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Source: Systematic Botany, 40(3):746-754.

Published By: The American Society of Plant Taxonomists

URL: <http://www.bioone.org/doi/full/10.1600/036364415X689203>

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Paspalum minutispiculatum (Poaceae, Panicoideae): a New Annual Species from Central Brazil

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Communicating Editor: Benjamin van Ee

Abstract—*Paspalum* (Poaceae, Panicoideae) is a large genus comprising approximately 330 species, of which 210 are cited for Brazil. *Paspalum minutispiculatum* is similar to other related species of the informal “Humboldtiana” group, but this new species is annual and lacks rhizomes. This work establishes the distinction between this new species and related ones from this group, and contributes to a better characterization of the little known *P. bicilium*. Micromorphological characters of the upper anthecium and anatomical characters of the root, culm, and leaf of both species were analyzed. The new species appears to be endemic to the Federal District. It grows along trails in gallery forest, which is a rare habitat for this genus. The present work includes a key for species of the “Humboldtiana” group.

Keywords—*Ceresia*, Gramineae, Humboldtiana, leaf anatomy, Paspaleae.

Paspalum L. (Poaceae, Panicoideae) is predominantly an American genus, with approximately 330 species, with a few taxa growing in Asia and Africa (Zuloaga and Morrone 2005). Oliveira and Valls (2014) cited 210 species from Brazil, 75 of these exclusive to the country. Major centers of species diversity include central and southern Brazil, eastern Paraguay, north-eastern Argentina, Mexico, and the Andean region (Chase 1929). The genus can be recognized by the unilateral raceme-like partial inflorescences and plano-convex spikelets with the upper lemma on the adaxial side of the rachis, lower glumes frequently lacking, obtuse, indurate upper florets, and lemma margins involute at maturity, although one or more of these characters may be absent in some species (Chase 1929).

Chase (1929) accepted the subdivision of *Paspalum* into the subgenera *P. subg. Paspalum* and *P. subg. Ceresia* (Pers.) Rchb. She characterized the latter by its foliaceous rachis, with one to several racemes per inflorescence, and spikelets densely pilose, with long white hairs, mainly along the margins of the upper glume and lower lemma. Denham et al. (2002) reviewed the taxonomy and undertook phylogenetic studies in *P. subg. Ceresia* using morphological characters. Only homoplasious characters support *P. subg. Ceresia*: a membranous upper anthecium, upper lemma not entirely enclosing the palea, but with a gap at the apex, and a winged rachis (Denham et al. 2002). The new species described here agrees with all the above characters.

Denham et al. (2002) accepted 25 species in *P. subg. Ceresia*, distributed from Mexico to Argentina, Uruguay, and southern Brazil. Since the work of Denham et al. (2002), several new species and the re-establishment of others related to *P. subg. Ceresia* were published, all occurring in the Brazilian Cerrado vegetation. Oliveira and Rua (2005) described *P. vallsii* R. C. Oliveira & G. H. Rua. The species *P. vexillarium* G. H. Rua, Valls, Graciano-Ribeiro & R. C. Oliveira was published, and at the same time *P. spissum* Swallen. was reestablished by Rua et al. (2008). Ramos et al. (2011) described *P. rostratum* D. Ramos, Valls & R. C. Oliveira.

Brazil hosts the largest number of species of *Paspalum* subgenus *Ceresia*, 15 of which are endemic. Of these, 12 species are endemic to the Brazilian Cerrado. Rua et al. (2010) showed that *P. subg. Ceresia* is paraphyletic, and that *P. humboldtianum*

Flüggé, *P. polyphyllum* Nees ex Trin., and *P. bicilium* Mez, in *P. subg. Ceresia*, form a clade with *P. paucifolium* Swallen and *P. falcatum* Nees ex Steud. The morphological relationship between *P. humboldtianum*, *P. polyphyllum*, and *P. paucifolium* has already been shown by Parodi and Nicora (unpubl. manuscript), who included all these species in the informal group “Humboldtiana.” Previously, Scataglini et al. (2014) found this clade to be composed of five species: *P. polyphyllum*, *P. buchtienii*, *P. heterotrichon*, *P. humboldtianum*, and *P. ceresia*. These species coincide with clade B of Denham et al. (2002), with the exception of *P. ceresia*.

All these species are rhizomatous and have a pilose upper glume with ciliate margins. The upper glume of this new species is similar to that of other species in this clade, and is morphologically more closely related to the little known *P. bicilium*, on account of the smaller size of the spikelets and of the two longer cilia on the upper glume. Killeen (1990), Denham et al. (2002), and Zuloaga and Morrone (2005) considered *P. bicilium* as conspecific with *P. polyphyllum*. Filgueiras (1991), Peñaloza et al. (2008), Silva (2013), and Oliveira and Valls (2014) considered *P. bicilium* and *P. polyphyllum* independent species. Rua et al. (2010) found evidence with molecular data that these species are distinct.

Field collections made in the vicinity of the “Poço Azul” waterfall, near Brazlândia, Federal District, Brazil revealed a new species of *Paspalum*, which appears to be endemic to this area. This new species is herein described, illustrated, and compared macromorphologically with *P. bicilium* and *P. polyphyllum*. Characters of the upper anthecium observed with scanning electron microscopy, and of the anatomy of roots, culms and leaves of *P. minutispiculatum* and *P. bicilium* are provided. A key for closely related species discussed in this study is also included.

MATERIALS AND METHODS

The new species was collected along trails in gallery forest at the “Poço Azul” area of the Brasília National Park, Brazil (Fig. 1). Descriptions and illustrations were based on field populations and herbarium material (UB). For the traditional taxonomic study, the specimens were analyzed and measured using a stereoscopic microscope (Leica EZ4D).

