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# Simonachne, a new genus for Australia segregated from Ancistrachne s.l. (Poaceae: Panicoideae: Paniceae) and a new subtribe Cleistochloinae

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#### **ABSTRACT**

A new genus, Simonachne E.J.Thomps. is described and Ancistrachne maidenii (A.A.Ham.) Vickery is transferred to it as Simonachne maidenii (A.A.Ham.) E.J.Thomps. The new subtribe Cleistochloinae E.J.Thomps. is described and is composed of four genera, Calyptochloa, Cleistochloa, Dimorphochloa and Simonachne, united by distinctive morphology that is associated with reproductive dimorphism. Phenetic analyses were used to examine the similarities of taxa and to test the consistency of results with variation in analysis inputs. Input variations included the dataset in terms of composition of the samples and morphological characters, and the cluster analysis algorithms, viz. classification, ordination and association measure. A baseline dataset was used for comparison of results and comprised 24 samples and 161 characters relating to anatomy, micro- and macromorphology of spikelets, leaves and fertile culms. Three major clusters were resolved, Cleistochloinae ('the cleistogamy group'), Neurachninae in its original sense, and a cluster referred to as the 'paniculate inflorescence group' composed of Ancistrachne s.s., Entolasia and Panicum s.s. The results were congruent with a recent phylogenetic study that showed that Ancistrachne s.l., Cleistochloa s.l. and Dimorphochloa s.l. were not monophyletic. The process provided an array of morphological characters for descriptions of species and for distinguishing taxa at multiple ranks in natural groups, components of alpha and beta taxonomy respectively.

**Keywords:** Ancistrachne, Ancistrachne maidenii, Calyptochloa, Cleistochloa, Cleistochloinae, clusters, Dimorphochloa, morphology, Neurachninae, phenetics, similarity, Simonachne, 'the cleistogamy group'.

#### Introduction

A recent phylogenetic reconstruction of the panicoid grass subtribe Neurachninae sensu Morrone et al. (2012) showed that it consists of three major clades, viz. 'the cleistogamy group', the 'paniculate inflorescence group' and Neurachninae s.s., and, additionally, that the genus Ancistrachne sensu Vickery (1961b) is not monophyletic (Thompson and Fabillo 2021). 'The cleistogamy group' was represented by Ancistrachne maidenii (A.A.Ham.) Vickery and placed sister to Calyptochloa C.E.Hubb., Cleistochloa s.l. and Dimorphochloa s.l., with all taxa united by several morphological characters and, most notably, by a rare dimorphic breeding system (Table 1). Ancistrachne s.s. was sister to Entolasia Stapf. in the 'paniculate inflorescence group', defined by the terminal inflorescences in panicles and several characters relating to spikelets. Neurachninae in its original sense as circumscribed by Clayton and Renvoize (1986) comprised Neurachne R.Br. (syn. Paraneurachne (Hack.) S.T.Blake) and Thyridolepis S.T.Blake that share a definitive combination of characters (Clayton and Renvoize 1986; Thompson and Fabillo 2021).

The dimorphic breeding system of the 'the cleistogamy group' has a distinctive morphology in tribe Paniceae. It consists of two types of inflorescences, terminal and axillary, in different locations on the same plant (amphigamy), with corresponding spikelet dimorphism (Fig. 1, 2). The spikelet dimorphism across the genera is either of

Table I. Major differences in morphological characters between Simonachne maidenii (=Ancistrachne maidenii) and other putative genera in subtribe Cleistochloinae and Ancistrachne s.s.

Character	Simonachne	Calyptochloa <sup>A</sup>	Cleistochloa <sup>B</sup>	Dimorphochloa	Dimorphochloa spp.	Ancistrachne
Growth habit <sup>F</sup>		Stoloniferous		hizomotous	Tufted, decumbent	Rhizomotous
Terminal inflorescence	Reduced spike-like panicle	Raceme	Reduced	spike-like panicle 5?	Raceme	Open panicle, secondary branches reduced or absent <sup>G</sup>
		(Primary	branches reduced or abse	ent <sup>G</sup> )		reduced or absent
Axillary inflorescences <sup>F</sup>		Axillary raceme	sometimes reduced to o		Absent	
		(Dimorphochloa rigida has much i	reduced CL racemes apica	al on subordinate branches)		
Terminal and axillary spikelets	Similar	Distinctly differen	ent	Similar	Distinctly different	Terminal only
Long white hairs on leaf sheath and proximal margin of leaf blades	Present					Absent
Leaves disarticulating D,F; relative duration of leaves	Yes, ph	notosynthesising in culms; long	Ye	Yes, photosynthesing in culms; very short		
Ligule		A fringe of hairs <sup>F,G</sup>				Fringed membrane; membrane and cilia approximately equal length or shorter
Upper margin of leaf sheath			Ciliate			Glabrous
Contraligule present <sup>C</sup>		Yes		No	Yes	No
Spikelet compression	Dorsi-ventral	Strongly dorsi-ventral		Dorsi-ventral		Little to none
Orientation of CH spikelets with respect to culm			Adaxial			Abaxial <sup>E</sup>
Orientation of terminal spikelets on the pedicel <sup>C</sup>		Transverse				Oblique
CH spikelet indumentum <sup>F</sup>	Woolly <sup>C</sup>		Straight			Uncinate <sup>C</sup>
CH lower glume <sup>F</sup>		Small to vestigial		Absent	Small	50% of spikelet length
CH lower glume enveloping the upper glume at the base			No			Yes

Table I. (Continued)

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Character	Simonachne	Calyptochloa <sup>A</sup>	Cleistochloa <sup>B</sup>	Dimorphochloa	Dimorphochloa spp.	Ancistrachne
Texture of the margins of the upper glume		D		Same as body		
Number of veins on the upper glume	7	7	5	5–7	5	7–11
CH upper glume vs lower lemma length	Subequal	Longer	Shorter	Subequal to equal		Subequal to equal
Trichomes on lower lemma in distal portion of ventral surface	Glabrous	Sparse, ~40 μm long, tubular	Dense, to 120–220 µm long, flattened	Common, to ~100 μm long, tubular	Dense, to ~50 μm, flattened	30–45 μm long, tubular; ~30 μm long prickles
Indumentum on dorsal surface of CH upper glume vs lower lemma	Similar	Dissimilar		Similar		Similar
Lower palea			Absent <sup>G</sup>			~1/3 lower lemma
Relative length of the upper lemma to the lower lemma	Shorter	Much shorter		Shorter		Equal to shorter
Rachillar stipe below upper floret <sup>C</sup>				Present		
Apex of CH upper lemma <sup>F</sup>	Mucronate	Awned	Awned	Mucronate		
Germination lid on CH upper lemma	Inconspicuous crescent-shaped depression between a raised portion of the two veins	Conspicuous crescent-shaped depression	Inconspicuous			
Texture of upper lemma		Chartaceous	Thin, hardened, glossy; tightly clasping caryopsis			
Texture of margin of upper lemma at apex		Tapering from slig	Hard and brittle throughout			
CH upper lemma minutely longitudinally ridged			No			
CH upper lemma with papillae	No		Yes			Yes
CH upper palea creased at apex		No				Yes

Table I. (Continued)

Character	Simonachne	Calyptochloa <sup>A</sup>		Cleistochloa <sup>B</sup>	Dimorphochloa	Dimorphochloa spp.	Ancistrachne
Orientation of CL spikelets		Adaxial			Abaxial	Adaxial	Axillary CL spikelets absent
CL lower lemma with an elaiosome		Absent		Large	Medium	Small	Axillary CL spikelets absent
CL upper glume and lower lemma	Chlorophyllous		With or without chlorophyll	Lacking chlorophyll	Chlo	rophyllous	Axillary CL spikelets absent
CH caryopses							
Longitudinal outline	Elliptical		Elliptical	Elliptical	Ovate	Ovate	Ovate
Cross-sectional shape	Narrow elliptical		Narrow elliptical	Narrow elliptical	Narrow elliptical	Narrow elliptical	Elliptical
Stylopodium	Vestigial		Vestigial	Narrowly fused	Narrowly fused	Vestigial	Broadly fused
Shape of scutellum	Elliptical		Elliptical	Elliptical	Broadly elliptical	Elliptical	Broadly elliptical
Surface pattern of pericarp	Finely longitudinally striate Smooth		Coarsely longitudinally striate		Smooth	Coarsely longitudinally striate	
Photosynthetic pathway <sup>F</sup>	C <sub>3</sub> (for			rest shade species)			C <sub>3</sub> (forest shade species)
Radiate mesophyll <sup>F</sup>	Distinct		Indistino	t or incomplete		Relatively distinct	Indistinct or incomplete

Characters shown in bold considered by Vickery (1961) to discriminate A. maidenii. CH, chasmogamous; CL, cleistogamous.

<sup>&</sup>lt;sup>A</sup>Includes Cleistochloa sp. (Duaringa K. B. Addison 42).

<sup>&</sup>lt;sup>B</sup>Cleistochloa sensu Hubbard (1931).

<sup>&</sup>lt;sup>C</sup>Uncommon character in Paniceae.

<sup>&</sup>lt;sup>D</sup>Not used in character list by Thompson and Fabillo (2021).

<sup>&</sup>lt;sup>E</sup>Used by Morrone et al. (2012) to define subtribe Neurachninae.

FUsed by Kellogg (2015) for to define Cleistochloa.

<sup>&</sup>lt;sup>G</sup>Used by Kellogg (2015) for to define Neurachninae.



**Fig. 1.** Inflorescences of *Simonachne maidenii* (=*Ancistrachne maidenii*). (a) Fertile culm of showing amphigamous inflorescences comprising terminal reduced spike-like panicles (TI) and axillary racemes (AI). (b) Terminal inflorescence. (c) Axillary inflorescence. All from *Thompson EJT936* (BRI). Photos: Queensland Herbarium. Scale bar: 10 cm.

two forms, namely similar or very different (Fig. 2). Ancistrachne maidenii and Dimorphochloa rigida S.T.Blake are similar in that both species have relatively inconspicuous spikelet dimorphism compared with other species in the group (Fig. 2). Conversely, Calyptochloa, Cleistochloa s.l. and Dimorphochloa s.l. have axillary spikelets that are very different from their corresponding terminal spikelets. All species in 'the cleistogamy group' share anther dimorphism where anthers in the axillary spikelets are considerably smaller than those in the terminal spikelets. The axillary spikelets, if appraised independently, could be considered to belong to a different taxon in a similar perspective as remarked by Chase (1918, p. 255) in her assessment of

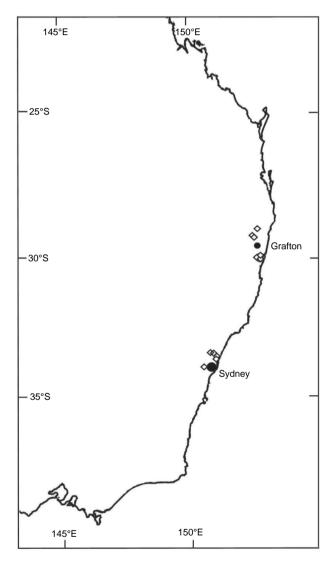
several North American grasses with dimorphic breeding systems. The spikelets in the axillary inflorescences are obligately cleistogamous (CL), i.e. pollination takes place within a closed flower that never opens, whereas the spikelets in the terminal inflorescences are usually chasmogamous (CH), as for most grasses, or uncommonly mixed with CL spikelets in some species.

