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The true identity of *Stylosanthes seabrana* B.L. Maass & L. 't Mannetje (Leguminosae Papilionoideae)

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Abstract — Samples of the legume, *Stylosanthes seabrana* B.L. Maass & L. 't Mannetje, were collected in Bahia, Brazil and have been described taxonomically. They have been compared with *S. scabra* Vogel and *S. hamata* Taubert. Fruits and plants of *S. seabrana* cv. Unica was studied, the seed were germinated and root samples were studied used for chromosome counts. All herbarium specimens examined corresponded with *S. scabra* Vogel. Root studies of *S. seabrana* cv. Unica indicated that both diploid and tetraploid cytotypes of *S. scabra* were present, and we conclude that it is a synonym of *S. scabra* Vogel.

Key words: cytology, Leguminosae, Southamerica. *Stylosanthes*, taxonomy.

INTRODUCTION

Stylosanthes scabra is widely distributed in South America, occurring in Colombia, Ecuador, Venezuela, Peru, Brazil and the north-eastern, eastern and southern regions of Bolivia, Mohlenbrock (1957). It also occurs in the dry Chaco and eastern regions of Paraguay, Vanni & Fernandez, in this paper, and Burkart (1939) reported the species from Argentina, where it was found exclusively in the north-west.

Stylosanthes seabrana was first recognised as a distinct taxon in the 1980s by CSIRO's Plant Introduction group at the Davies Laboratory, Townsville, Australia and by CIAT's Genetic Resources group in Colombia. In both cases, the species was considered as being intermediate in morphology and growth between *Stylosanthes hamata* Taubert and *Stylosanthes scabra* Vogel. Maass (1989) referred to this 'new' species as 'cf. scabra-type' and Jansen and Edye (1996) as *S.* sp.aff. *S. scabra*. Edye and Maass (1997) reported on the three species *S. scabra*, *S. hamata* and *S. seabrana* although the latter still had not been formally published. Maass and 't Mannetje (2002) finally described the species *Stylosanthes seabrana* B.L. Maass & 't Mannetje, indicating that it was related to *Stylosanthes scabra*. They presented a key for separating the new species from both *S. scabra* and *S. hamata* Taubert. In addition, they reported different levels of ploidy in *S. scabra*, 2n = 40 chromosomes and *S. seabrana*, 2n = 20 chromosomes. According to a previously published analysis, *S. scabra* likewise recorded sa having 2n = 40 chromosomes (Cameron 1967). Two cultivars "Primar" and "Unica" were subsequently selected from a series of accessions of *Stylosanthes seabrana* in Australia and CIAT, Colombia and were released in Australia.

In an endeavour to clarify the relationship between *S. seabrana* and other *Stylosanthes* species, we examined material from *Stylosanthes* accessions collected in South America and Australia.

MATERIALS AND METHODS

The herbaria reviewed were those of the New York Botanical Garden, (NY), the Royal Botanical Garden Kew (K); Museum National d'Histoire Naturelle (P); the Nationale Plantentuin van België (BR), the Botanischer Garten und

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Botanisches Museum of Berlin-Dahlen (B); and the Botanische Staammlung, München (M). We also had access to a significant amount of South American material such as samples of *S. hamata* TAUBERT, (1890) author of the most important monograph of *Stylosanthes*. Another important contributions is MOHLENBROCK (1957). The following paratypes of *S. seabrana* were studied: Coradín et al. 1172 (K), Coradín et al. 5186 (K), Coradín. LC 5208 (K), Coradín et al. 5221 (K), Coradín et al. 1234 (K), Bastos 75 (K).

A seed sample of *Stylosanthes seabrana* cv. Unica (commercially obtained from Heritage Seeds, Australia) was sent to us by Dr Albrecht Glatzle, INTTAS, Loma Plata, Paraguay. We compared the plant material grown from this seed with the herbarium specimens of will source *S. seabrana* from Bahia.

All taxonomically relevant characteristics of these samples have been compared with the following specimens of the *Stylosanthes scabra – S. seabrana* – complex, covering all the material available in the most prestigious herbariums in South America and worldwide.