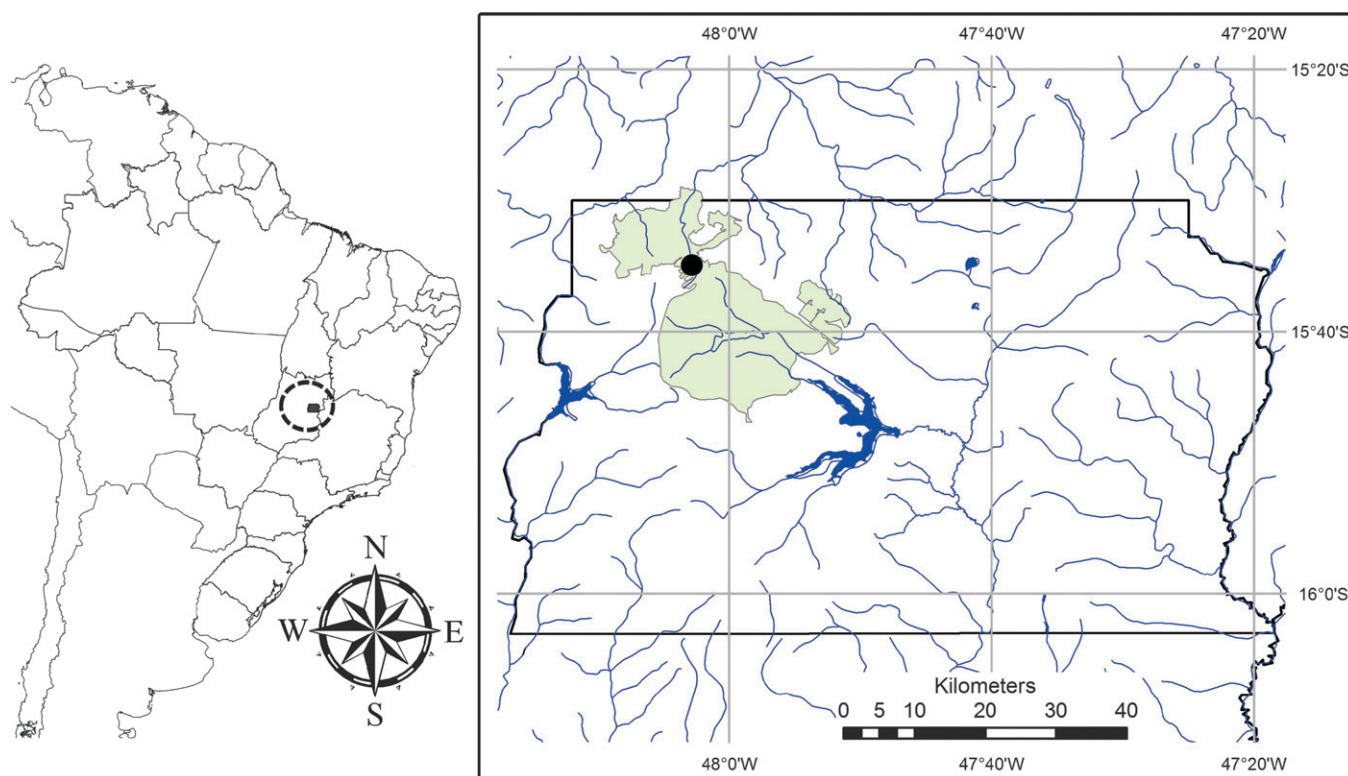


FIG. 1. Map showing the limits of Brasília National Park with the location of the population of the new species, *Paspalum minutispiculatum*.

Field data on *P. bicilium* were based on Silva (2013, unpubl. data) and personal observations. Anatomical data on *P. bicilium* were based on Silva (2011, unpubl. data). The voucher used for the micromorphological study of this species was Oliveira *et al.* 2612 (UB).

For the anatomical study, the root, culm (internode), and leaf lamina were rehydrated by boiling in distilled water (voucher Oliveira *et al.* 2612). Transverse sections of the middle third of the culm (internode) and of the leaf lamina, and the root at ca. 5 cm from the tip, were made on a table microtome (Reichert-Jung model 26896, Heidelberg, Germany). The sections were clarified in 50% sodium hypochlorite for 1–3 hrs, washed in distilled water, stained with alcian blue and safranin (4:1), and dehydrated in an ethanol and butyl acetate gradient (Johansen 1940). Paradermal sections were prepared in 30% Franklin solution (acetic acid and hydrogen peroxide), washed, dehydrated, and stained with alcian blue. Permanent slides were prepared with Acrilex® crystal colorless varnish (Paiva *et al.* 2006). The anatomical descriptions followed the style of the taxonomic ones (without verbs) and the terminology was based on Türpe (1967) and Ellis (1976, 1979).

The upper antheria of both species were analyzed with scanning electron microscopy (SEM) without previous treatment. The samples were mounted directly onto aluminum specimen stubs with double-sided carbon tape and then sputter coated with gold. Micrographs of the abaxial surface of the lemma and palea were obtained using a scanning electron microscope (model JEOL JSM 7001F).

TAXONOMIC TREATMENT

Paspalum minutispiculatum P. A. Reis, R. C. Oliveira & Valls, sp. nov. – TYPE: BRAZIL. Distrito Federal: Brazlândia, “Poço Azul,” 15°34'54"S, 48°02'50"W, gallery forest, rocky substrate, strongly corrugated, 08 Apr 2011, R. C. Oliveira, A. S. Silva, S. H. Santana & M. W. S. Souza 2612 (holotype: UB!; isotypes: CEN!, IBGE!, SP!, US!).

Similar to *P. bicilium* and *P. polyphyllum*, but differing in the more delicate habit, the annual lifecycle, narrower spikelets, two-nerved upper glume, with two longest tri-

chomes appressed at the apex, and lower lemma two-nerved and glabrous.