Ancistrachne, in its original sense as circumscribed by Blake (1941), is unique in the Australian Paniceae. The genus is identifiable by a combination of characters, including spikelets that are tilted on the pedicels, adorned with hooked hairs, and exhibit abaxial orientation (Webster 1987; Simon 2002). Ancistrachne sensu Blake (1941) comprises



Fig. 2. (Caption on next page)

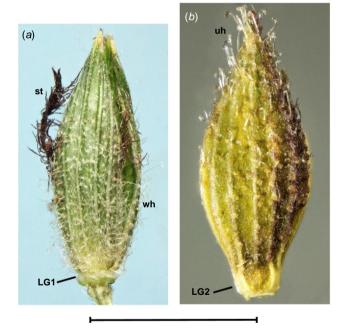
three species, *viz. A. uncinulata* (R.Br.) S.T.Blake, an Australian endemic occurring along the eastern coast of Australia, and two species from the tropical Asia–Pacific region, *A. ancylotricha* (Quisumb. & Merr.) S.T.Blake and *A. numaeensis* (Balansa) S.T.Blake, recorded in the Philippines and New Caledonia respectively. *A. ancylotricha* was omitted from the study by Thompson and Fabillo (2021) becaue of poor quality of extracted DNA and polymerase chain-reaction failures. However, *Ancistrachne maidenii*, a rare species from the eastern coast of Australia (Fig. 3)



**Fig. 3.** Distribution of Simonachne maidenii (=Ancistrachne maidenii) on the basis of herbarium specimen point data.

transferred to *Ancistrachne* from *Eriochloa* in 1961 (Vickery 1961*a*), has woolly hairs and lacks the hooked hairs and differs from *Ancistrachne s.s.* in other morphological characters (Hamilton 1913; Vickery 1961*a*, 1961*b*; Simon 2002; Thompson and Fabillo 2021; Table 1, Fig. 4).

The affinities and placement of Ancistrachne s.l. (i.e. including A. maidenii) have differed in several studies using various methods of analysis of morphological or molecular data (Table 2). Ancistrachne s.l., Calyptochloa, Cleistochloa S.T.Blake and Dimorphochloa rigida have frequently been grouped together and occasionally Entolasia has been allied to these genera (Table 2). In the intuitive diagram of morphological relationships of genera in subtribe Setariinae by Clayton and Renvoize (1986), Ancistrachne s.l. had close affinity to Panicum L., reflecting the basionym for A. uncinulata, Panicum uncinulatum R.Br., and Calyptochloa and Cleistochloa s.s. were grouped with Entolasia. In molecular phylogenetic reconstructions, Ancistrachne s.l. was positioned in subtribe Neurachninae s.l. with Calyptochloa, Cleistochloa s.s. (subsuming D. rigida), Neurachne and



**Fig. 4.** Lower lemmas from chasmogamous spikelets from terminal inflorescences: (a) Simonachne maidenii; (b) Ancistrachne uncinulata; (a) from Thompson EJT936 (BRI) and (b) from Thompson EJT1018 (BRI). Photos: E. J. Thompson. LGI, lower glume; st, stigma emergent at half way along spikelet; wh, woolly hairs; uh, uncinate macrohairs. Scale bar: 2 mm.

Fig. 2. Spikelets of species from subtribe Cleistochloinae ('the cleistogamy group') with view of the lower lemma. (a and b) Simonachne maidenii (=Ancistrachne maidenii) from Thompson EJT936. (c and d) Dimorphochloa sp. (Miles E. J. Thompson EJT906) from E. J. Thompson MOR931. (e and f) Calyptochloa cylindrosperma from Thompson EJT786. (g and h) Dimorphochloa rigida from Thompson EJT959. (i and j) Cleistochloa sclerachne from Thompson EJT981. Photos: E. J. Thompson. el, remnants of elaiosome. Scale bar: 5 mm.

Table 2. The main sources of morphological data and taxa relationships used in this study.

				B 1			
References	Number of morphological	Paniceae taxa		Context	Sampling	Remarks	
	characters	Species	Genera				
Hilu and Wright (1982)	87		35	Poaceae	Au, Cg, Csub, Dr	The four genera clustered with Entolasia and Ichnanthus	
Clayton and Renvoize (1986)	Not specified		All	Poacaeae	Au, Am, Dr	The three genera placed in subtribe Setariinae showing affinity to <i>Panicum L.</i> , but <i>Ancistrachne</i> being distant from <i>Calyptochloa</i> and <i>Cleistochloa s.l.</i> grouped with <i>Entolasia</i> . Setariinae characterised by mostly hardened upper floret.	
Webster (1987)	199	308	45	Australian Paniceae	Au, Am, Dr		
Watson and Dallwitz (1992)	425		All	Poacaeae	Au, Am, C s.s., Dr		
Zuloaga et al. (2000)	67	110	105 <sup>A</sup>	Paniceae	Au, Cg, Dr	Overlapping character sets; no character selection criteria.	
Morrone et al. (2012)	57	208	113	Paniceae	Au, Cg, Dr	Calyptochloa and Dimorphochloa rigida clustered with Entolasia.	
Thompson and Fabillo (2021)	104	22	П	Neurachninae	Au, Am, Ca (3) <sup>B</sup> , Cl (3) <sup>B</sup> , Dr, D. spp. (2) <sup>B</sup>	'The cleistogamy group' represented by the 10 putative species. Ancistrachne s.s. grouped with Entolasia in the 'paniculate inflorescence group'.	

Am, Ancistrachne maidenii; Au, A. uncinulata; Ca, Calyptochloa; Cg, C. gracillima; Cl, Cleistochloa s.l.; Csub, C. subjuncea; Dr, Dimorphochloa rigida; D. spp., Dimorphochloa spp. with phrase names.

Alncludes subgenera and sections for Panicum and Paspalum.

<sup>&</sup>lt;sup>B</sup>Number of species.

Thyridolepis (Christin et al. 2012; Grass Phylogeny Working Group II 2012; Morrone et al. 2012; Soreng et al. 2015, 2017). In these studies, Ancistrachne uncinulata was used as the sole representative of Ancistrachne s.l., usually with one other species from each of the other four genera in Neurachninae s.l. (Table 2). Morrone et al. (2012) considered Neurachninae to have two uniting characters, viz. inflorescences with reduced secondary branching, and abaxial spikelet orientation. However, most of the taxa in the clade have reduced primary branching and lack secondary branches, although Ancistrachne s.s. sometimes bears secondary branching. Ancistrachne s.s., Neurachne and Thyridolepis have abaxial spikelet orientation, whereas members of 'the cleistogamy group' have adaxial spikelet orientation (Webster 1987; Thompson and Fabillo 2021; Table 1). All genera in Neurachninae s.l. have C3 photosynthetic pathway, although some species of Neurachne have been recorded as having C<sub>3</sub>-C<sub>4</sub> intermediacy (Hattersley et al. 1982; Christin et al. 2012). On the basis of the phylogenetic reconstructions by Morrone et al. (2012) and Grass Phylogeny Working Group II (2012), Kellogg (2015) synonomised Ancistrachne s.l. and Calyptochloa with Cleistochloa s.s. and listed several uniting characters (Table 1). Byn contrast, Ancistrachne uncinulata, Calyptochloa, Cleistochloa s.l. and Dimorphochloa s.l. have been maintained by Brown and Bostock (http://data.qld.gov.au/dataset/census-of-thequeensland-flora-2020/, accessed 15 May 2021), although the latter two genera are not monophyletic (Thompson and Fabillo 2021).

Thompson and Fabillo (2021) refined the distinction of taxa in the 'the cleistogamy group', comprising five putative genera and 10 species, by the inclusion of 37 characters that relate to the CL inflorescences and spikelets (Fig. 1, 2, Tables 1, 2). Prior to this, very few CL characters were used in keys and descriptions (Tothill and Hacker 1983; Clayton and Renvoize 1986; Webster 1987; Watson and Dallwitz 1992; Simon 2002; Simon and Alfonso, see http://ausgrass2.myspecies.info/, accessed 21 September 2020). For example, of the 157 inflorescence and spikelet characters used by Webster (1987), two relate to CL. Dimorphochloa rigida was subsumed in Cleistochloa s.s. because Dimorphochloa was considered to lack merit as a genus in the context of generic concepts used in Paniceae despite the five characters recognised by Blake (1941), Webster (1987), Clayton and Renvoize (1986) and Clayton (1987). Furthermore, only one of the characters used by Zuloaga et al. (2000) and Morrone et al. (2012) relates to CL.

The taxonomy of *Ancistrachne maidenii* has also vacillated. Vickery (1961a) considered the generic affinities of *Eriochloa maidenii* A.A.Ham. to be ambiguous but subsumed it in *Ancistrachne* on the basis of several characters (Table 1). Her opinion was that the generic circumscription of *Ancistrachne* should be broadened to encompass the specific variation found in *A. maidenii*. Vickery (1961a) considered that *A. maidenii* had some resemblance to *Entolasia*,

Cleistochloa and Dimorphochloa whereas Simon and Alfonso (http://ausgrass2.myspecies.info/, accessed 21 September 2019) regarded A. maidenii to be of 'questionable taxonomic status' indicating that in Simon's consideration of the morphology was inconsistent with the diagnosis of Ancistrachne by Blake (1941).

The study by Thompson and Fabillo (2021) provided a set of morphological characters that enabled recognition of putative taxa at several ranks and they examined the consistency of their results by analysing multiple datasets. Since their work, research on the morphology of 'the cleistogamy group' has been continued by the first author, resulting in expansion of the character set. In the present work, an updated dataset was statistically analysed with the aim of exploring the morphological similarities and dissimilarities of the taxa within 'the cleistogamy group' and among its allies (Ancistrachne s.s., Entolasia, Neurachne, Panicum and Thyridolepis). The consistency of results from analyses using different cluster algorithms and datasets, varying in composition of morphological characters and samples, were tested against a baseline. On the basis of the congruency of findings from this study and Thompson and Fabillo (2021), 'the cleistogamy group' and Ancistrachne maidenii were considered to have definitive taxonomic morphology and, as a consequence, a new subtribe and new genus respectively, are diagnosed herein.

#### Materials and methods

#### Taxon sampling

The baseline sample set was composed of 24 putative species modified from Thompson and Fabillo (2021) by (1) the addition of *Dimorphochloa* sp. (Mt Cooper R.J. Cumming 18623), and (2) the use of *Panicum s.s.* as the sole outgroup to which was added *Panicum mitchellii* Benth. and *Panicum queenslandicum* Domin (to provide morphological variation present in Australian species), and *Walwhalleya subxerophylla* (Domin) K.E.Wills & J.J.Bruhl. was removed (Appendix 1).

An image of the type specimen of *Ancistrachne uncinulata* was obtained from the Natural History Museum, London, and examined. For detailed examination of spikelet morphology, eight specimens from the Queensland Herbarium (BRI) were selected as topotypes for *A. uncinulata* (Appendix 2). Selection of topotypes was based on the location of the type collection of *A. uncinulata* from Keppel Bay, Queensland.

Ancistrachne ancylotricha was not sampled owing to restrictions placed on destructive sampling of the borrowed specimens. A. ancylotricha is known only from three collections and is presumed extinct.

#### Nomenclature and terminology

Botanical nomenclature follows Brown and Bostock (http://data.qld.gov.au/dataset/census-of-the-queensland-flora-2020/, accessed 15 May 2021).

General botanical terminology follows Harris and Harris (1994) and Beentje (2010). Terminology relating to inflorescences and spikelet morphology follows Tothill and Hacker (1983), Jacobs *et al.* (2008), Gibson (2009) and Thompson (2021). The spikelet is viewed here as a reduced inflorescence where florets are enveloped by bracts (Kellogg 2006; Endress 2010). Terminology relating to grass anatomy and micromorphology follows Ellis (1976, 1979) and Dengler *et al.* (1994).

# Acquisition of data and classification of morphological characters

Characters scored for this study include macromorphological, anatomical and micromorphological characters of inflorescences, spikelets, leaves and culms (Table 3). Information was gathered from fresh material from cultivated and field plants, herbarium specimens and images. Character groups classified include the following:

- · inflorescence structure
- spikelet bracts (glumes, lemmas, paleas), stigmas, anthers, lodicules and caryopses
- · leaf shape, transverse section, abaxial surface and ligules
- · culm transverse section and surface.

Plants were cultivated in pots under nursery conditions in semi-shade at a latitude of 27.5°S, in Brisbane, Australia, to provide fresh material of leaves, culms and spikelets. For *A. maidenii*, plants were propagated from portions of the root stock of *ex situ* plants, and for *A. numaeensis*, plants were established from caryopses obtained from New Caledonia. Plants of *A. uncinulata* were established from *ex situ* plants as well as propagated from caryopses taken from several herbarium specimens to examine potential variation over its broad latitudinal range (12.6–32.5°S).

Fresh material was used for free-hand transverse sections of leaves and inflorescence culms. Sections were prepared following the free-hand sectioning method used by Thompson (2017), modified from the method described by Frohlich (1984).

Leaf-surface micromorphology was examined from replicas of the abaxial surface of fresh leaves obtained using the 'impression method' described by Hilu and Randall (1984).