The synonymy was based on the following finding:

Stylosanthes scabra Vogel

Vogel, Linnaea 12: 69. 1838. TYPE: "Hbt. in Brasil merid.: *Sellow*. in Serra da Maeda" (Phototype 2248 F!). Brasilia, Bahia, *Sellow* 332 (isosyntype B!) Brasilia, *Sellow* s.n. (isosyntype K!). Brasil: Bahia, *Luschnath* 149 (isosyntype BR!), *Stylosanthes seabrana* B.L. Mass & L. 't Mannetje, Novon 12(4): 497-500. 2002. TYPE: Brasil, Bahia Rodovia Irecê-Xique-Xique (BA-052) 80 km. to, 10º56'S, 42º29'W, 29 June 1983, L. Coradin, G.P. Silva, J.G.A. Vieira, F.B. de Souza, R.M. Harley, S. Linintong & R. Baker 6261 (holotype CEN, isotype K!, M!)

Iconography: Burkart, 1939: 239, fig. 14, a-g; lam. 13; Mohlenbrock, 1957: 301, fig. 1, 6.

Branched erect perennial herb to 50 cm tall., ramified, branches with an indumentum of weak, white hair, of variable density and setae of up to 2 mm long. Leaves trifoliolate. Stipules united to the leaf base free at their linear apex, where they are, glabrous with setae. Petioles 4-6 mm long., with whitish hair. Leaflets 8,5-15 mm long. x 2-5 mm wide, elliptic, leaflet lower surface gland-dotted, with marked, white or green veins more undoubtedly the rest of the sheet, with setae on the veins and the margins, upper surface of leaflets glabrous. Inflorescence in dense spike, always developed in the axil of a leaf, up to 5-flowered, each flower protected by a bract, this similar to the leaves, but smaller, each flower with two bracteoles for and a feathery estipite, extrafloral. Flowers yellow, 5,5 mm long., hypantium 5 mm long. Calyx campanulate, 2 mm long., tube 1 mm long., 5 teeth, the two upper teeth rounded upper, united to each other, the lower three theth larger and ciliate, 1 mm long. Standard, 5 mm long., rounded, emarginate; the wings oblong, keel slightly curved inwards. Fruit with 2 articles, each 11 mm long. x 2,5 mm wide., with dense, weak, white hairs and distinct eins, the upper article with a 2 mm long beack., this curved at the apex.

Distribution. Colombia, Ecuador, Venezuela, Peru, Brazil, Bolivia. BURKART (1939), recorded the species for Argentina. Not known in Paraguay.

Inhabits fields, and hillsides on sandi soil in 420-1900 m s.m.

Specimens examined

VENEZUELA. In savanna Nagrea, 1891, Warming 101 (C).

ECUADOR. Loja, 5 Km NE of La Loma, 29-IV-2001, J.E. Madsen et al. 7789 (K).

PERU. **Tumbes.** Tumbes, La Cruz, 28-V-1992, Sagástegui 14615 (NY).

BRASIL. Amazonas Glaziou 9733 (C). **Bahia**. Bahia, in orilloris aridi, 1830, Salzman s.n. (P). Lagoa da Eugenia, near Camaleao, Harley et al 16248 (P). Jacobina, 3-IV-1967, Krapovickas 12823 (CTES). Mun. de Lancois, Rio Lancois, 10-IV-1992, Hatschbach et al. 56954 (CTES). Jaguaquara, estrada BR. 4, 30-III-1967, Krapovickas 12777 (CTES). Chapada occidental da Bahia, islets and branchs of rio Corriente, by Correntina, 580 m s. m., 44º38'W 13º20'S, 23-IV-1980, Harley et al. 21622 (NY). **Distrito Federa**l:Brasil inter, Martius 1124 (P). Brasilia. Warming 3168 (C). BR. 20, entrada a Formosa, 2-II-1990, Arbo et al. 3441 (CTES). Estrada entre Taguatinga-Brazlandia, 15º46'S, 48º4'W., 27-IV-1983, Kirkbride 1640 (CTES). **Rio de Janeiro**: Rio de Janeiro, 1867, Glaziou 1375 (BR). **Sao Paulo**. Mun Campinas, beira rodovia D. Pedro, 1 km. ao trevo p/ Magi-Mirin, 20-II-1976, H. Freitas Leitao Filho 1787 (NY). Campos das sete Lagoas, 3,7 Km NNW of Padua Sales, 22º18'S 47º10'W, cerrado, 21-III-1961, Eiten et al. 2742 (NY). **Minas Gerais**: Chapada Praevium Martius 757 (M). 1838, Clausen 905 (P). Gouveia, rib. do Tigre 13-XI-1971, Hatschbach

56954 (NY). Prope Caza Blanca, 18-X-1924, Riedel 735 (P). **Mato Grosso do Sul**: Alrededores de Campo Grande, camino a Sidrolandia, 10-II-1979, Krapovickas et al. 34531 (CTES).