Annuals with culms 6–34 cm tall, 0.3–0.8 mm diam, erect, unbranched or little-branched on the lower nodes; internodes glabrous; nodes glabrous, purple-tinged. Leaf sheaths 1.5–3 (–5.2) cm long, papillose-pilose at apex, the trichomes up to 2.5 mm long; ligules 0.8–1 mm long, membranous, with dorsal long trichomes; pseudopetiole absent; leaf blades 2–4 (–6) cm long × (1.2–) 2–2.5 (–3) mm wide, flat, linear to lanceolate, acute or acuminate at apex, slightly attenuate at base, long-pilose on both surfaces. Inflorescences mostly exserted; ‘racemes’ (1–) 2 (–4), the oldest (1–) 2–5 (–6) cm long, alternate along the inflorescence axis, separated by an internode 5 (–20) mm long; pulvini pilose, rachis of the racemes 0.2–0.5 mm wide, winged, green or purple on the wings, glabrous or with few trichomes, ending in a terminal spikelet. Pedicels pilose, pair of spikelet pedicels unequal, basal spikelet 0.2 mm long, upper one to 0.4–1.0 mm long. Spikelets 1.5–1.8 mm long × 0.4–0.6 mm wide, lanceolate or narrowly elliptic, plano-convex, paired, sometimes one spikelet of the pair aborted, 2-seriate. Lower glume absent. Upper glume as long as the spikelet, 1.5–1.8 mm long × 0.4–0.6 mm wide, elliptic, hyaline, with two lateral nerves, the middle nerve absent, dorsally pilose, mainly at the proximal portion, and ciliate with short cilia, 0.4–1 mm long and with two much longer cilia, generally on the distal portion, 0.8–1.7 mm long. Lower lemma 1.3–1.8 mm long × 0.2–0.5 mm wide, linear-lanceolate, acute, flat, glabrous, with two lateral nerves, hyaline. Upper antherium 1.3–1.5 mm long × 0.3–0.5 mm wide, narrowly elliptic, dorsiventrally compressed, stramineous, the youngest hyaline, with few scattered trichomes at the apex, margins of the upper lemma flat, not enclosing the apex of the upper palea; lodicules 2; stamens 3, anthers ca. 1 mm long,

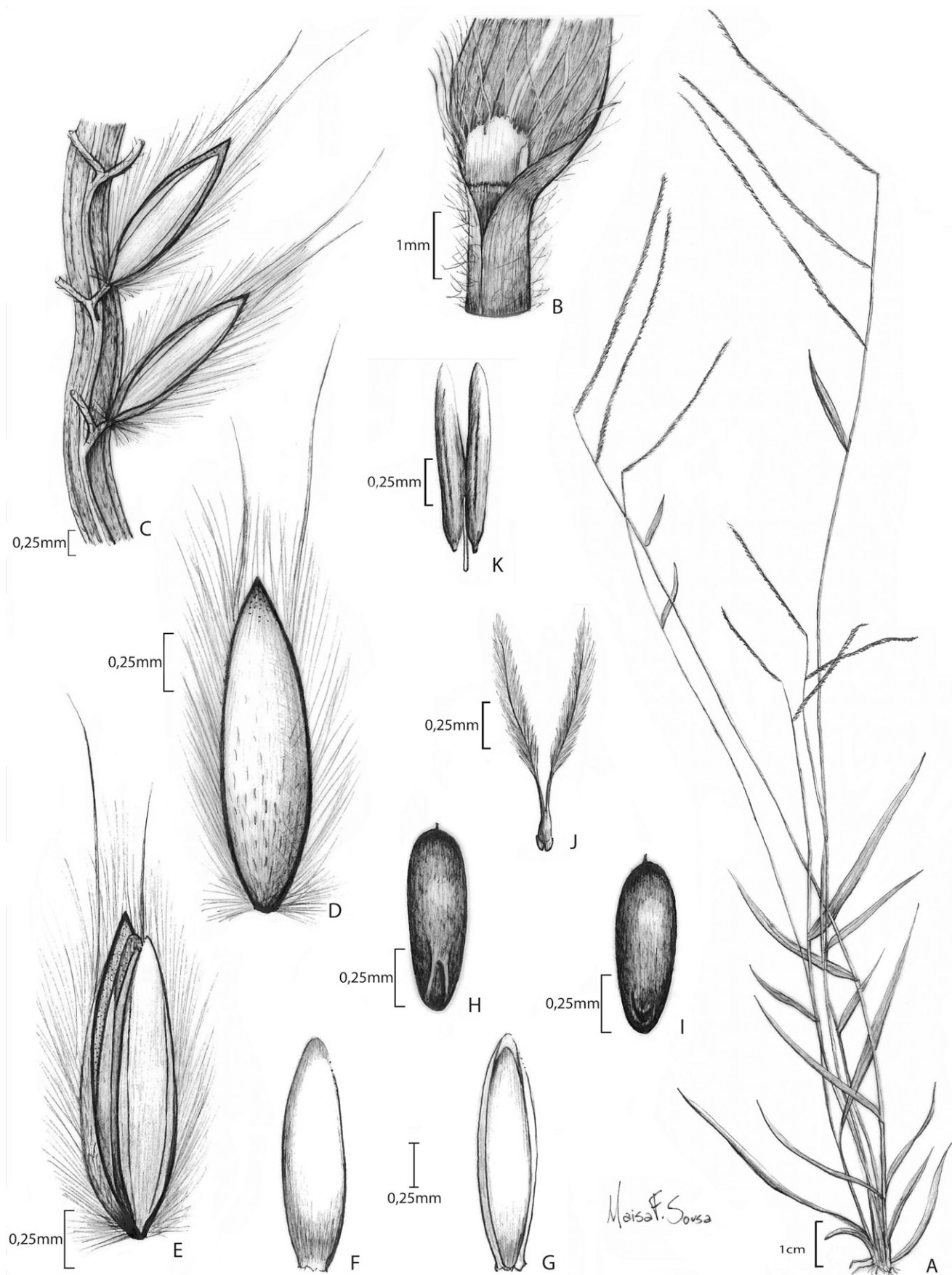


FIG. 2. *Paspalum minutispiculatum*. A. Habit. B. Ligular region of the leaf. C. Detail of the winged rachis with spikelets and pilose pedicels. D. Upper glume, abaxial view. E. Lower lemma, abaxial view. F-G. Upper anthercia, with abaxial views of the lemma (F) and palea (G). H-I. Caryopsis: embryo and hilum side view, respectively. J. Gynoeceum and lodicules. K. Stamen. (Oliveira et al. 2612, caryopsis from Oliveira & Fagg 2745).