Leaf and culm anatomy and micromorphology including stomata, silica bodies and micro-hairs were classified and recorded following classifications used by other authors, including De Wet (1960), Metcalfe (1960), Twiss *et al.* (1969), Carolyn and Jacobs (1973), Ellis (1979), Renvoize (1987), Watson and Dallwitz (1992), Siqueiros-Delgado and Herrera-Arrieta (1996), Piperno and Pearsall (1998), Krishnan *et al.* (2000), Siqueiros-Delgado (2007), Lu *et al.* (2009) and Jattisha and Sabu (2015) (Table 3, 4, Appendix 1).

Silica bodies, stomata, epidermal long cell walls, microhairs and macrohairs on lower lemmas and upper lemmas and paleas were classified following other authors (Hsu 1965; Jirasek and Jozifova 1968; Ellis 1979; Valdes-Reyna and Hatch 1991; Snow 1996; Acedo and Llamas 2001; Liu *et al.* 2010; Mashau *et al.* 2015; Olonova *et al.* 2016; Neumann *et al.* 2017). Lodicules on fresh material were classified as plicate or non-plicate (Hsu 1965; Jirasek and Jozifova 1968; Guedes and Dupuy 1976).

Stigma characters included the position of emergence from the spikelet, overall shape and colour, and characteristics of the lobes such as shape of apex, relative length and tilt (Thiele *et al.* 1996; Table 3).

Caryopsis morphology was scored for characters relating to hilum, scutellum, spermaderm and stylopodium using characters and states by other authors (Kennedy 1899; Reeder 1957; Brown 1959, 1960; Watson and Dallwitz 1992; Klak 1994; Kosina 1995; Snow 1998; Liu *et al.* 2005, 2015; Table 3).

Classification of inflorescence types, ligules and leaf bases follows Jacobs *et al.* (2008), Tothill and Hacker (1983), and Webster (1987).

#### **Imagery**

Imagery was used as the major source of information, especially in the studies of anatomy and micromorphology and to illustrate differences across the taxa. Images were taken using light and scanning electron microscopy. Photographs for anatomy and micromorphology were taken using two light microscopes (Nikon SMZ25 binocular microscope with Nikon DS-Ri1 camera and images were viewed using imaging software NIS-Elements BR (ver. BR 5.11.000 64-bit, Laboratory Imaging, USA, see http://www.lim.cz, accessed 15 December 2019); Leica DMLB compound binocular microscope with an industrial digital camera and images were viewed using imaging software ToupView (ver. x64 4.7.14326.20190401, Touptek, China, see http://www. touptek.com, accessed 20 September 2019). Scanning electron micrographs (SEMs) were obtained using a Phenom G2 5kev SEM with backscatter detector, without sputter coating samples.

#### Data sets and phenetic analyses

Variations in inputs to analyses included algorithm (ordination, classification, association measure), sample composition in terms of number of samples and replications, and the format of characters following findings made by other authors (Clifford and Goodall 1967; 't Mannetje 1967; Clifford and Williams 1973; Austin and Belbin 1982; Hilu and Wright 1982; Johnson 1982; Stevens 1991; Thiele 1993; Wills *et al.* 2000; Scotland *et al.* 2003; Wortley *et al.* 2005; Pereira *et al.* 2007; Newmaster *et al.* 2008; Zuloaga *et al.* 2014; Peichoto *et al.* 2015; Aliscioni 2016; Table 5). Datasets were established from the baseline dataset composed of 24 samples and 161 morphological characters, modified from the data matrix and character list by

#### Table 3. Morphological characters, states and codes based on Thompson and Fabillo (2021).

#### I. Vegetative - growth habit and culms

- 1. Growth habit: decumbent (0), caespitose (1), rhizomotous (2), stoloniferous (3) [009]
- 2. Culm internode indumentum: glabrous (0), woolly (1) [102]
- 3.<sup>A</sup> Culm nodes pubescent: no (0), yes (1), yes or no (2 [124]
- 4. Long straight white tubercle-based hairs perpendicular on leaf sheaths and usually on the proximal margins of leaves: absent (0), present (1) [072]
- 5.<sup>A</sup> Presence of contraligule: no (0), yes (1) [085]

#### II. Chasmogamous (CH) inflorescences (terminal on culms)

- 6. Terminal inflorescence type: panicle (0), raceme, spikelets ascending and overlapping (1), raceme, spikelets appressed and barely touching (2), spike-like panicle (branches much reduced with spikelets and branches appressed) (3) [001]
- 7. Racemes, including racemose branches of panicles, with spikelets distichous: no (0), yes (1) [070]
- 8. Pulvinii in the inflorescence branch axes: no (0), yes (1) [093]
- 9. Shape of pedicel apex: shallow cup with thin walls (0), wine glass shaped with thick walls (1), broad saucer to truncate (2), shallow bowl (3) [087]
- 10.<sup>A</sup> Inflorescences a mixture of sessile spikelets (terminal on short branches with a pulvinus at the base) and shortly pedicellate ones: no (0), yes (1) [152]

#### III. Macromorphology of CH spikelets

- 11. Spikelet tilted: no (0), yes (1) [151]
- 12. Spikelet callus angle: transverse (0), oblique (1) [155]
- 13. Spikelet orientation: adaxial (0), abaxial (1) [094]
- 14. Spikelet disarticulation: below glumes (0), above glumes (1) [139]
- 15. Spikelet compression: none to minimal (0), dorsi-ventral (1), strongly dorsi-ventral (2) [019]
- 16. Hairy callus, a ring of hairs at the base of the spikelet: absent (0), present (1) [118]
- 17. Racemes composed of heteromorphic spikelets increasing in size apically: no (0), yes (1) [146]
- 18.<sup>B</sup> Lower glume development: absent (0), vestigial to small (<~1/10 of spikelet length) (1), >~1/5 spikelet length (2) [061]
- 19. Relative length of lower glume to upper glume: different (0), approximately equal (1) [119]
- 20. Lower glume with a window: absent (0), present (1) [150]
- 21. Lower glume enveloping the upper glume at the base: no (0), yes (1) [147]
- 22. Shape of apex of upper glume: obtuse (0), acute (1), attenuate (2), elongated with blunt tip (3) [008]
- 23. A Shape of body of upper glume: elliptical (0), ovate (1), spatulate (2) [007]
- 24. Texture of the margins of the upper glume: same as body (margin usually appearing as inrolled) (0), tapering (1) [084]
- 25. Submargin of upper glume with distinctive trichomes: absent (0), long cilia (1) tuberculate-based bristles (2) [111]
- 26. Upper glume with 2-keeled margins: absent (0), present (1) [123]
- 27. Upper glume prominently ribbed on the back: no (0), yes (1) [135]
- 28. Upper glume with a small proximal elaiosome: no (0), yes (1) [134]
- 29. Relative length of the upper glume to the lower lemma: sub-equal to equal (0), shorter (1), longer (2) [054]
- 30. Relative similarity (shape, size, texture, indumentum, chlorophyllous) of upper glume and lower lemma: similar (0), dissimilar (1) [116]
- 31. Similarity of the indumentum (type and distribution) on the upper glume to that on the lower lemma: similar or absent (0), dissimilar (1), very dissimilar (2) [014]
- 32. Shape of apex of lower lemma: truncate (0), obtuse (1), broadly acute (2), narrowly acute (3) [003]
- 33. A Shape of body of lower lemma: elliptical (0), ovate (1), spatulate (2) [088]
- 34. Texture of margin of lower lemma: normal (same as body) (0), tapering (1), hyaline at least proximally (2) [020]
- 35. Type of tuberculate-based macrohairs on the back of the lower lemma: macrohairs absent (0), short and straight (1), long and straight (2), short and crinkled, (3) uncinate (4) [002]
- 36. Length of strap-shaped trichomes on distal margin of lower lemma: absent (0), short, <99 µm (1), long, >100 µm [015]

#### Table 3. (Continued)

#### III. Macromorphology of CH spikelets

- 37. Back of lower lemma with epicuticular wax: absent (0), film (1), platelets (2) [132]
- 38. Lower lemma with 2-keeled margins: absent (0), present (1) [143]
- 39. Lower lemma with a proximal elaiosome (oily residue present on dry specimens): absent (0), small and relatively inconspicuous (1), conspicuous (2) [129]
- 40. Lower lemma saccate: absent (0), pouched (shallow to very conspicuous) (1) [142]
- 41. Back of lower lemma channelled: no (0), yes (1) [154]
- 42.<sup>B</sup> Lower palea: absent (0), present (1) [083]
- 43. Lower palea thinner than the lower lemma: no (0), yes (1) [149]
- 44. Lower palea minutely ciliate on margin at apex: palea absent (0), no (1), yes (2) [057]
- 45. Lower floret male: no (0), yes (1) [117]
- 46. Lower floret anthers larger than upper floret ones: absent (0), no (1), yes (2) [110]
- 47.<sup>A</sup> Relative position of emergence of stigmas from upper floret: half way (0), 2/3-3/4 (1), apex (2) [069]
- 48.<sup>A</sup> Outline shape of stigma: relative squat (0), elongated (1) [138]
- 49. Colour of stigma: clear or white (0), pink or purple (1) [131]
- 50.<sup>A</sup> Number styles: two (0), three, third reduced to a point or ridge (1) [141]
- 51. Apex of upper lemma: muticous (0), mucronate (1), awned (a bristle like structure greater than 0.5 mm long) (2) [010]
- 52. A Shape of body of upper lemma: elliptical (0), ovate (1), spatulate (2) [097]
- 53. Texture of body of upper lemma relative to lower lemma: less firm; hyaline, loosely covering caryopsis (0), less firm; membranous, loosely covering caryopsis (1), approximately equal to slightly firmer; chartaceous to cartilaginous, slightly hardened papery texture, loosely overlapping caryopsis (2), much harder; thin, indurated, pliable but brittle, glossy, strongly clasping caryopsis (3) [076]
- 54. Relative length of the upper lemma to the lower lemma: subequal to equal (0), shorter (1), much shorter (2) [090]
- 55. Upper lemma with a germination lid: absent (0), inconspicuous (1), conspicuous crescent-shaped depression between a raised portion of the two veins (2) [006]
- 56. Rachillar stipe below upper floret: absent (0), present (1) [121]
- 57. Prickles or hooks on the back of the upper lemma: no (0), yes (1) [168]
- 58.<sup>B</sup> Texture of margin of upper lemma: same as body (0), tapering; thinner than body (1) [126]
- 59. Macrohairs on submargin of upper lemma: absent (0), present (1) [156]
- 60. Length of trap-shaped trichomes on distal margin of upper lemma: absent (0), <99 μm (1), >100 μm long (2) [042]
- 61. Macrohairs on the back of upper lemma and palea: absent (0), appressed pilose with flattened multicellular hairs (1) [133]
- 62.<sup>A</sup> Microhairs on the back of the upper palea: absent (0), present (1) [167]
- 63. Shape of apex of upper palea: arc (0), obtuse (1), truncate (2), triangle (3) [080]
- 64. Margins of upper palea overlapping at apex: no (0), yes (1) [153]
- 65. A Cross-sectional shape of CH caryopses: slightly compressed, x-section broadly elliptical (0), moderately dorsi-ventrally compressed, shallowly convex on both sides (1), strongly compressed, plano-convex (2), surface more or less flat on both sides (3) [041]
- 66. Longitudinal outline of CH caryopses: ovate (0), elliptical (1), obovate (2) [071]
- 67. Relative length of scutellum to total length of caryopsis: <half (0), >half (1) [121]
- 68. A Shape of scutellum: broadly elliptical (0), elliptical (1) [077]
- 69. A Shape of hilum: broadly elliptic (0), elliptic (1), obovate (2) [021]
- 70. A Relative length of hilum (length ratio of hilum: caryopsis): short, (<0.18) (0), medium (0.19-0.29) (1), long (>0.30) (2) [120]
- 71. Surface pattern of pericarp: smooth (0), finely longitudinally striate (1), coarsely longitudinally striate (2), rugulose (3) [068]
- 72.<sup>A</sup> Shape of embryonic axis of CH caryopsis from dorsal view: spatulate (0), oblong (1) [065]
- 73. A Stylopodium: absent (0), narrow fusion (1), broad fusion (2) [055]

