTTGTGTGTTAGTGGAACTCCTTCTAAATTTGAGAAAGAAGGGCTTTATACATC
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UNIO L ATTACGTTGTGTGTTAGTGGAACTCCTTCTAAATTTGAGAAAGAAGGGCTTTATACATC

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CALO2 TAATAT----AGGTTCTTTATT-CTTTTTTAG-----
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POOLD NNN
RUF TAATAT----AGGTTCTTTATT-CTTTTTTAG-----
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UNIOL TAATAT----AGGTTCTTTATT-CTTTTTTAG-----

AFFJU -----
AFFLA -----
AFMON -----
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BIDEN -----
BRACH -----
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CALOT -----
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ERECT -----
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GYMNA -----
ITATI -----
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MINOR -----
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RUF -----
SCABR -----
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UNIOL -----

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POOLD NNN
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UNIOL CTAGTAAAAGGAAAATCCGTCGACTTTATAAGTCGTGAGGGTTCAAGTCCC-TCTATCCC

AFFJU NNN
AFFLA CANNN
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BRASI TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
BROMU TCACAAAGGAATGCGAANAGA AACTCAATGG-ATCTTATCC-----TAGAA-TAGATTC
CALO2 TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
CALOT TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
CBRIZ TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
ERECT TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
ERIAN TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
GYMNA TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
ITATI TCCCAAAGGAGGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
JUERG TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
LAMAR TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
LAMSC TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
MACRO TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
MAXIM TCTCAAAGGAGTGGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
MEDIA TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TATATTT-
MINOR TACCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TATATTT-
MONAN TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTTA
PALEA TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
PAROD TCCAAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
POAAN TCACAAAGGAGTGCGAAGAGA AACTCAATGG-ATCTTATCC-----TAGAA-TATATTT-
POABR TCACAAAGGAGTGCGAAGAGA AACTCAATGG-ATCTTATCC-----TAGAA-TATATTT-
POAEM TCCAAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
POOID NNN
RUF A TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
SCABR TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
SPNOV TCCCAAAGGAGTGCGAAGAGA AATAATGG-ATCTTATCC-----TAGAA-TAGATTC
SUBAR TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC
UNIOL TCCCAAAGGAGTGCGAAGAGA AACTAAATGG-ATCTTATCC-----TAGAA-TAGATTC

AFFJU TTTTT-ATTGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
AFFLA TTTTT-ATTGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
AFMON TTTTT-ATTGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
AMPHI TTTTT-ATTGAGTATCGGGAAGG-----AATCCCGG-TTATTCATTCTAT-TTTTA
ASALA TTTTTTATTGAGTATCGGGAAGG-----AATCCCGGTTNTTACTCTAT-TTTTA
BIDEN TTTTT-ATTGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
BRACH TTTTT-ATTGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
BRASI TTTTT-ATTGAGTATCGGGAAGG-----AATCNGG-TATTCACTCTAT-TTTTA
BROMU TTTTT-ATTAGATATCGGCAAAAATCTTGTTATGG-TTATTCACTCTAT-TTTTA

CALO2 TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
CALOT TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
CBRLZ TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
ERECT TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
ERIAN TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
GYMNA TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
ITATI TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
JUERG TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
LAMAR TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
LAMSC TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
MACRO TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
MAXIM TTTTTG-ATTCGAGTATCGGGAAGGGAAGG--AATCCCGG-TTATTCATTCTAT-TTTTA
MEDIA TTTTTT-ATTCGAGTATCGGGAAGGGAAGG--AATCCCGG-TTATTCACTCTAT-TTTTA
MINOR TTTTTT-ATTTGAGTATTTGGGAAGGGAAGG--AATCCCGG-TTATTCACTCTAT-TTTTA
MONAN TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
PALEA TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
PAROD TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
POAAN TTTATT-AT---AGTATCGGGAAGG-----AATCCCGG-TTATTCATCTATTTTTTTC
POABR TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCATCTCTTTTTTTC
POAEM TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
POOID NNN
RUF A TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
SCABR TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
SPNOV TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTTACTCTAT-TTTTA
SUBAR TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA
UNIOL TTTTTT-ATTCGAGTATCGGGAAGG-----AATCCCGG-TTATTCACTCTAT-TTTTA

AFFJU AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
AFFLA AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
AFMON AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
AMPHI AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
ASALA ANNATTATT-AATTAAGCCNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN
BIDEN AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
BRACH AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
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BROMU AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
CALO2 AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
CALOT AGTATTATT-AMGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
CBRLZ AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
ERECT AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
ERIAN AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
GYMNA AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
ITATI AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
JUERG AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
LAMAR AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
LAMSC AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
MACRO AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
MAXIM AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
MEDIA AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
MINOR AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
MONAN AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
PALEA AGTATTATT-ACGTAAGCCATATACACTGCGTAGGACTACCCCC--ATTTTCAAATTT
PAROD AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
POAAN AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
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POAEM AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
POOID NNA
RUF A AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
SCABR AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
SPNOV AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
SUBAR AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT
UNIOL AGTATTATT-AAGTAAGCCATATACAATGCGTAGGACTACCCCC--ATTTTCAAATTT

AFFJU CGAATTTGAAATACTTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
AFFLA CGAATTTGAAATACTTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATANATACAA
AFMON CGAATTTGAAATACTTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
AMPHI CGAATTTGAAATACTTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
ASALA NNN
BIDEN AGAATTTGAAATACTTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA