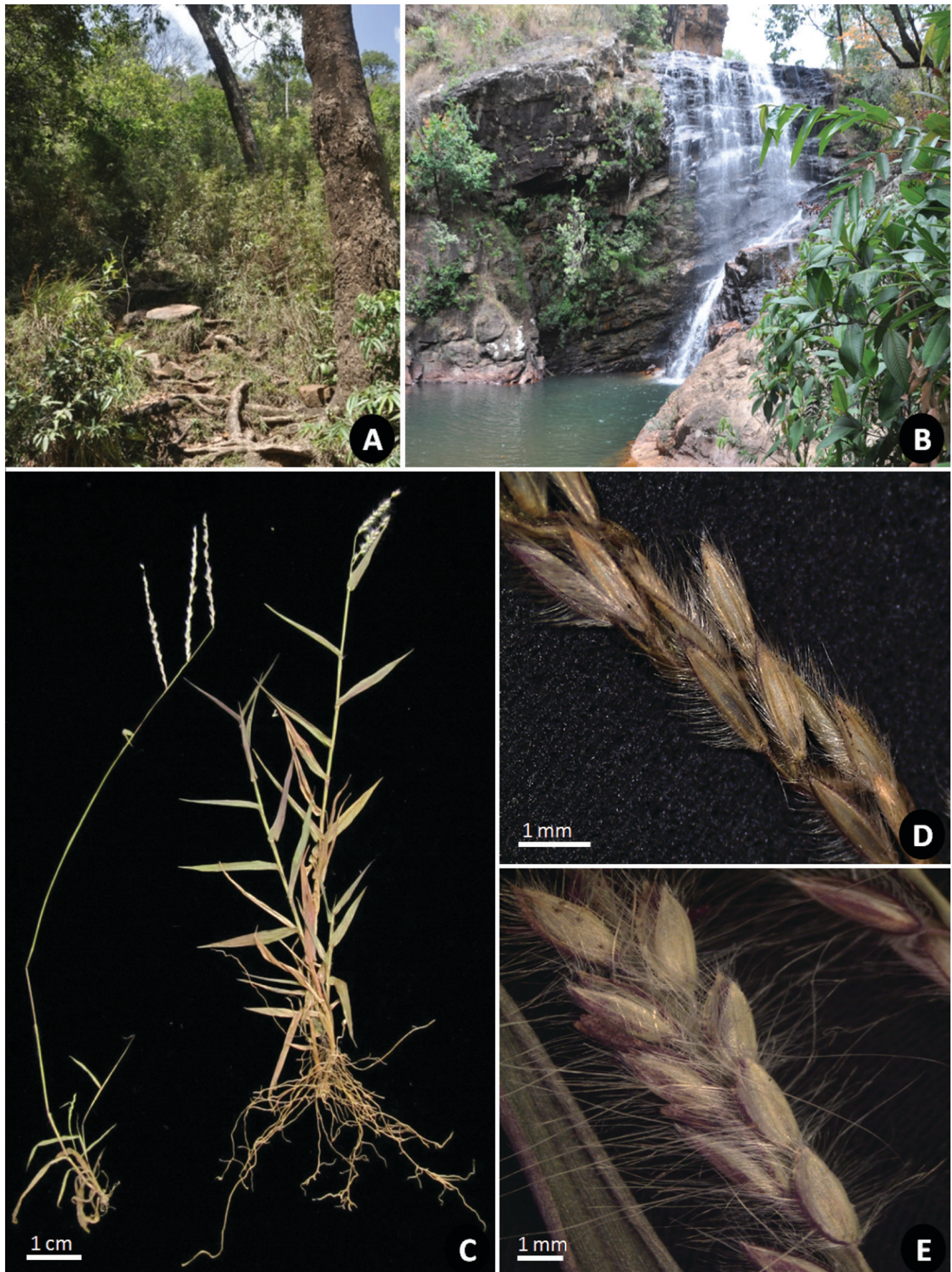


FIG. 3. Habitat and morphology of the two species of *Paspalum*. *P. minutispiculatum*: A. Disturbed gallery forest habitat. C (left). Habit. D. Spikelets. *P. biciliatum*: B. Habitat at the “Poço Azul” waterfall. C (right). Habit. E. Spikelets.

yellow or purple or yellow with purple marks; stigmas 2, purple, plumose. Caryopsis 0.9 mm long × 0.3 mm wide, oblong, dark purple, hilum inconspicuous, probably linear. Figures 2, 3C-left, 3D.

Paratypes—BRAZIL—Distrito Federal: Brasília National Park, “Poço Azul,” 15°34′54″S, 48°02′50″W, 20 Apr 2012, R. C. Oliveira & C. W. Fagg 2745 (CEN, UB); BRAZIL. Distrito Federal: Brasília National Park, “Poço Azul,” 15°34′54″S, 48°02′50″W, 10 Apr 2013, P. A. Reis & A. R. O. Ribeiro 203 (BAA, CEN, CTES, K, P, RB, UB).

Etymology—The epithet of this new annual species refers to the small size of the spikelets.

Habitat, Conservation, and Other Aspects—*Paspalum minutispiculatum* are delicate plants with culms generally lacking branches or with few branches on the lower nodes (Figs. 2A, 3C-left). This new species has been found along shaded trails, in disturbed gallery forest (Fig. 3A); this habitat is rare for *Paspalum* species, since it is primarily a grassland genus. So far it is known only from the “Poço Azul” waterfall in the Brasília National Park, Federal District, Brazil (Fig. 1). *Paspalum minutispiculatum* may have a greater distribution in this area, but has not been collected before, perhaps due to the very short flowering period and the small size of the plants.

The most remarkable differences between *P. minutispiculatum*, *P. bicilium*, and *P. polyphyllum* (Table 1) involve the more delicate habit, the annual cycle, the narrower spikelets, the upper glume with the two longest trichomes appressed and positioned at the apex, the two-nerved glume, and the lower lemma two-nerved and glabrous. Table 1 summarizes eight external morphological differences between the three species.

Paspalum minutispiculatum displays the typical characters of *P.* subg. *Ceresia* sensu Denham et al. (2002): a membranous upper anthecium, palea opening at the apex, not covered by the upper lemma, and a winged rachis. The key below includes annual species allied to *P.* subg. *Ceresia* (Denham et al. 2002) and species closely related to *P. humboldtianum* (Parodi, unpubl. data; Denham et al. 2002; Rua et al. 2008, 2010; Scataglini et al. 2014).