#### Table 3. (Continued)

#### III. Macromorphology of CH spikelets

- 74. Length of hairs on distal margin of upper palea: absent (0), <99 µm (1), long >100 µm long (2) [005]
- 75. Cross-sectional shape of upper palea near the apex: flat to convex (0), sharply creased (1) [106]
- 76.<sup>A</sup> Lodicule type: non-plicate (0), plicate (1) [112]
- 77. A Lodicule symmetry of plicate type: not applicable (0), relatively symmetrical (1), asymmetrical (2) [082]
- 78. Type of cleistogamy present in upper floret of terminal inflorescence: absent (0), monomorphic anthers (1), dimorphic anthers (2) [108]

#### IV. Cleistogamous (CL) inflorescences (presence of obligately cleistogamous spikelets in a second type of inflorescence, mostly axillary)

- 79. Composition of the CL inflorescences: absent (0), mostly a single axillary spikelet (1), usually paired (2), I (-4) spikelets terminal on short leafy branches on differentiated culms (3), short axillary raceme with up to ~4 spikelets (4) [036]
- 80. Nature of the leaf sheath at maturity subtending the CL spikelets: absent (0), revolute (1), gaping (2) [022]
- 81. CL culm nodes: not disarticulating (0), disarticulating at maturity (1) [161]

#### V. Macromorphology of CL spikelets

- 82. Compression of CL spikelets, cross-sectional shape: spikelets absent (0), ±circular (1), elliptical (2), concavo-convexo (3) [041]
- 83. Orientation of CL spikelets: spikelets absent (0), adaxial (1), abaxial (2) [027]
- 84. Pedicels of basal CL spikelets: absent (0), pedicellate only (1), sessile only (2), both sessile and pedicellate (3) [025]
- 85. Relative similarity of shape and size of CH and CL spikelets: absent (0), relatively similar (1), very dissimilar (2) [049]
- 86. Presence of lower glume: spikelets absent (0), absent (1), small to vestigial (2) [062]
- 87. Shape of upper half to third of upper glume: disc (0), obtuse (1), truncate (2), acute (3) [043]
- 88. Upper glume margins at apex: spikelets absent (0), flat (1), clasping (2) [044]
- 89. Texture of margin of upper glume at apex: spikelets absent (0), same as body (1), tapering to membranous (2) [026]
- 90. Back of upper glume 2-keeled: spikelets absent (0), no (1), yes (2) [022]
- 91. Elaiosome at base of upper glume: spikelets absent (0), absent (1), present (2) [064]
- 92. Similarity of the CH and CL upper glumes: spikelets absent (0), similar (1), dissimilar (2) [045]
- 93. Similarity of the texture of the lower lemma to the upper glume: spikelets absent (0), similar (1), dissimilar (2) [047]
- 94. Relative length of the upper glume to the lower lemma: spikelets absent (0), shorter (1), equal (2) [050]
- 95. Similarity of the shape of the upper glume to the lower lemma: spikelets absent (0), similar (1), dissimilar (2) [052]
- 96. Margins of lower lemma at apex: spikelets absent (0), not overlapping (1), overlapping (2) [030]
- 97. Margin of the lower lemma with texture different from the body: spikelets absent (0), relatively narrow width of taper (1), broad width of taper graduating to hyaline (2) [037]
- 98. Hook microhairs on the back of the lower lemma: spikelets absent (0), no (1), yes (2) [058]
- 99. Macrohairs on the back of the lower lemma: spikelets absent (0), absent (1), tuberculate-based prickles (2) [059]
- 100. Texture of the body of the lower lemma: spikelets absent (0), chartaceous (1), woody (2) [048]
- 101. Lower lemma with a proximal elaiosome: spikelets absent (0), absent (1), basal crescent shaped without oily residue when dry (2), basal with oily residue when dry (3), covering most of body with oily residue when dry (4), covering most of body without oily residue when dry (5) [017]
- 102. Back of lower lemma channelled: spikelets absent (0), no (1), yes (2) [028]
- 103. Back of upper lemma ribbed: spikelets absent (0), no (1), yes (2) [053]
- 104. Similarity of the shape of the CH and CL lower lemmas: spikelets absent (0), similar (1), dissimilar (2) [046]
- 105. Apex of the upper lemma: spikelet absent (0), mucronate (1), awned (2) [024]
- 106. Relative length of upper lemma to lower lemmas: spikelets absent (0), shorter (1), equal (2) [021]
- 107. Submargin of upper lemma pilose: spikelets absent (0), no (1), yes (2) [051]
- 108. Shape of apex of upper lemma: spikelets absent (0), absent (1), apex obtuse to truncate (2), apex acute (3) [018]
- 109. Short-barbed hooks on back of upper lemma: spikelets absent (0), absent (1), present (2) [063]

#### Table 3. (Continued)

#### V. Macromorphology of CL spikelets

- 110. Surface of back of upper lemma with minute longitudinal ridges: spikelets absent (0), absent (1), present (2) [023]
- 111. Upper lemma with a germination lid: spikelet absent (0), indistinct (1), distinct (2) [038]
- 112. Distal margin of upper lemma ciliate with strap-shaped trichomes: spikelet absent (0), absent (1), present (2) [060]
- 113. Shape of apex of upper palea: absent (0), triangle (1), narrow triangle (2) [027]
- 114. Cross-sectional shape of CL caryopses: not applicable (0), slightly compressed, x-section broadly elliptical to circular (1), moderately dorsi-ventrally compressed, shallowly convex on both sides (2) [024]
- 115.<sup>A</sup> Longitudinal shape of CL caryopsis: not applicable (0), ovoid (1), elliptical (2), linear (3) [029]

#### VI. Leaf micromorphology and anatomy

- 116.<sup>B</sup> Photosynthesis pathway: C3 (0), C4 (1) [127]
- 117. Sheath of primary vascular bundles of leaves: one ring of cells (0), two rings of cells (1) [159]
- 118. A Kranz type sheath of primary vascular bundles of leaves: absent (0), one ring (1) [128]
- 119. Sheath of primary vascular bundles of leaves with inner ring: absent (0), complete with thin-walled cells (1), an adaxial arc of thin-walled cells (2) complete with thick-walled cells (3) [130]
- 120. Configuration of outer parenchyma sheath of primary vascular bundles of leaves: a ring (0), discontinuous abaxially (1), discontinuous adaxially and abaxially (2) [109]
- 121. Adaxial sclerenchyma of primary vascular bundles: girder (0), strand (1) [145]
- 122. Abaxial sclerenchyma of primary vascular bundles: girder (0), strand (1) [160]
- 123. A Leaf and culm stomata: similar (0), different (inflated) (1) [148]
- 124. Shape of subsidiary cells of stomata on abaxial leaf surface: parallel sided, narrow (0), parallel sided, broad (1), low triangular (2) [081]
- 125. Shape of predominant silica bodies on abaxial leaf surface in costal zone: crosses (circular to elliptical with or without slits) (0), bilobates (dumbbell-shaped, constricted at centre, concave or convex at ends) (1) [113]
- 126.<sup>A</sup> Polylobate silica bodies on abaxial leaf surface: absent or uncommon (0), common (1) [114]
- 127. Two types of silica bodies on surface in costal zone on abaxial leaf: no (0), yes (1) [067]
- 128. A Shape of anticlinal walls of long cells on abaxial leaf surface:  $\Omega$ -shape (0), U-shaped (1) [125]
- 129. Shape of intercostal long cells on abaxial leaf surface: long and narrow, <1:5 (0), short and broad >1:7 (1) [115]

#### VII. Culm anatomy and micromorphology of surface

- 130. Chlorenchyma shape from x-section of culm: rectangular blocks 2–4 cells wide (0), continuous or discontinuous strips 2 or 3 cells wide (1), C<sub>4</sub> type with two small areas, ±connected, either side of a tertiary vascular bundle (2) [098]
- 131.<sup>A</sup> Separation of chlorenchyma and tertiary vascular bundles from x-section of culm: touching (0), by clear cells (1), by sclerenchyma (2) [101]
- 132. Shape of chlorenchyma cells from x-section of culm: all more or less circular (0), radially oblong on outer layer, inner layers more or less circular to slightly oblong (1), all radially oblong, outer layer more elongated (2) [091]
- 133. A Peripheral sclerenchyma from x-section of culm: radial (0), tangential (1) [095]
- 134. Shape of radial sclerenchyma from x-section of culm: absent (0), linear (1), rectangular (2), triangular (3) [004]
- 135. Peripheral sclerenchyma separated from adjacent tertiary vascular bundle from x-section of culm: no (0), yes, by a row of clear cells (1), yes, by chlorenchyma cells (2) [013]
- 136. Inner sclerenchyma from x-section of culm: absent to uncommon (0), discontinuous bands (1), continuous ring (2) [011]
- 137. Sheath of primary vascular bundles from x-section of culm with xylem separated from parenchyma by: a ring of thick-walled cells (0), an arc of parenchyma (1), a ring of parenchyma (2) [030]
- 138. Vascular bundles from x-section of culm: secondary and primary (0), tertiary, secondary and primary (1) [086]
- 139.<sup>A</sup> Silica bodies on surface of culms: absent or very sparse (0), present (1) [066]
- 140.<sup>A</sup> Presence of bicellular microhairs on surface of culms: absent or very sparse (0), present (1) [078]

#### Table 3. (Continued)

#### VIII. Spikelet micromorphology (CH unless otherwise indicated)

- 141. Distribution of papillae on the back of the upper lemma and palea: absent (0), all over (1), at the apex only (2) [144]
- 142. Arrangement of papillae on the back of the upper lemma and palea: papillae absent (0), not in distinct rows and unevenly spaced (1), in distinct longitudinal rows and regularly spaced (2) [016]
- 143. Position of the papillae on the back of the upper lemma and palea: ridges or papillae absent (0), in the furrows (1), along the ridges (2) [103]
- 144. Surface of the back of the upper lemma and palea with minute longitudinal ridges: absent (0), ridges narrower than furrows (1), ridges broader than or equal to furrows (2) [079]
- 145. Surface of the back of the upper lemma and palea on axillary CL spikelets: axillary CL spikelets absent (0), similar to CH spikelets (1), different, with minute ridges present (2) [043]
- 146. Type of simple papillae over the back of the upper lemma and palea: absent (0), nipple-shaped (1), volcano-shaped (2), beak-like apex (3), Neurachninae type (4) [096]
- 147. Compound papillae on upper palea: absent (0), mostly 2 or 3 prongs (1), mostly 4 or more prongs (2) [137]
- 148. Compound papillae on the back of the upper palea: absent (0), nipple-shaped (1) volcano-shaped (2) [140]
- 149. Type of bicellular microhairs on the back of the upper palea: absent (0); type 1: distal cell much larger than proximal, apex round (1); type 2: equal, elongated distal cell with acute apex (2) [100]
- 150. A Shape of anticlinal walls of long cells on upper lemma and palea: Ω-shape (0), U-shaped (1), elongated U-shaped (2) [056]
- 151.<sup>A</sup> Silica bodies present on apex of upper palea: no (0), yes (1) [107]
- 152. Microhairs near apex of upper palea: glabrous (0), needle-like microhairs to ~60 μm long (1) [105]
- 153. A Microhairs near apex of upper palea: glabrous (0), flattened type (1) [075]
- 154.<sup>A</sup> Stomata at apex of upper palea: absent (0), present (1) [104]
- 155. Shape of silica bodies on lower lemma: crosses (more or less circular outline) (0), bilobate (dumb-bell shaped, constricted at centre, indented at ends) (1), saddles (circular to elliptical with or without slits) (2) [089]
- 156. Bicellular microhairs: type 1: distal cell much larger than proximal, apex round (0); type 2: equal, elongated distal cell with acute apex (1) [073]
- 157. A Shape of anticlinal walls of long cells on lower lemma: Ω-shape (0), U-shaped (1) [092]
- 158. Lower lemma with dense covering of more or less paired papillae in longitudinal rows: no (0), yes (1) [136]
- 159. Shape of apex of stigma lobes: obtuse or truncate (0), broad rounded (1), narrow rounded (2) [099]
- 160.<sup>A</sup> Tilt of apex of stigma lobes (apical 4-5): more or less appressed (0), ascending (1) [074]
- 161.<sup>A</sup> Relative length of stigma lobes (apical 4–5): vestigial (0), short (1), long (2) [012]

Characters used by other authors are shown as: Anot used by Thompson and Fabillo (2021), Bnot used by Morrone et al. (2012) for taxa that overlap with this study. Some of these characters have been modified in the list. Order of most discriminating characters is shown in brackets after each character. Order in terms of highest to lowest Kruskal–Wallis values was generated from PATN analysis of the baseline dataset composed of 24 samples and 161 morphological characters, using Gower association measure at the 10-group level.