BOLIVIA. **Santa Cruz**: Gutierrez, orillas del río Piray, 8 km. E de Portachuelo, 20-IV-1977, Krapovickas et al. 31561 (CTES). Chiquitos, 2 km. W de San José, km 263, del ferrocarril, 60º47'W, 17,47'S, 25-IV-1980, Krapovickas et al. 36531 (CTES). Ñuflo de Chávez, 3 Km S de Ascención de Guarayos, 63º6'W, 15º43'S, Ayo. San Joaquín, 27-IV-1977, Krapovickas et al. 31801 (CTES). Andrés Ibañez, ciudad de Sta. Cruz, 420 m s.m., 12-VI-1989, Coimbra 548 (NY). **Tarija**: Santa Cruz, ruta Tarija Villa Montes, Entre Ríos, 21-V-1971, Krapovickas et al. 19087 (CTES). **Chuquisaca**: Tomina, Los Naranjos, 1900 m s.m., 12-XII-1992, Saravia Toledo 11114 (CTES).

PARAGUAY. **Amambay**: 22º45'S, 56º15'W, ruta 3 y ruta 5, 23-VIII-1980, Schinini et al. 20524 (CTES). Cerro Corá, 17-VIII-1980, Schinini et al. 20284 (CTES). Ruta 3, 35 km SE de Bella Vista, 24-VIII-1980, Schinini et al. 20568 (CTES). **Canindeyú**: Reserva biológica Mbaracayú, 7-X-1991, Caballero Mármori 2091 (CTES). **Alto Paraguay**: Pto. Casado, 16-V-1974, Arenas 729 (CTES). **Chaco**: Cerro León, lomada al E de la meseta central, 17-V-1988, Charpin. & Ramella AC 21661 (CTES). Cap. Pablo Lagerenza, 20º20'S 60º20'W, 14-IV-1989, Ramella 2170 (CTES). **Pte. Hayes**. Est. Exp. Chaco Central M.A.G.G.T.Z. Col de leguminosas introducidas CSIRO-110361, indentif. *S. aff scabra*, 13-IV-1997, Vanni & Lopez 3988 (CTES). Boquerón, Filadelfia, Isla Poí. "Unica", componente 2 de la mezcla, 27-I-2006, Glatzle s.n. (CTES).

ARGENTINA. **Jujuy:** Dep. Ledesma: Ledesma, 11-III-1905, Spegazzini (Min. Agr. 13943) (SI). **Salta**: Dep. Capital, lomadas 1º ayo. ruta Salta a San Lorenzo, 21-III-1979, J.Fernández s.n. (CTES). 3 km de Salta, camino a San Lorenzo, La Loma, 10-IV-1980, Krapovickas et al. 35971 (CTES). Lomas del Mecidor, 1200 m s.m., II-1925, Inostroza s n (LIL).

For chromosome analysis, the seeds were scarified in hot water at 75ºC for 10 minutes. After germination, roots of the seedlings were treated in 8-oxiquinoleína for 3 hours and fixed in acid lactic absolute-alcohol for 24 hours (Fernandez, A. 1973), and stored in 70% alcohol The roots-tips were hydrolyzed for 8 min in 1N HCL at 60º and were stained in Feulgen (FERNANDEZ *et al.* 1991). The roots were macerated and subsequently modified in a lacto-acetic acid of orceína. The material was examined under an optical microscope in the metaphase.

RESULTS

All materials examined from the herbariums corresponded with *S. scabra* Vogel. No taxonomically relevant distinctions could be detected between the specimens studied. All characteristics of all materials examined fully agreed with the standard description for *S. scabra*:

The study of roots from seeds germinated in the laboratory showed that cultivar Unica was made up of diploid and tetraploid cytotypes of *S. scabra* (Figures 1 A, 1B).