Anatomy—Roots with uniseriate epidermis; one layer of subepidermal parenchyma cells with few sides (4–6 sides in cross section), sometimes these two tissues absent at maturity; a biseriate exodermis with thick cell walls and small cell lumen, followed by one layer of parenchyma cells with few sides (4–5 sides in cross section); endodermis with U-shaped walls, and few passage cells; a uniseriate pericycle with lignified and non-lignified cell walls; vascular cylinder with one to four protoxylem poles; pith lignified (Fig. 4A). Culm with a uniseriate lignified epidermis; one to two subepidermal layers

of cortical sclerenchyma; vascular bundles 18–25, in two levels, with one sheath with thin-walled fibers, and two metaxylem vessels; parenchyma pith fistulous at maturity (Fig. 4B). Leaf blade with a uniseriate epidermis; amphistomatic, subsidiary cells of the stomata cupule-shaped; macrohairs unicellular, filiform, and rigid; microhairs absent; bristles on the leaf margin and the costal region; rows of silica cells in the costal region of both surfaces, and uncommon in the intercostal region (Fig. 4F–G); adaxial surface: hexagonal bulliform cells in the intercostal region, two to four times longer than wide, with anticlinal walls mostly straight; cork cells isolated or in pairs either with another cork cell or with a silica cell, oval or elliptical (Fig. 4F); abaxial surface: bulliform cells absent; papillae uniformly distributed (Fig. 4E), with anticlinal walls mostly straight (Fig. 4G); mesophyll homogeneous with radiate chlorenchyma; vascular bundles 60–80, circular, first order bundles totally locked, second order bundles semi-locked and the remaining bundles free; incomplete parenchyma sheath in first order bundles, complete in other bundles, and sometimes with cells isolated or in pairs and without vascular bundles (Fig. 4E, indicated with arrows); midrib plano-convex, with adaxial subepidermal sclerenchyma fascicles, cortical parenchyma cells larger than the bulliform cells, and with 8–12 vascular bundles (Fig. 4C–D). (Oliveira et al. 2012).

Upper Anthecium—In both *Paspalum minutispiculatum* and *P. bicilium* the upper lemma and palea have sparse macrohairs on the distal portion and at the apex of the abaxial surface (Fig. 5B, G). The adaxial surface of the palea and lemma in both species display few informative characters.

Paspalum minutispiculatum (Fig. 5A–E): abaxial epidermis of lemma with cells 2–4 times longer than wide, surface strongly bullate and densely papillose, each cell with one low electron dense papillose structure at the distal region of the majority of cells (Fig. 5B, indicated with arrow), with anticlinal cell walls markedly sinuous, all the sinuses strongly bullate, and silica cells absent; abaxial epidermis of palea as in lemma, except for the cells 1–2 times longer than wide and sinuses not bullate near the margin (region covered by lemma) (Fig. 5C–E).

Paspalum bicilium (Fig. 5F–I): abaxial epidermis of lemma with cells 8–12 times longer than wide, not bullate, and surface scarcely papillose, some cells with one low electron dense papillose structure at the distal region (Fig. 5G, indicated with arrow), anticlinal cell walls markedly sinuous, sinuses not bullate, and cruciform silica cells present; abaxial epidermis of palea as in lemma (Fig. 5H–J).

The differences in the abaxial epidermis of the lemma and palea between *P. minutispiculatum* and *P. bicilium*

TABLE 1. Morphological differences between *Paspalum minutispiculatum*, *P. bicilium*, and *P. polyphyllum*, based on Silva (2013) and the present work.

Character	<i>P. minutispiculatum</i>	<i>P. bicilium</i>	<i>P. polyphyllum</i>
Life cycle	Annual	Perennial	Perennial
Culm branching	Without branching or with sparse branching at the base	Branching at the base	Branching at the apex
Rachis width	0.2–0.5 mm	0.5–1 mm	1.0–1.5 (2.0) mm
Spikelet length	1.5–1.8 mm	1.2–2.0 (2.5)	2.5–3.5 (4.0) mm
Spikelet width	0.4–0.6 mm	0.8–1.0 mm	1.0–1.5 mm
Nerves of the upper glume and lower lemma	2	5	5
Glume: longest trichome length	0.8–1.7 mm	4–8 mm	2–5 mm
Glume: longest trichome position	Appressed, inserted at apex	Patent, inserted at middle	Patent, inserted along the glume