Thompson and Fabillo (2021). The characters comprised 5 vegetative characters, 73 characters relating to CH spikelets and inflorescences, 38 characters relating to axillary CL spikelets, and 45 relating to leaf anatomy (Table 3, Appendix 1). The dataset consisted of 69 binary and 92 multistate characters. Characters and character states were evaluated in terms of homology, ambiguities, reliability, plasticity, practicality, repeatability and standardisation of assessment or measurement using considerations by other authors (Hillis 1987; Wagner 1989; Smith 1990; Lipscomb 1992; Hillis and Wiens 2000; Poe and Wiens 2000; Scotland *et al.* 2003; Wiens 2004; Smith and Turner 2005; Thompson and Fabillo 2021).

Phenetic analyses were conducted using PATN (ver. 4.00, Australia; Blatant Fabrications, see <a href="http://patn.org">http://patn.org</a>, accessed 20 February 2021). Cluster analyses used the unweighted

pair method using arithmetic mean (UPGMA), and dendrograms were generated using agglomerative hierarchical fusion with unweighted pair group method with Beta value of -0.1. Classifications involved two association measures, Gower metric and Czekanowski (Somerfield 2008; Bray and Curtis), where the differences between states of polymorphic characters are considered equal and unequal respectively (L. Belbin, pers. comm.). Three-dimensional ordination plots were generated using semi-strong hybrid multi-dimensional scaling (SSH). Ordination stress value (OSV) was used as the measure of closeness of fit: stress values of <0.05= excellent, <0.1= good, <0.1-0.15 may be OK, <0.2= not good (see https://patn.org/). Discriminating characters were generated for each analysis from Kruskal–Wallis (KW) values at two group levels, namely,

Table 4. Summary of differences in micromorphology and anatomy of Simonachne maidenii and Ancistrachne uncinulata.

Character		Simonachne maidenii	Ancistrachne uncinulata		
Upper lemma of	Surface texture	Minutely longitudinally striate	Smooth		
terminal spikelet	Papillae	Absent	Truncate apex type, in regularly to irregular spaced in longitudinal rows		
	Anticlinal walls of long cells	Elongated U-shape	U-shape		
Stigma lobes		Appressed	Ascending		
Lower lemma of	Stomata	Subsidiary cells triangular	Absent		
terminal spikelet	Silica bodies	Bilobate and polylobate, dumb bell type, I I–20 $\mu m$ long, approximately as wide as long, narrow waste and indented ends	Bilobate, dumb bell type, 9–15 $\mu$ m, approximately as wide as long, broad waste and rounded ends; similar to those on culms		
	Microhairs	Bicellular only, 50–70 μm long	Bicellular, 40–50 $\mu$ m long, proximal cell 40% of length of distal; densely covered in prickles		
	Epicuticular wax	Film	Platelets		
Leaf x-section	'		Two rings; inner incomplete, a few adaxial thin- walled cells, several thick-walled cells and interrupted abaxially by sclerenchyma; outer ring of clear thin-walled cells largest in the centre, interrupted abaxially by several thick-walled cells		
Bundle sheath of tertiary veins		A single ring of thin-walled clear cells; cells relatively even sized mostly as large as the largest cells in the midvein bundles	Double ring; inner composed of small thin-walled cells; outer, adaxial half thin-walled clear cells and abaxial half thick-walled cells		
	Dorsal sclerenchyma	I or 2 cells deep × 4 or 5 cells wide	2 or 3 cells deep × 5–8 cells wide		
	Chlorenchyma	Radiate adjacent to outer bundle sheath; palisade adjacent to bulliform cells	Not or indistinctly radiate adjacent to bundle sheath; radiating adjacent to bulliform cells		
Fertile culm Peripheral x-section sclerenchyma		Tangential, <10 cells wide × 1–3 cells deep; strand	Radiate, 3 or 4 cells wide × ~8 cells deep; girder		
	Chlorenchyma	~8 cells wide × 2 or 3 cells deep; fringed by clear parenchyma on the inner margin; ring of sclerenchyma absent	~8 cells wide × 4–7 cells deep; usually fringed on the inner margin by a band of sclerenchyma one to several cells wide and pale green parenchyma		
	Vascular bundles (vbs)	Three sizes of vbs, tertiary, secondary and primary; circular; all vbs encircled by a ring of clear thinwalled cells and with an inner ring of sclerenchyma; vbs separated by thin-walled clear cells; secondary vbs adjacent to chlorenchyma	Two sizes of vbs, tertiary and primary; circular; vbs surrounded by sclerenchyma		
Abaxial leaf surface	Stomata	40–42 μm long, subsidiary cells low triangular	$1832\mu m$ long, subsidiary cells parallel sided with rounded ends		
	Silica bodies	$\sim\!\!50\%$ trilobed dumbbell type; constricted at centre, indented at ends	Dumbbell type; wide central area, rounded ends		
	Intercostal long cells	<130 µm long	<70 μm long		
	Anticlinal walls of costal long cells	$\Omega$ -shape	U-shape		
Fertile culm surface	Stomata	Subsidiary cells low triangular, guard cells visible	Ends of subsidiary cells nodular, guard cells recessed, often obscure; different from those on leaves		
	Silica bodies	Absent or sparse	Abundant; saddle-shaped, $8-14\mu m$ long, wide central area, rounded at ends; different from those on leaves		
	Indumentum	Scabrid with spicules and muriculate with prickles	Sparsely muriculate with hooks		

**Table 5.** Summary of variations in inputs to phenetic analyses including datasets (composition of samples and morphological characters) and algorithms (classification, ordination, association measure) used to test the effect on results.

Analysis	Number of	Dataset	Association	Analysis
	samples	Number of characters and format	measure	
Baseline	24	161, binary and multistate	Czekanowski, Gower	Classification, ordination
Test I	10, 30	161, binary and multistate	Gower	Classification, ordination
Test 2	10, 24	Of the 161-baseline character set, 92 are multistate which were converted to binary creating 365 binary	Gower	Classification
Test 3	21	102, from Thompson and Fabillo (2021). Binary and multistate.	Gower	Classification
Test 4	24	Fifteen datasets composed of batches of characters compiled from a list of most discriminating characters in a descending order based on Kruskal–Wallis values (Test 4). Datasets were created by successively removing batches of data for the next 10 least discriminating characters starting from 150 characters, and the next 150, 140, 130 and so on. Binary and multistate.	Czekanowski, Gower	Classification
Test 5	10, 24	Fifteen from Morrone et al. (2012) that apply to taxa in this study. Binary and multistate.	Czekanowski, Gower	Classification

The baseline dataset consists of 24 samples, representing 24 putative species, and 161 morphological characters.

10-group level and three-group level respectively. The 10-group level corresponds to the 10 putative genera (Ancistrachne s.s., Ancistrachne maidenii, Calyptochloa, Cleistochloa s.s., Dimorphochloa rigida, Dimorphochloa spp., Entolasia, Neurachne, Panicum s.s., and Thyridolepis) and the three-group level was represented by the three main clusters identified by Thompson and Fabillo (2021), i.e. 'the cleistogamy group', Neurachninae s.s. and the 'paniculate inflorescence group'.

Twenty-two variations in datasets for five types of tests were analysed to generate topologies for comparison with those from analysis of the baseline dataset (Table 5). The variations in datasets were as follows:

- Test 1. Variation in sample size using 10 and 30 samples with the 161-character set. The sample of 10 was represented by a single species for each of the 10 putative genera and the 30 samples included replicates, so all genera have three samples.
- Test 2. The 92 multistate characters expressed as binary, resulting in 365 characters
- Test 3. Character set consisted of 102 characters and 21 samples from Thompson and Fabillo (2021).
- Test 4. Fifteen datasets were established using a list of the most discriminating characters in a descending order, on the basis of KW values generated from analysis of the baseline dataset with Gower association measure and number of groups set at 10. Datasets were created by successively removing batches of 10 characters with the lowest KW values, starting from 150, followed by 140, 130, and so on (Table 6).
- Test 5. An example of *a priori* character selection. Two tests were conducted, using the characters that overlap

with Morrone *et al.* (2012). Of the 57 morphological characters used by Morrone *et al.* (2012), 15 apply to the seven genera that overlap with this study (Table 3). Two datasets were established using the 10 and 24 sample sets. The samples used in these analyses differ from Morrone *et al.* (2012) by the substitutions, *Neurachne munroi* for *Neurachne alopecuroidea* R.Br. and non-native species of *Panicum s.s.* with an Australian species. Analyses were also run using characters expressed as binary.

#### Results

The three major clusters, viz. 'the cleistogamy group', the 'paniculate inflorescence group' and subtribe Neurachninae s.s. each with distinctive morphology, were consistently recovered from the cluster analyses. The topologies generated from the analyses showed that Ancistrachne s.l., Calyptochloa, Cleistochloa s.l. and Dimorphochloa s.l. are not monophyletic and Ancistrachne s.s. and A. maidenii are placed in separate clusters, viz. the 'paniculate inflorescence group' and 'the cleistogamy group' respectively (Table 5, Fig. 5, 6, Appendices 3-5). Ancistrachne maidenii and A. uncinulata differ in macro- and micromorphological and anatomical characters relating to upper and lower lemmas, surface of leaves and culms, stigmas, caryopses and leaf and culm crosssections (Tables 1, 4, Fig. 4, Appendices 6-19). An unusual feature of A. uncinulata is the distinctly different stomata on the leaves and culm surfaces (Table 4, Appendices 14, 18).

The topologies generated from cluster analyses in this study showed differences from that for the baseline, being affected by the algorithm (ordination, classification and association measure) used and the dataset with respect to

Table 6. Summary of results from cluster analyses.

Variable	Description of variation	Figure	Remarks
Algorithm	Classification analysis using baseline dataset of 24 samples and 161 characters using two assocaition measures, Czekanowski and Gower	Fig. 5	Topologies using the two association measures are congruent.
	Ordination using dataset of 24 samples and 161 characters with Gower association measure	Fig. 6	Relationships of clusters compatible with those from the classification analysis for the same dataset.
Sample set composition	Datasets with 10, 24 and 30 samples dataset and 161 characters and classification analysis using Gower association measure (Test 1)	Fig. 5, Appendix 3	All topologies agree with results from analysis of baseline dataset.
Character format	All characters in binary format comprising dataset of 365 characters and 24 samples and classification analysis using Gower association measure (Test 2)	Fig. 5	Topology congruent with that for the baseline dataset.
Character set composition	102 characters from Thompson and Fabillo (2021) and classification analysis using Gower association measure (Test 3)	Fig. 5	Topology shows the same relationships of taxa as for baseline dataset. Taxon relationships congruent with cladogram by Thompson and Fabillo (2021).
	Fifteen datasets compiled from the most discriminating characters and classification analyses using Gower association measure (Test 4)	Appendix 4	For the top 30 most discriminating characters, the three major clusters were the same as for the baseline dataset. Results for the top 130–150 characters were congruent with the baseline results.
	A priori set of 15 characters from Morrone et al. (2012), datasets with 8 and 24 samples and	Appendix 5	Topology for the 8-sample set congruent with the baseline dataset at 3-group level.
	classification analyses using Gower and Czekanowski association measures (Test 5)		Topology for 24-sample set not congruent with baseline at 3-group level using Gower association measure, congruent using Czekanowski. Clusters for Cleistochloinae and Neurachninae congruent with baseline.

variations in composition of samples and characters (Table 5). The dataset required the presence of only a single species for each of the 10 genera to achieve consistent topologies with the baseline (Fig. 5, Appendix 3). Consistency in resolution of subclusters within the 'paniculate inflorescence group' was achieved when the dataset included 130 or more most discriminating characters based on the highest KW values (Table 7, Appendix 4). The composition of the clusters generated from analysis at the three-group level was consistent with the baseline topology, except in the case of the small character sets used in Test 4 (Table 5).