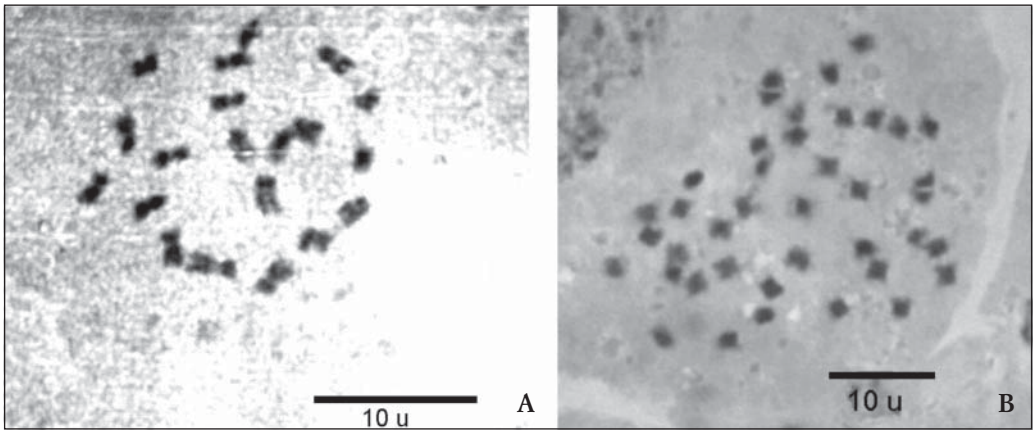

Fig. 1 — Somatic chromosomes of *S. scabra:* A. 2n= 20; B. 2n= 40.

DISCUSSION

From this study, it was not possible to separate the examined *S. seabrana* specimens from *S. scabra* Vogel.

While it is a general agreed *S. hamata* can be distinguish from the *S. scabra* – *S. seabrana* – complex by fruit morphology, we disagree with EDYE and MAASS (1997) and DATE *et al.* (2010) who consider the form of leaflets, the absence or presence of bristles and hairs on stipules and leaflets, and their venation as distinctive characteristics that separate the two species. Typically in taxonomy these morphological traits are not considered sufficient to separate species (MAASS et al. 2002). Likewise ploidy levels are not valid criteria for species distinction in the genus *Stylosanthes*, as *S. scabra* has been reported to be one of the few species with diploid (2n = 20) and tetraploid (2n = 40) genotypes CAMERON (1967). This is in agreement with our findings within a single commercial seed sample, declared as *S. seabrana* Unica (Figure 1). *Scabra* at CSIRO Lansdown Research Station in north Queensland, Australia, including 15 accessions of *S. seabrana* used by JANSEN and EDYE (1996), 18 additional *S. seabrana* and 11 *S. scabra* accessions collected by L.A. Edye and R.A. Date and 30 accessions of *S. scabra* from CIAT (Maass aff.-scabra-type), 2 *S. scabra* cultivars (Seca and Siran) and *S. hamata* cv. Amiga. On the basis of morphological and agronomic characteristics, he classified them into 5 groups. Group 1 contained 35 members, of which 33 were listed as *S. seabrana* and 2 as *S. scabra*; Group 2 contained 25 members, of which 23 were listed as *S. scabra*, Group 4 had 14 members, including the 2 *S. scabra* cultivars (Seca and Siran). All members of this group were *S. scabra* and most had provenances of Minas Gerais in Brazil. Group 3 had 2 members and Group 5 had only 1. Sixty-six of the new accessions originated from the state of Bahia in Brazil, while the remaining 10 were from the neighbouring state of Minas Gerais. Most of those in Group 1 were from the northern half of the region and those in Group 2 were from the southern half.

DATE *et al.* (2010) studied accessions almost exclusively from Brazil and particularly Bahia. No information is given whether or not the paratypes of *S. scabra* and *S. seabrana* had been included in their study. Leaflet and stipule characteristics, such as shape, presence and absence of bristles had been used for species separation, which we do not consider sufficient caracters. Nor the trend to *Bradyrhizobium* specificity between distinguishable groups of ecotypes of the *S. scabra* – *S. seabrana* – complex, observed by Date et al. (2010) do justify the distinction of two different species.

Having analyzed materials form the various (sub)tropical regions of the American continent, (Vanni, 2008 inéd. Vanni, 2009) we consider having covered most of the natural variability of the *S. scabra* – *S. seabrana* – complex which is not appropriate to separate two different species.

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