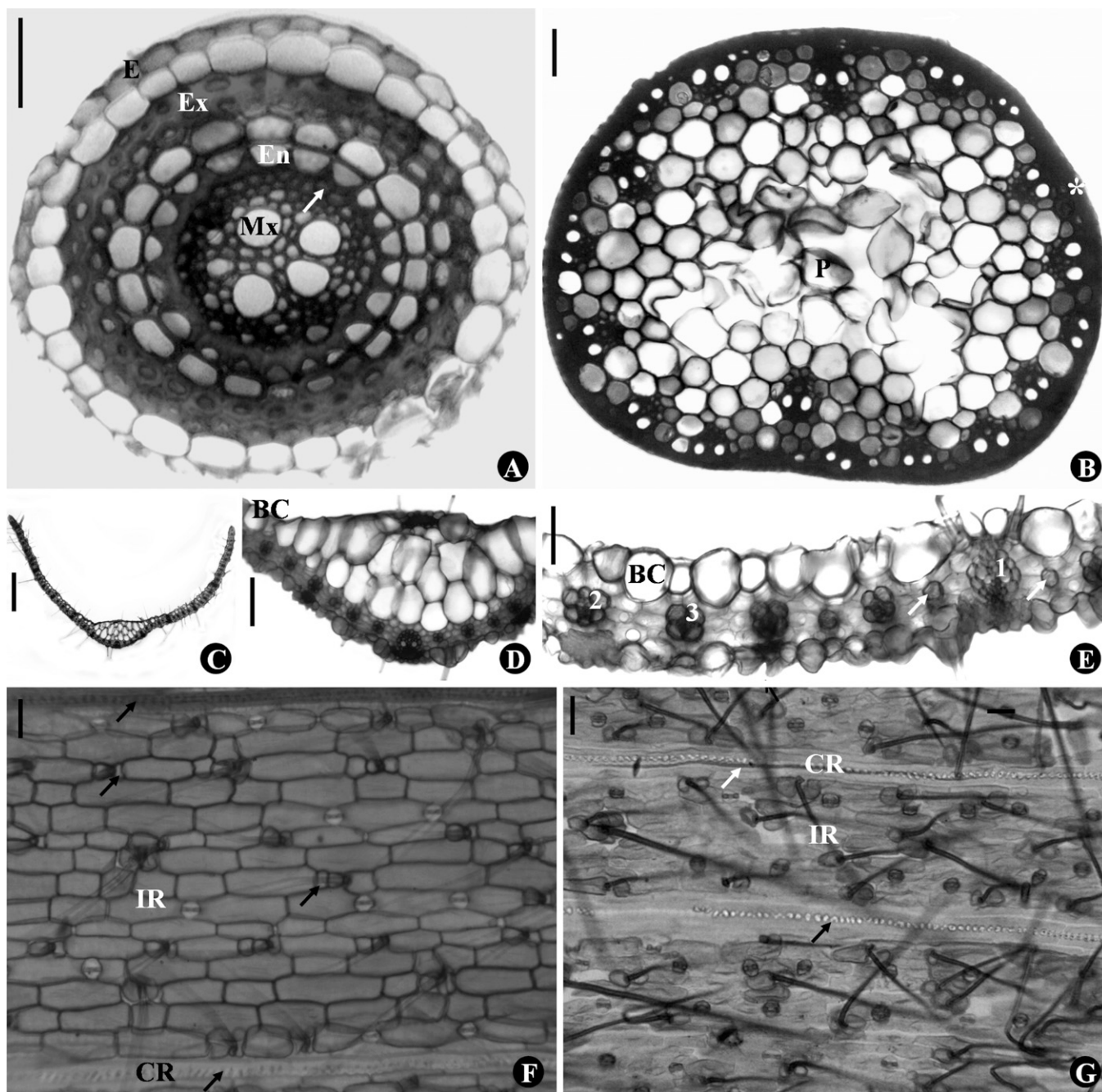


FIG. 4. Anatomy of *Paspalum minutispiculatum* (A–E, transverse sections; F–G, paradermal sections). A. Root; endodermis with U-shaped walls and lignified pericycle (arrow). B. Culm internode with 1–2 subepidermal sclerenchyma layers (asterisk) and fistulous pith. C–E. Leaf blade (D–E, details of C); midrib with cortical parenchyma cells larger than the epidermal bulliform cells (C–D); vascular bundles of first (1), second (2), and third (3) orders, and rare isolated sheath cells (arrows). F–G. Leaf epidermis in adaxial (F) and abaxial (G) views; silica cells (indicated with arrows) in pairs with cortical cells and in rows in the costal region; stomata with cupule subsidiary cells. BC = bulliform cell; E = epidermis; En = endodermis; Ex = exodermis; CR = costal region; IR = intercostal region; Mx = metaxylem; P = pith. Scale bars: A–B, D–E = 50 μ m, C = 200 μ m, F–G = 100 μ m.

are: the presence of cruciform silica cells, the presence of strongly bullate sinuses in the cells, and distribution of papillose structures on the surface of the lemma and palea.

The anatomical and micromorphological comparison between *P. minutispiculatum*, *P. biciliatum*, and *P. polyphyllum* are shown in Table 2, with a total of 14 differences between the three species.

KEY TO THE ANNUAL SPECIES OF *P. SUBG. CERESIA* AND SPECIES CLOSELY RELATED TO *P. HUMBOLDTIANUM*

- | | |
|---|--------------------------|
| 1. Spikelets awned | <i>P. longiaristatum</i> |
| 1. Spikelets awnless | 2 |
| 2. Spikelets 4–4.3 mm long; upper glume winged with flat margins; upper antherium stipitate | <i>P. cachimboense</i> |
| 2. Spikelets 1.5–3.5 (4.0) mm long; upper glume not winged; upper antherium not stipitate | 3 |