Analysis of datasets for *a priori* characters and two sample sizes generated different topologies (Test 5; Table 7, Appendix 5). The topology for the eight-sample dataset was congruent with the baseline. However, in the topology for the 15-sample dataset, the position of some of the subclusters within the 'paniculate inflorescence group' was affected (Appendix 5).

The most discriminating characters generated from the analyses were dependent of the composition of the sample and the group level. From analysis of the baseline dataset, the following were recorded for the top 40 most discriminating characters (Table 8):

1. The list comprised six categories of characters, *viz.* vegetative, terminal inflorescences, macromorphology of

- spikelets in the terminal inflorescences, micromorphology of obligately cleistogamous spikelets in axillary racemes, culm anatomy and surface micromorphology, and spikelet micromorphology.
- 2. Of the 10 most discriminating characters generated at the 3- and 10-group levels (corresponding to the three major clusters and the 10 putative genera respectively), there were three characters common to both, *viz*. terminal inflorescence type, characters of the upper glume and the germination lid on the upper lemma.
- 3. Nine of the characters were additions to the 104-character set from Thompson and Fabillo (2021).
- 4. Three of the fifteen characters from Morrone *et al.* (2012) were represented.

#### Discussion

The combination of processes in this study with Thompson and Fabillo (2021) provided a balance of information for alpha, beta and gamma taxonomy (Guerra-Garcia *et al.* 2008; Wood 2010). The comprehensive dataset of characters and samples permitted detailed description of the taxa at various ranks, and the erection of classifications that acknowledge natural groups (Clayton and Renvoize 1986; Wiens 2004; Kellogg 2006; (Guerra-Garcia *et al.* 2008;

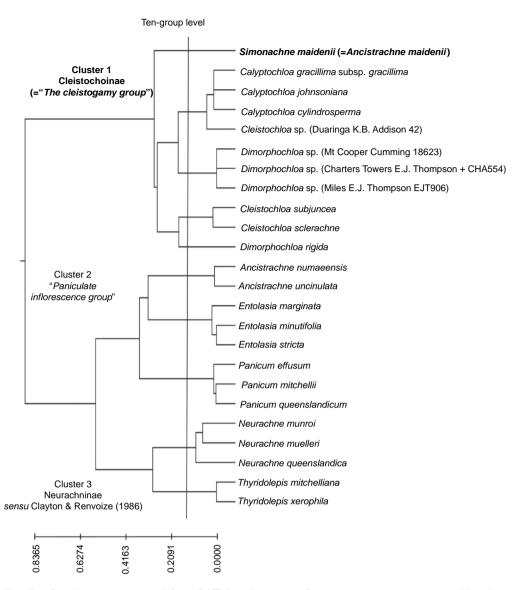


Fig. 5. Dendrogram generated from PATN analysis using Gower association measure and baseline dataset comprising 24 samples and 161 morphological characters. Three main clusters were resolved, viz. subtribes Cleistochloinae and subtribe Neurachninae sensu Clayton and Renvoize (1986) and 'paniculate inflorescence group' and 'paniculate inflorescence group'. Classification strategy set at flexible UPGMA agglomerative hierarchical fusion technique with Beta = −0.10. Size of symbols and letters indicates depth of field.

Wood 2010). The processes also provided information for accountable and transparent taxonomic decision making that included conducting impact assessments to determine the consistency of results from analyses of multiple datasets using different algorithms. Furthermore, the process provided information about the content of datasets required to achieve repeatable results. The phenetic analyses generated topologies that show the morphological similarities among taxa of various ranks and provided lists of the most discriminating characters to elucidate differences in the taxa.

The results from this study and Thompson and Fabillo (2021) support the intuitive evaluation of the taxonomic

status of *A. maidenii* by Simon and Alfonso (http://ausgrass2.myspecies.info/, accessed 21 September 2019) by revealing that *Ancistrachne s.l.* is not monophyletic. *A. maidenii* and *Ancistrachne s.s.* each bear combinations of characters that are distinctive in Poaceae and that distinguish them from each other. The distinguishing characters possessed by *A. maidenii* include a dimorphic reproductive system with similar CH and CL spikelet morphology, woolly indumentum on the upper glume and lower lemma, and presence of a contraligule (Campbell *et al.* 1983; Webster 1987; Watson and Dallwitz 1992; Thompson and Fabillo 2021; Appendix 7). Distinctive characters for *Ancistrachne* 

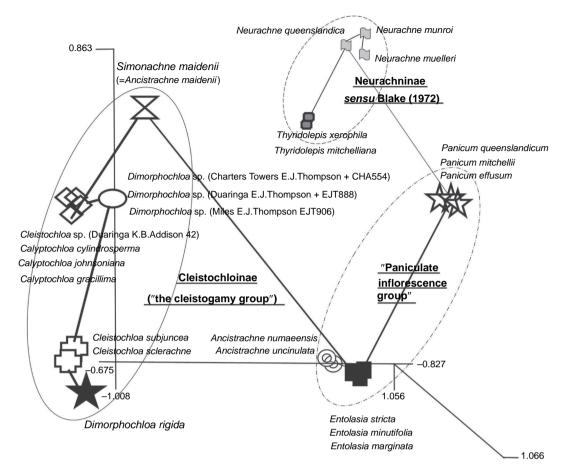


Fig. 6. Three-dimensional ordination generated from PATN analyses using Gower association measure, and dataset with 24 samples and all 161 morphological characters. Ordination stress value = 0.0711. Ordination from principal-component analysis using semi-strong hybrid multidimensional scaling (SSH).

s.s. that are not shared by A. maidenii but are found in other taxa include the following:

- 1. Uncinate tuberculate-based hairs on the spikelets are rare in Panicoideae but are very similar to those found in the Australian genera *Leptaspis* R.Br. and *Scrotochloa* Judz. (subfamily Pharoideae). However, the African genus *Pseudechinolaena* Stapf (subtribe Boivinellinae) has hooked hairs (Clayton 1974; Clayton and Renvoize 1982), also referred to as hooked spines (Watson and Dallwitz 1992), on the upper glumes of mature spikelets. These trichomes differ from those of *Ancistrachne s.s.* by their development (immature vs mature), shape (flattened when dry) and size (longer and thicker). Observations of specimens for this study showed that hairs on immature spikelets of *Pseudechinolaena* have an appearance different from mature the ones with the former appressed and without the hook.
- 2. Spikelets tilted on the pedicel also occurs in *Lasiacis* A.Hitchc. (Boivinellinae), *Panicum hirtum* Lam. (subtribe Panicinae), and *Tatianyx* Zuloaga & Soderstr. (tribe Paspaleae);

3. The rachillar stipe below the upper floret (Appendix 8), referred to as a proximal beak on the upper lemma by Thompson and Fabillo (2021), is very similar to that found in *Arundinella* Raddi (tribe Arundinelleae) and similar to *Entolasia* (Boivinellinae) but different from that found in *Yakirra* Lazarides & R.Webster and some species of *Panicum* (Lazarides and Webster 1984; Zuloaga 1986; Zuloaga *et al.* 2018).

The broad range of morphological characters used in this study showed similarities and differences in the taxa that support the definition of groups at several taxonomic ranks recognised by Thompson and Fabillo (2021). Nevertheless, although the results from the phenetic analyses indicate that small character sets such as the *a priori* set of 15 characters can be sufficient for segregation of clusters in topologies, they were inadequate for satisfying identity of taxa at all ranks.

'The cleistogamy group' is diagnosed here as the new subtribe, Cleistochloinae E.J.Thomps., and *Ancistrachne maidenii* is transferred to a new genus, *Simonachne* E.J.Thomps. Furthermore, *Cleistochloa* sp. (Duaringa K. B. Addison 42) is recognised as a new species of *Calyptochloa* 

Investigation of the effect of sample size using successive samples of the highest-ranking discriminating characters in terms of highest to lowest Kruskal-Wallis values generated from PATN analysis of the baseline dataset of 24 samples and 161 characters with Gower metric association measure.

		300		
Number of	Cluster assemblage	lage		Dendrogram -
discriminating characters	Subtribe Cleistochloinae	Neurachninae sensu Clayton and Renvoize (1986)	isu 'paniculate inflorescence group'	Appendix 4
10–20	Partially resolution of Cleistochloinae; Simonachne clustered with Ancistrachne Both clusters inconsistent with baseline.	Both clusters incon	sistent with baseline.	٧
30	As above		As above	В
40-60	Cleistochloinae segregated but genera relationships incongruent with baseline	Resolved	Relationships of genera inconsistent with baseline	U
70	Broadly similar to above. S <i>imonachne</i> clustered with Cleistochloa, incongruent with baseline. Dimophochloa spp. clustered with Calyptochloa in agreement with baseline		As above	Ω
80–161	Relationships of taxa within Cleistochloinae agree with baseline with Simonachne present as distinct cluster	As above	Consistent with baseline with Entolasia clustered with Ancistrachne.	ш

**Table 8.** The 40 most discriminating characters on the basis of Kruskal–Wallis values at the 10-group level generated from PATN cluster analysis by using the baseline dataset of 24 samples × 161 characters.

Character code	Character description
	Vegetative
I	Growth habit
	Terminal inflorescences
6* <sup>#</sup>	Terminal inflorescence type
	Macromorphology of spikelets in terminal inflorescences (mostly chasmogamous (CH))
15 <sup>B</sup>	Spikelet compression
22*#	Shape of apex of upper glume
23 <sup>A</sup>	Shape of body of upper glume
31	Similarity of the indumentum (type and distribution) on the upper glume to that on the lower lemma
32	Lower glume enveloping the upper glume at the base
34	Texture of margin of lower lemma
35	Type of tuberculate-based macrohairs on the back of the lower lemma of CH spikelets
36	Length of strap-shaped trichomes on distal margin of lower lemma
51 <sup>B</sup>	Presence of awn on the upper lemma of CH spikelets
55*#	Upper lemma with a germination lid
69 <sup>A</sup>	Shape of hilum
74	Length of hairs on distal margin of upper palea of CH spikelets
	Macromorphology of obligately cleistogamous (CL) spikelets mostly from leaf axils
79*	Composition of the CL inflorescences
80*	Nature of the leaf sheath at maturity subtending the CL spikelets
82*	Compression of CL spikelets, cross-sectional shape
83*	Orientation of CL spikelets
84* <sup>B</sup>	Pedicels of basal CL spikelets
89*#	Texture of margin of upper glume at apex
90*	Back of upper glume 2-keeled on CL spikelets
96*#	Margins of lower lemma of the axillary CL spikelets overlapping at apex
97*#	Margin of the lower lemma of the axillary CL spikelets with texture different from the body
101*	Lower lemma of CL spikelets with a proximal elaiosome

Table 8. (Continued)

Character code	Character description
102*	Back of lower lemma of axillary CL spikelets channelled
105*	Apex of the upper lemma of the axillary CL spikelets
106*	Relative length of upper lemma to lower lemmas of axillary spikelets
108*	Shape of apex of upper lemma of CL spikelets
110*	Surface of back of upper lemma of CL spikelets with minute longitudinal ridges
111*	Axillary CL spikelets with upper lemma with a germination lid
113*	Shape of apex of upper palea of axillary CL spikelets
II4 <sup>A</sup>	Cross-sectional shape of axillary CL caryopses
115 <sup>A</sup>	Longitudinal shape of axillary CL caryopses
	Culm anatomy and surface micromorphology
134 <sup>A</sup>	Shape of radial sclerenchyma from x-section of culm
135 <sup>A</sup>	Peripheral sclerenchyma separated from adjacent tertiary vascular bundle from x-section of culm
136 <sup>A</sup>	Inner sclerenchyma from x-section of culm
137 <sup>A</sup>	Sheath of primary vascular bundles from x-section of culm with xylem separated from parenchyma
	Spikelet micromorphology
142	Arrangement of papillae on the back of the upper lemma and palea
145*	Surface of the back of the upper lemma and palea on axillary CL spikelets
161 <sup>A</sup>	Relative length of stigma lobes

The 10-group level corresponds to the 10 putative genera. Character codes shown in bold represent the top 10 most discriminating characters at the 10-group level. Of the characters shown in this list, the most discriminating characters at the three-group level are indicated by an asterisk (\*) and of those overlapping with the 10 most discriminating characters at 10-group level are indicated by a number sign (#). The three-group level corresponds to the three main clusters, viz. Cleistochloinae ('the cleistogamy group'), Neurachninae sensu Clayton and Renvoize (1986) and the 'paniculate inflorescence group'.