BRACH CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
BRASI C-AATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATA-ATACAA
BROMU TGAATTTGAAATACTTTT- ---AATTGATTTTTTAGTCCCTTAATTGACATANATACAA
CALO2 CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
CALOT CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
CBRIZ CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
ERECT CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
ERIAN CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
GYMNA CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
ITATI CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
JUERG CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
LAMAR CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
LAMSC CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACANTANATACNA
MACRO MGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
MAXIM CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACAGAGATACAA
MEDIA CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
MINOR CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
MONAN CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
PALEA AGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
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POABR AGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
POAEM CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATARATACAA
POOID CGAATWYSMAAATASTTTAT-TYAAWWSATRRWTTAGTCCWMAAATGMCATAGATACAA
RUFU CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
SCABR CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
SPNOV CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
SUBAR CGAATTTGAAATACTTTAT-TTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA
UNIOL CGAATTTGAAATACTTTATCTTAATTGATTTTTTAGTCCCTTAATTGACATAGATACAA

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AFFLA ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCA-TTGGTAGAGCAGAGG
AFMON ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
AMPHI ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
ASALA NNN
BIDEN ATCCTCTACTAGGATGATGCAGAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
BRACH ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
BRASI ATCCTCTACTAG-ATGATGCA--AAAAAGGTCAGGATAGCTCAGTTGGNAGAGCAGAGG
BROMU ATCCTCTACTAGGATGATGCACAANAAGAAAAGGTCAGGATAGCTCAGTTGGTAGANAGG
CALO2 ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
CALOT ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
CBRIZ ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
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ERIAN ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
GYMNA ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
ITATI ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
JUERG ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
LAMAR ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
LAMSC ATCCNCTACTAGGANGANGCACAANAAGGNCAGGATAGCTCANTTGGNAGANCANANG
MACRO ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
MAXIM ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
MEDIA ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGC
MINOR ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
MONAN ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
PALEA ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGAAGGTAGAGCAGAGG
PAROD ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
POAAN ATACTCTACTAGGATTATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
POABR ATACTCTACTAGGATTATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
POAEM ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
POOID ATMSTSTAYYWGGAWGATGSRGARGAAMAGGTCAGGAGRGCTCAGNNNNNNNNNNNNNN
RUFU ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
SCABR ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
SPNOV ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
SUBAR ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGGATAGCTCAGTTGGTAGAGCAGAGG
UNIOL ATCCTCTACTAGGATGATGCACAAGAAAAGGTCAGKAYAGCTCAGKTGGTAGAGCAGAGG

AFFJU ACTGAAAATCCTCGTGT
AFFLA ACTGAAAATCCTCGTGT
AFMON ACTGAAAATCCTCGTGT

AMPHI	ACTGAAAATCCTCGTGT
ASALA	NNNNNNNNNNNNNNNN
BIDEN	ACTGAAAATCCTCGTGT
BRACH	ACTGAAAATCCTCGTGT
BRASI	ACTGAAAATCCTCGTGT
BROMU	ACTGAAAATCCTCGTGC
CALO2	ACTGAAAATCCTCGTGT
CALOT	ACTGAAAATCCTCGTGT
CBRIZ	ACTGAAAATCCTCGTGT
ERECT	ACTGAAAATCCTCGTGT
ERIAN	ACTGAAAANNNNNNNNN
GYMNA	ACTGAAAATCCTCGTGT
ITATI	ACTGAAAATCCTCGTGT
JUERG	ACTGAAAATCCTCGTGT
LAMAR	ACTGAAAATCCTCGTGT
LAMSC	ACTGAAAATCCTCNTGN
MACRO	ACTGAAAATCCTCGTGT
MAXIM	ACTGAAAANNNNNNNNN
MEDIA	NNNNNNNNNNNNNNNN
MINOR	ACTGAAAATCCTCGTGT
MONAN	ACTGAAAATCCTCGTGT
PALEA	NNNNNNNNNNNNNNNN
PAROD	ACTGAAAATCCTCGTGT
POAAN	ACTGAAAATCCTCGTGT
POABR	ACTGAAAATCCTCGTGT
POAEM	ACTGAAAATCCTCGTGT
POOID	NNNNNNNNNNNNNNNN
RUFA	ACTGAAAATCCTCGTGT
SCABR	ACTGAAAATCCTCGTGT
SPNOV	NNNNNNNNNNNNNNNN
SUBAR	ACTGAAAATCCTCGTGT
UNIOL	ACTGAAAATCCTCGTGT

2. Matrizes para análise dos ISSRs (Ref. Cap. II)

2a. Matriz ISSR Complexo 1

1 114 21L 1 9

li7 li8 hw5029 hw5040 li10 li11 li12 li19 li202 li54 li57 li13 li18 hw5038 hw5056 sc li201 z8119 z8173 a359 w10768

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2b. Matriz ISSR Complexo 2

1 78 7L 1 9
li280L li281L li278L li279L hw8061 li372L li373L
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0011000
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0100000
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1100100
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1100101
1110111
0110000
1001011
0110100
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0100101
0100100
1011001
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0010000
0001001
1111111
0101111
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0001000
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0010100
1111101
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0011111
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0100100
0100000
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1119191
1119090
1111119

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2.c. Matriz ISSR Complexo 3

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1 76 15L 1 9
hw8062 li115 li308 hw5041 li192 hw9662 hw8749 hw8713 li222 li186 li132 li242 lfl200 hw5019 li190
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2.d. Matriz ISSR Complexo 4

1 94 7L 1 9
poo li55 li106 li317D li175 mona BS
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0001000
0000010

1000010
1111101
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1111110
0010100
1000000
0001100
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1911119