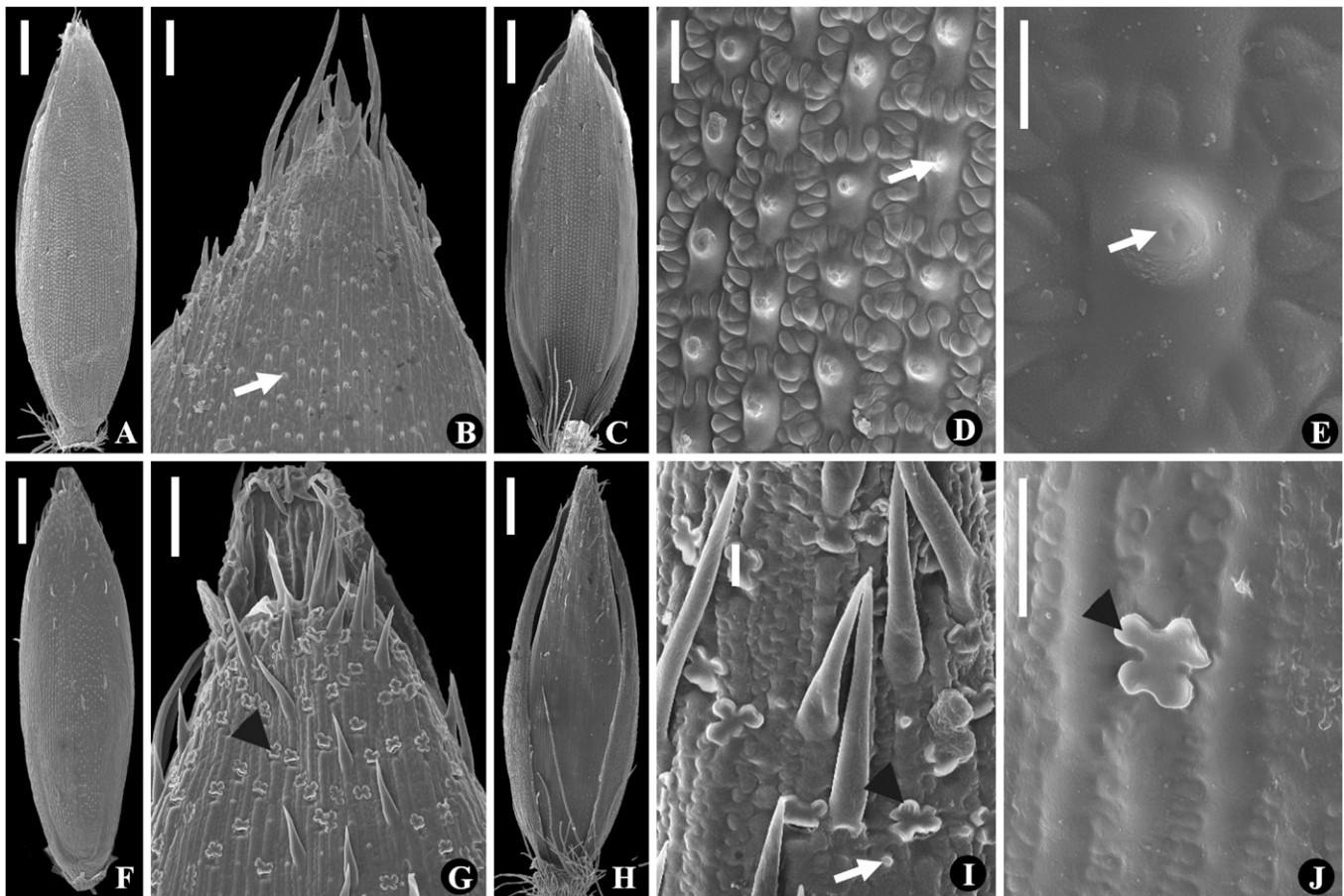


FIG. 5. Upper anthercium of *Paspalum minutispiculatum* (A-E) and *P. bicilium* (F-J) seen in SEM. A-B, F-G. Lemma; ciliate apex, surface with papillose projections (B, indicated with arrow), and cruciform silica cells (G, indicated with black arrowhead). C-E, H-J. Palea; cells with papillose projections (D, E, I, indicated with arrows), bullate sinuses (D) at the middle of the palea, and non-bullate sinuses (E) at the palea margins, and cruciform silica cells (I-J, indicated with black arrowheads). (Oliveira et al. 2612). Scale bars: A, C, F, H = 200 μ m; B, G, I = 50 μ m; D, J = 10 μ m; E = 5 μ m.

3. Rachis 6–10 mm wide 4
4. Spikelets 1.2–1.4 mm wide; upper glume flat; lower lemma 93–97% as long as the upper glume, ca. 1 mm wide, flat to slightly convex, acute, pilose throughout on the abaxial surface; culms freely branching *P. ceresia*
4. Spikelets ca. 1 mm wide; upper glume with inrolled margins and slightly recurved apex; lower lemma 64–84% as long as the upper glume, 0.4–0.5 mm wide, concave, obtuse, glabrous, and glossy on the abaxial surface; culms unbranched or branching just at the lowermost nodes *P. vexillarium*
3. Rachis 0.5–3 mm wide 5
5. Rachis falcate; spikelets glabrous or sparsely pubescent *P. falcatum*
5. Rachis straight; spikelets conspicuously pilose 6
6. Upper glume with unequal cilia 7
7. Annual; spikelet 0.4–0.6 mm wide; upper glume two-nerved, with the two longest trichomes 0.8–1.7 mm long, inserted at apex of glume margin; lower lemma 2-nerved *P. minutispiculatum*
7. Perennial; spikelet 0.8–1.5 mm wide; upper glume 5-nerved, with two, four or several trichomes 2–8 mm long at middle or inserted along the glume margin; lower lemma 5-nerved 8
8. Rachis 0.5–1 mm wide; spikelet 0.8–1.0 mm wide; upper glume with two longest trichomes (eventually with four longest trichomes, but two longer than the other two), at the margin of the middle of the glume *P. bicilium*
8. Rachis 1–2.5 mm wide; spikelet 1–1.5 mm wide; upper glume with several longest trichomes, all of equal length, along the entire glume margin 9
9. Pseudoligule with a ring of hairs; blades narrowing to the base; rachis 2–2.8 mm wide, ending in a naked point; southern Mexico to Nicaragua *P. cymbiforme*
9. Pseudoligule absent; blades wider at the base; rachis 1–1.5(2) mm wide, usually ending in a spikelet; South America *P. polyphyllum*
6. Upper glume with equal cilia or without cilia 10
10. Rachis triquetrous; upper glume not ciliate; upper anthercium closed at the apex *P. paucifolium*
10. Rachis winged; upper glume ciliate; upper anthercium open at the apex 11
11. Racemes (4) 7–11, arcuate; rachis 0.8–1.2 (–1.4) mm wide; spikelets 0.8–1 (–1.3) mm wide, sparsely distributed; blades papyraceous *P. buchtienii*
11. Racemes 2–6, not arcuate; rachis 1.2–1.5 mm wide; spikelets 1.2–1.4 mm wide, imbricate; blades herbaceous *P. humboldtianum*

TABLE 2. Anatomical and micromorphological differences between *Paspalum minutispiculatum* and *P. bicilium*, based on Silva (2011) and this study.

Character	<i>P. minutispiculatum</i>	<i>P. bicilium</i>
Culm epidermis	Lignified	Not lignified
Culm subepidermal sclerenchyma layers	1–2	4–6
Culm pith	Fistulose	Solid
Culm vascular bundle number	18–25	25–31
Leaf midrib shape	Plano-convex	Plane
Leaf total number of vascular bundles	60–80	70–90
Leaf midrib parenchyma cell sizes	Different sizes	Same size
Leaf midrib parenchyma cell sizes compared with bulliform cell size	Larger	Smaller
Leaf costal regions: bristles	Present	Absent
Leaf abaxial surface	Densely papillose	Very sparsely papillose
Leaf abaxial epidermis: anticlinal cell walls	Straight	Sinuuous
Upper lemma and palea abaxial epidermis - silica cells	Absent	Present, cruciform
Lemma: cell length of abaxial epidermis	2–4 times longer than wide	8–12 times longer than wide
Upper palea abaxial epidermis - sinuses	Strongly bullate	Not bullate

DISCUSSION

The ciliate margin of the upper glume of *Paspalum minutispiculatum* resembles that of the informal group “Humboldtiana” (proposed by Parodi and Nicora, unpubl. manuscript). The small size of the spikelets and the two longest cilia in the upper glume (Figs. 2D–E, 3D) suggest that *P. minutispiculatum* is morphologically closer to *P. bicilium* (Figs. 3C-right, E), according to the circumscription of the latter species as cited by Filgueiras (1991), Oliveira and Valls (2014), and supported by Peñaloza et al. (2008), and not as a mere synonym of *P. polyphyllum* (Silva 2013). On the other hand, *P. minutispiculatum* differs from *P. bicilium* and *P. polyphyllum* by several characters (Fig. 3; Tables 1, 2), as well as from the other members of the “Humboldtiana” clade and the rest of *P. subg. Ceresia*, by the annual life cycle. There are at least 22 differences between *P. minutispiculatum*, *P. bicilium*, and *P. polyphyllum* (Tables 1, 2), and this fact evidences they are good species.