<sup>A</sup>Indicates characters additional to those used by Thompson and Fabillo (2021). <sup>B</sup>Indicates characters used by Morrone et al. (2012).

and the three species of *Dimorphochloa* with phrase names are considered new species of a new genus that will be circumscribed elsewhere. It is suggested from interpretation of the results that *Ancistrachne s.s.* should be placed in Boivinellinae with *Entolasia*.

### **Taxonomic treatment**

Cleistochloinae E.J.Thomps. subtrib. nov.

Type: Cleistochloa C.E.Hubb

'The cleistogamy group' of Thompson and Fabillo (2021).

Plants stoloniferous, rhizomatous or decumbent. Collar of leaves with or without a contraligule. Inflorescences of two types; Type 1 terminal, a reduced spike-like panicle or raceme, and Type 2, reduced racemes, clandestine axillary, or exposed apical on short sub-branches. Spikelets of corresponding inflorescences relatively similar or markedly dissimilar; terminal spikelets chasmogamous or uncommonly mixed with cleistogamous spikelets; Type 2 spikelets cleistogamous. Spikelets slightly dorsi-ventrally compressed; lower glume absent or vestigial, upper glume and lower lemma, chartaceous, usually as long as the spikelet; lower floret barren without a palea; upper lemma chartaceous to cartilaginous, slightly hardened, papery texture, loosely overlapping the caryopsis. Anatomical type: C<sub>3</sub>.

## Key to the genera of Cleistochloinae

Three species with phrase names: *D.* sp. (Charters Towers *E. J. Thompson + CHA554*), *D.* (Miles *E. J. Thompson EJT906*), *D.* (Mt Cooper *R. J. Cumming 18623*).

Plants not stoloniferous, bushy with some decumbent branching; upper lemma subequal to lower lemma.......Dimorphochloa spp.

# Simonachne E.J.Thomps., gen. nov.

Type: Simonachne maidenii (A.A.Ham.) E.J.Thomps. (= Eriochloa maidenii A.A.Ham.)

Stoloniferous perennials with ascending fertile culms. Culm with pith. Leaf sheath with one margin pilose. Ligule and contraligule a fringe of hairs. Leaf blades lanceolate, base truncate, pseudopetiolate, proximal margins white, ciliate and one margin undulate. Inflorescences of two types, the terminal ones spike-like panicles with short proximal branches or raceme, and axillary racemes partly concealed within leaf sheath. Axes of inflorescence branches lacking pulvinii. Pedicel apices a shallow cup with thin walls. Spikelets of two similar types, falling entire except for proximal ones of axillary racemes trapped in leaf sheath terminal spikelets chasmogamous and axillary spikelets cleistogamous; adaxial, slightly dorsally compressed, elliptical in outline. Lower glume much reduced; upper glume 7-veined, woolly with mostly appressed tubercular-based hairs. Lower lemma

5-veined, similar in shape, size and indumentum to upper glume. Lower palea absent. Upper lemma subequal to lower lemma; 5-veined, chartaceous, body glabrous; margins hyaline, apex with flattened cilia; minutely longitudinally ridged, without papillae; apex mucronate; germination lid a crescent-shaped depression. Upper palea 2-veined. Anthers 3. Caryopsis dorsi-ventrally compressed; hilum punctiform.

#### **Etymology**

The genus is named in memory and honour of Bryan Kenneth Simon (1943–2015), curator of Poaceae at BRI for nearly 40 years and author of numerous publications on grasses, and *achne* from the Greek for scale in reference to the spikelets.

# Simonachne maidenii (A.A.Ham.) E.J.Thomps., comb. nov.

Eriochloa maidenii A.A.Ham., Proc. Linn. Soc. N.S.W. 37: 709 (1912); Ancistrachne maidenii (A.A.Ham.) Vickery, Contrib. N.S.W. Natl. Herb. 3(2): 83 (1961). Type: New South Wales. Hawkesbury River, 3 May 1912, A. A. Hamilton s.n. (holo: NSW [NSW52514, JSTOR photo !]; iso: BRI [AQ540088, !], NSW [NSW520052, JSTOR photo !]).

Fertile branches ascending to 40 cm high, copiously branched with up to ~12 nodes. Fertile culm internodes up to 4.5 cm long. Mature fertile leaf sheaths retained, convolute, pilose; outer margin ciliate with ascending tuberculate-based simple trichomes upto 0.5 mm long. Leaf blades upto 4.5 cm long and 7 mm wide; lower margin with tuberculate-based simple trichomes upto 3 mm long. Ligule 0.3 mm long. Contraligule 0.1 mm long. Mid-culm leaf blades 2.5–4.5 cm long, 2.5–4.0 mm wide, apex tapering, base truncate, margins white; both surfaces sparsely pubescent with simple trichomes upto 0.8 mm long. Terminal inflorescences on axes 1.5-5.5 cm long, 8-32-flowered; 0-several branches, appressed, rachis upto 1 cm long, 1-6-flowered. Spikelets 2.6-2.9 mm long, 1.0–1.1 mm wide; lateral pedicels 0.4–1.0 mm long, ultimate pedicel 0.5–2.0 mm long. Lower glume lunar, ~0.2 mm long, apex obtuse. Upper glume ovate, 2.6-2.9 mm long, apex acute, margins inrolled. Lower lemma ovate, 2.6-3.0 mm apex acute, margins inrolled. Upper lemma 2.4-2.6 mm long; apical cilia 60-90 µm long, mucronate to shortly awned. Lodicules ~0.2 mm long. Upper palea 2.4–2.6 mm long; apex acute. Anthers 1.5 mm long. Caryopsis ~1.7 mm long, ~0.8 mm wide, rarely present. Axillary inflorescences usually present below apical 2 or 3 internodes; 3- or 4-flowered, lowest 1 or 2 enclosed in leaf sheath. Spikelets 2.8-3.2 mm long, 1.2-1.3 mm wide. Lower glume vestigial upto 0.1 mm long. Upper glume ovate, 2.8-3.2 mm long, chartaceous, villous with woolly hairs upto 0.2 mm long; apex obtuse. Lower lemma ovate, 2.7–3.2 mm long; apex revolute. Upper lemma 2.2–2.4 mm long; apex revolute, cilia 60-90 µm long, mucronate to shortly awned. Upper palea 2.2-2.4 mm long; apex revolute. Anthers 0.2 mm long. Caryopsis 1.4–1.9 mm long, 0.7–0.9 mm wide; surface minutely longitudinally striate; light brown. Fig. 1, 2, Appendix 19.

#### Illustrations

D. J. B. Wheeler, S. W. L. Jacobs and B. E. Norton, *Grasses of New South Wales* 93 (1982); S. W. L. Jacobs and C. A. Wall in G. J. Harden (ed.), *Flora of New South Wales* 4: 452-3 (1993).

#### Additional specimens examined

NEW SOUTH WALES. Along an ephemeral creekline close to Old Great North Road, Devines Hill, 29 January 1999, P.Pike s.n. (NSW 427980); Singleton Road, Wiseman Ferry, 23 January 1999, P.Pike s.n. (NSW 427981); Erskine Creek downstream from Jack Evans track in the Blue Mountains National Park, 6 March 1999, R. G. Coveny 17686 & Hind (BRI AQ681893); 20 m E of Glenreagh - Grafton road, 7 km N of Glenreagh, 6 July 2001, G.Elks s.n. (NSW 488035); ~5 km along Stockyard Creek Road from Punchbowl Road, ~25 km NW by N of Grafton, 28 November 2005, Edwards 3 (NSW 618071); on left-hand side of track along Marramarra Creek ~20 m from Smugglers Ridge and Marramarra Ridge track exits, 17 November 2006, P. Pike s.n. (NSW 772920); Sandstone Drive, S of Bull paddock, Orara River, 3 January 2011, G. Clancy s.n. (NSW 882378); Porto Ridge, SW of Brooklyn, May 2011, R. G. Coveny 19454 (NSW 891751); Stockyard Creek, along Stockyard Creek Rd, ~30 km NNW of Grafton, 5 June 2013, E. J. Thompson EJT936 (BRI AQ1012262); near Whiporie, along Summerland Way, ~55 km N of Grafton, 5 June 2013, E. J. Thompson EJT939 (BRI AQ1012263); Coaldale, edge of Coaldale Rd nr. junction with Stockyard Creek Rd, ~31 km NNW of Grafton, 22 June 2013, E. J. Thompson EJT945 & G. P. Guymer (BRI AQ1012265); along Benowie walking track adjacent to Sams Creek, Berowa, September 2015, E. J. Thompson EJT1038 & G. P. Guymer (BRI AQ 971047). CULTIVATED. Ashgrove (ex Stockyard Creek Road), April 2017, E. J. Thompson MOR815 (BRI AQ1022226).

#### **Distribution**

Occurs in two disjunct populations on the New South Wales coastline (Fig. 3).

#### Habitat and ecology

Under a canopy of trees or shrubs on sandy soils derived from sandstone.

#### **Phenology**

Dates of herbarium collections indicate that *S. maidenii* flowers throughout the year. Cultivated plants flower mostly in summer.

#### **Conservation status**

Listed as *Vulnerable* under the *Biodiversity Conservation Act* 2016, New South Wales.

#### **Notes**

Simonachne maidenii shares morphological characters with all of the other taxa in subtribe Cleistochloinae (Table 1). Notable similarities include S. maidenii and Calyptochloa

spp. having a contraligule and the stoloniferous growth habit, giving plants a very similar appearance in the field.

#### **Breeding system**

The type of CL manifested by *Simonachne maidenii* fits the category of dimorphic anthers with amphigamy in the classification of CL by Thompson (2017). Axillary CL in *S. maidenii* is obligate, whereas in *Ancistrachne*, CL is facultative with the CL and CH anthers, the same size fitting the type 'monomorphic CH and CL anthers' on the same plant as defined by Thompson (2017).

# Micromorphology and macromorphology of the lemmas and palea

Simonachne maidenii and Ancistrachne s.s. differ in the lower lemmas by S. maidenii having absence of prickles and film of epicuticular wax. Upper lemmas of S. maidenii have minute longitudinal ridges and lack papillae. Upper lemmas of the terminal spikelets of S. maidenii are chartaceous to slightly hardened and loosely clasping the caryopsis, whereas for Ancistrachne s.s. they are hardened, glossy and tightly clasping the caryopsis. Simonachne maidenii, as for other species in Cleistochloinae, lacks a lower palea, whereas species of Ancistrachne, members of Neurachninae s.s. and the species of Panicum have a lower palea. Simonachne maidenii differs from Ancistrachne by the indumentum on the upper glume and lower lemma and differs from other members of Cleistochloinae by having an indistinct germination flap on the upper lemma (Tables 1, 3, Fig. 4; Appendices 8, 10–13).

#### Abaxial leaf-blade epidermis

Costal – intercostal zonation conspicuous. Papillae absent. Costal long cells rectangular, much narrower than intercostal; anticlinal walls of intercostal long cells  $\Omega$ -shaped. Anticlinal walls of intercostal long cells moderately undulating, often irregular with short-wave length. Stomata 38–43 µm long with low triangular subsidiaries, in 2 rows separated by 5–6 files of long cells. Bicellular microhairs 52–56 µm long, proximal cell longer than distal, occasional. Silica bodies in single rows, bilobate and polylobate, 16–25 µm long, common. Hooks present (Appendices 14, 15).

The shape of the anticlinal walls of long cells in the upper lemmas and the abaxial leaf surface in *A. maidenii* differ from those in *A. uncinulata*, and, in *A. maidenii*, the shapes of these walls differ from each other (Table 3). Such differences in the shape of anticlinal walls of long cells have been reported for some other panicoid grasses (Lu *et al.* 2009; Harun *et al.* 2020).

#### Transverse section of leaf blade

C<sub>3</sub>; XyMS+. Mesophyll with radiate chlorenchyma; adaxial pallisade chlorenchyma present. Midrib not prominent; with

a double bundle sheath; outer complete ring of parenchyma cells and partial inner ring of thick-walled cells with adaxial arc of clear parenchyma cells. Bulliform cells in discrete regular groups; in simple fans. Sclerenchyma accompanying all vascular bundles as adaxial strands and abaxial girders (Appendix 16).