Paspalum bicilium is a riparian species, and occurs in the Cerrado biome (Silva 2013; Fig. 3B), while the new species grows along trails in gallery forest (Fig. 3A), a rare habit in this genus, and appears to be endemic to the Federal District. At the “Poço Azul” waterfall, *P. bicilium* can be found at the river margin, whereas *P. minutispiculatum* occurs exclusively along shaded trails in the adjacent gallery forest. Populations of *P. bicilium* are separated by only a few meters from those of *P. minutispiculatum* (Fig. 3A–B). We assess *P. minutispiculatum* as “data deficient” (DD) according to the IUCN red list criteria (IUCN 2001), on account of the insufficient information available.

In Poaceae, leaf anatomy assembles many characters that have been used for taxonomic purposes, however, we believe that root and stem anatomy are under-explored. The root and culm internode of *Paspalum minutispiculatum* were anatomically analyzed here, and these organs showed differences from other species of *Paspalum*. The new species presents two or more exodermal layers with highly lignified cells in the root cortex, endodermis with U-shaped walls, and thin lignified walls in the pith cells (Fig. 4A), whereas in the root of *P. rostratum* D. Ramos, Valls & R. C. Oliveira the exodermis is uniseriate and slightly thickened, the endodermis has “O” shaped thickenings, and the pith cells have thick lignified walls (Ramos et al. 2011). As in *P. minutispiculatum*, the endodermis of *P. dilatatum* Poir. has “U” thickenings, which has the particularity of a cortical aerenchyma (Vasellati

et al. 2001). Therefore, root anatomy can provide taxonomic characters for the distinction of *Paspalum* species.

Regarding the anatomy of the culm internode, *P. minutispiculatum* has vascular bundles in only two levels of depth in the ground tissue, which are not totally surrounded by the subepidermal sclerenchyma, which has 1–2 cell layers (Fig. 4B), while *P. rostratum* has vascular bundles with 3–4 levels, and the majority are surrounded by the subepidermal sclerenchyma, which has 10–14 layers (Ramos et al. 2011). As in the roots, stem anatomy can be more useful to the taxonomy of Poaceae than has been explored.

Studies using SEM have been useful in the characterization of *Paspalum* species. Morrone and Zuloaga (2003) showed that the distribution and shape of papillae, and the presence of bicellular microhairs in the upper antherium distinguish *P. plowmanii* Morrone & Zuloaga and *P. filgueirae* Morrone & Zuloaga. These two species differ from *P. minutispiculatum* and *P. bicilium* as presented here, since the latter two have papillose structures formed by longer and higher cells (Fig. 5), while the bicellular microhairs are absent in the lemma margin of the former two species. Therefore, the micromorphological characters of the upper antherium on the abaxial surface have proved to be of high taxonomic value for this genus.

According to Fahn (1990) and Evert (2006), a papilla is a trichome with a projection of the external periclinal wall and smaller than the hairs. In the present work, papillae were found on the abaxial leaf epidermis of *Paspalum minutispiculatum* (Fig. 4E). On the other hand, the surfaces of the antheria of both species studied here do not have papillae, but have a papillose surface, as the epidermal abaxial cells display a hemispherical or conical protuberance (Fig. 5, indicated with arrows).

Papillae and epidermal cells with a papillose surface have not been distinguished in the literature, regarding the upper antherium epidermis in Poaceae, and all cells have been referred to as papillae (Morrone and Zuloaga 2003, Morrone et al. 2004; Denham and Zuloaga 2006). The papilla has all the periclinal cell wall projected in a conical shape (Fig. 4E), and can be associated with silica crystals or not. On the other hand, the epidermal cells with a papillose surface display some areas with projections (Fig. 5, indicated with arrows), that are associated with silica crystals. The nature of both structures is different. The standardization of these characters is lacking in the literature, despite their taxonomic importance, and may have possible repercussions if they are used in phylogenetic analyses.

Both *Paspalum minutispiculatum* and *P. bicilium* have papillose cells in the abaxial epidermis of the lemma and palea, with one low electron dense structure, which has a closer distribution in *P. minutispiculatum* than in *P. bicilium*. Silica accumulated in the papillae-like protrusions are similar structures to those observed in the leaves of species of *Otachyrium* Nees (Panicoideae, Poaceae; Ribeiro et al. 2001), *Cyperus* L., and *Rhynchospora* Vahl (Cyperaceae; Estelita and Rodrigues 2012). The structures were described as “silica cells” and form conical projections of silica deposits in the inner periclinal wall of the abaxial epidermal cells that are surrounded by the outer periclinal wall (Estelita and Rodrigues 2012). Despite the presence of silica in such cells, they are restricted to the papillose protrusions, therefore we distinguished them from the cruciform silica cells at the apex of the palea and lemma of *P. bicilium* (Fig. 5G, I–J), as well as from all types of silica cells commonly described for Poaceae (Bonnett 1972; Ellis 1979), which have considerable value for the taxonomy of this family.

The leaves of *Paspalum minutispiculatum* are flat (Figs. 2A, 3C-left), but in transverse section they are concave, appearing involute (Fig. 4C). This is due to the dehydration of tissues, where the bulliform cells of the adaxial epidermis lose a greater volume than in the cells of the abaxial side.

ACKNOWLEDGMENTS. We are grateful to Maisa F. Sousa for the line drawings, to André Rodolfo Oliveira Ribeiro and Daniel Villarroel Cegarra for preparing the map, to the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Brasília, Brazil for financial support (474880/2012–5), for the Protax MSc. grants (561768/2010–2) awarded to two of the authors, and for a productivity grant (311844/2006–4) awarded to the fourth author.

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