#### Transverse section of culm

Culm examined 0.6 mm in diameter. Outer smallest vascular bundles adjacent to tangential girder sclerenchyma and imbedded in large-celled sclerenchyma. Vascular bundles with a ring of clear parenchyma; three sizes in separate circles, smallest to the periphery. Chlorenchyma in rectangular blocks, 2 or 3 cells deep by up to 10 cells wide; cells with regular size and shape, more or less circular. Inner ground tissue consisting of large thin-walled cells. (Appendix 17).

The distribution of the bundle sheath parenchyma of *S. maidenii* has similarities to the fresh culm sections of *Entolasia* spp. prepared for this study.

#### Surface of inflorescence culm

Pilose with tuberculate-based macrohairs up to 2 mm long, muriculate with hooks and scabridulous with prickles. Stomata frequent, similar to those on the abaxial leaf surface. Bicellar microhairs,  $\sim$ 44  $\mu$ m long, occasional. Silica bodies absent. (Appendix 18).

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Data availability. The data that support this study are available in Appendix 1.

Conflicts of interest. The author declares that he has no conflicts of interest.

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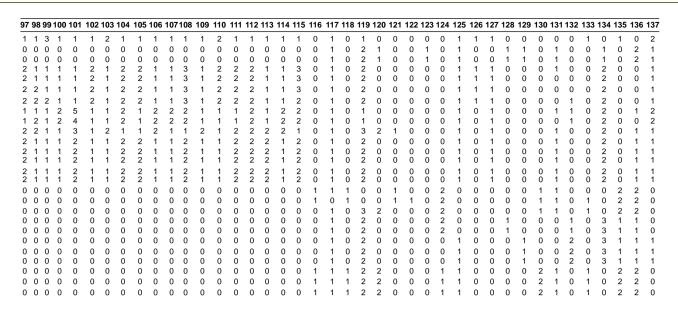
# Appendix I. Taxon and character-state data matrix for the 161 morphological characters for 24 samples

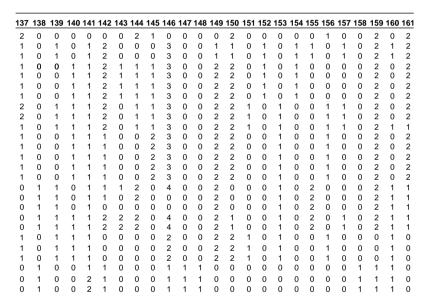
A number sign (#) indicates samples not used by Thompson and Fabillo (2021).

	1 2 3 4 5 6 7	8 9 10 11	12 13 14 1	16 17 18	19 20 21 22 2	3 24 25 26 27 2	8 29 30 31 32 33 34 35 36 37 38 39 40
Ancistrachne maidenii	3 0 0 0 1 3 0	0 0 0 0	0 0 0 0	0 0 1	0 0 0 1 0	1 0 0 0	0 0 0 0 2 0 1 3 1 1 0 0 0
Ancistrachne numaeensis	2000000	1 1 1 1	0 1 0 0	0 0 2	0 0 1 1	0 0 0 0	0 0 0 0 2 1 1 4 0 2 0 0 0
Ancistrachne uncinulata	2000000	1 1 1 1	0 1 0 0	0 0 2	0 0 1 1	0 0 0 0	0 0 0 0 2 1 1 4 0 2 0 0 0
Calyptochloa cylindrosperma	3 2 0 1 1 2 0	0 0 0 0	0 0 0 2	0 0 1	0 0 0 0	1 0 0 0	0 2 0 1 1 1 1 2 1 0 0 0 0
Calyptochloa gracillima	3 2 0 1 1 2 0	0 0 0 0	0 0 0 2	0 0 1	0 0 0 0	1 0 0 0	0 2 0 1 1 1 1 2 1 0 0 0 0
Calyptochloa johnsoniana	3 2 0 1 1 2 0	0 0 0 0	0 0 0 2	0 0 1	0 0 0 0	1 0 0 0	0 2 0 1 1 1 1 2 1 0 0 0 0
Cleistochloa sp. (Duaringa K.B. Addison 42)	3001120	0 0 0 0	0 0 0 2	0 0 1	0 0 0 0	1 0 0 0	0 2 0 1 1 1 1 2 2 0 0 0 0
Cleistochloa sclerachne	2001031	0 0 0 0	0 0 0 1	0 0 1	0 0 0 0	1 0 0 0	0 1 0 0 1 1 1 1 2 0 0 0 0
Cleistochloa subjuncea	2001031	0 0 0 0	0 0 0 1	0 0 1	0 0 0 0	1 0 0 0	0 1 0 0 1 1 1 1 2 0 0 0 0
Dimorphochloa rigida	2001031	0 0 0 0	0 0 0 1	0 0 0	0 0 0 0	1 0 0 0	0 0 0 0 1 0 1 1 2 0 0 2 0
Dimorphochloa sp. (Charters Towers E.J.Thompson + CHA554)	0 2 0 1 1 2 1	0 0 0 0	0 0 0 2	0 0 1	0 0 0 0	1 0 0 0	0 0 0 1 1 1 1 2 2 0 0 0 0
Dimorphochloa sp. (Miles E.J. Thompson EJT906)	0 2 0 1 1 2 1	0 0 0 0	0 0 0 2	0 0 1	0 0 0 0	1 0 0 0	0 0 0 1 1 1 1 2 2 0 0 0 0
Dimorphochloa sp. (Duaringa E.J. Thompson + EJT888) #	0 2 0 1 1 2 1	0 0 0 0	0 0 0 2	0 0 1	0 0 0 0	1 0 0 0	0 0 0 1 1 1 1 2 2 0 0 0 0
Dimorphochloa sp. (Mt Cooper R.J. Cumming 18623) #	0201121	0 0 0 0	0 0 0 2	0 0 1	0 0 0 0	1 0 0 0	0 0 0 1 1 1 1 2 2 0 0 0 0
Dimorphochloa sp. (Black Jack E.J. Thompson + CHA835)		0 0 0 0	0 0 0 2	0 0 1	0 0 0 0	1000	0 0 0 1 1 1 1 2 2 0 0 0 0
Neurachne muelleri	3 0 1 0 0 1 1	1 2 0 0	1 1 0 2	1 0 2	1 0 0 2 2	2 1 1 1 0	0 2 1 2 3 1 2 0 2 0 1 2 1
Neurachne munroi	2010011	0 2 0 0	1 1 0 2	1 0 2	1 0 0 2 2	2 1 1 1 0	0 2 1 2 3 1 2 1 2 0 1 0 1
Neurachne queenslandica	2 1 1 0 0 1 1	0 2 0 0	0 1 0 2	1 0 2	1 0 0 2 2	2 1 1 1 0	0 2 1 2 3 1 2 1 2 0 1 0 1
Thyridolepis mitchelliana	1 1 1 0 0 1 1	0 3 0 0	0 1 0 2	1 1 2	1 1 0 3 2	2 1 2 1 0	0 2 1 2 0 2 2 1 1 0 0 0 0
Thyridolepis xerophila	1 1 1 0 0 1 1	0 3 0 0	0 1 0 2	1 1 2	1 1 0 3 2	2 1 2 1 0	0 2 1 2 0 2 2 1 1 0 0 0 0
Entolasia marginata	2000000	1 0 0 0	0 0 0 0	0 0 2	0 0 0 1 0	0 0 0 0	1 0 0 0 2 0 0 0 0 0 1 0
Entolasia minutifolia	2000000	1000	0 0 0 0	0 0 2	0 0 0 1 0	0 0 0 0	1 0 0 0 2 0 0 0 0 0 1 0
Entolasia stricta	2000000	1 0 0 0	0 0 0 0	0 0 2	0 0 0 1 0	0 0 0 0	1 0 0 0 2 0 0 0 0 0 1 0
Panicum effusum	1020000	1 0 0 0	0 0 1 0	0 0 2	0 0 0 1	0 0 0 1	0 0 0 0 3 1 0 0 0 0 0 0 0
Panicum mitchellii <sup>#</sup>	1000000	1 0 0 0	0 0 1 0	0 0 2	0 0 0 1	0 0 0 1	0 0 0 0 3 1 0 0 0 0 0 0 0
Panicum queenslandicum <sup>#</sup>	1000000	1 0 0 0	0 0 1 0	0 0 2	0 0 0 2	0 0 0 1	0 0 0 0 3 1 0 0 0 0 0 0 0

41 42 43 44 43 46 47 48 49 30 31 32 33 34 3	33 36 37 36 39 60 61 62 63 64 63 66	07 08 09 70 71 72 73 74 75 76	11 18 19 80 81 82 83 84 85 86 87 88 89 90 91 92 93 94 95 96
0 0 0 0 0 0 1 0 0 0 1 0 2 1	1 0 0 1 0 1 0 0 2 0 1 1	0 1 1 0 1 1 0 1 0 1	2 0 4 1 0 1 1 1 1 2 3 2 1 1 1 1 1 2 1 2
0 1 0 2 0 0 1 0 1 0 1 0 3 0	1 1 1 1 0 1 0 0 2 1 0 0	0 0 2 0 2 0 1 1 1 1	2 1 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 1 0 2 0 0 1 0 1 0 1 0 3 1	1 1 0 1 0 1 0 0 2 1 0 0	0 0 2 0 2 0 1 1 1 1	2 1 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 0 0 0 0 0 0 0 1 0 2 1 2 2 3	2 0 0 1 0 2 0 0 3 0 1 1	0 1 1 0 0 1 0 2 0 1	2 0 1 1 0 3 1 2 2 2 2 1 2 2 1 2 1 1 1 2
0 0 0 0 0 0 0 0 1 0 2 1 2 2 2	2 0 0 1 0 2 0 0 3 0 1 1	0 1 1 0 0 1 0 2 0 1	2 0 1 1 1 3 1 2 2 2 2 1 2 2 1 2 1 1 1 2
0 0 0 0 0 0 0 0 1 0 2 1 2 2 3	2 0 0 1 0 2 0 0 3 0 1 1	0 1 1 0 0 1 0 2 0 1	2 0 2 1 0 3 1 2 2 2 2 1 2 2 1 2 1 1 1 2
0 0 0 0 0 0 0 0 1 0 2 1 2 2 2	2 0 0 1 0 2 0 0 3 0 1 1	0 1 1 0 0 1 0 2 0 1	2 2 4 1 0 1 1 2 2 2 2 1 2 2 1 2 1 1 1 2
0 0 0 0 0 0 0 0 1 0 1 1 2 0 2	2 0 0 1 0 2 0 0 3 0 1 1	0 1 1 0 2 1 1 3 0 1	2 0 1 2 0 2 2 1 2 2 2 1 1 2 1 2 2 1 2 1
0 0 0 0 0 0 0 0 1 0 1 1 2 0 2	2 0 0 1 0 2 0 0 3 0 1 1	0 1 1 0 2 1 1 2 0 1	2 0 1 2 0 2 2 1 2 2 2 1 1 2 1 2 2 1 2 1
1 0 0 0 0 0 0 0 1 0 1 1 2 0 2	2 0 0 1 1 2 0 1 3 0 1 0	0 0 1 0 2 1 1 2 1 1	2 0 3 2 0 2 2 1 1 1 1 2 1 1 2 1 1 2 1 1
0 0 0 0 0 0 0 0 1 0 2 1 2 1 3	2 0 0 1 0 2 0 0 3 0 1 0	0 1 1 0 0 1 0 2 1 1	2 0 2 1 0 1 1 2 2 2 2 1 1 1 1 2 1 1 1 2
0 0 0 0 0 0 0 0 1 0 2 1 2 1 3	2 0 0 1 0 2 0 0 3 0 1 0	0 1 1 0 0 1 0 2 1 1	2 0 2 1 0 1 1 2 2 2 2 1 1 1 1 2 1 1 1 2
0 0 0 0 0 0 0 0 1 0 2 1 2 1 3	2 0 0 1 0 2 0 0 3 0 1 0	0 1 1 0 0 1 0 2 1 1	2 0 2 1 0 1 1 2 2 2 2 1 1 1 1 2 1 1 1 2
0 0 0 0 0 0 0 0 1 0 2 1 2 1 3	2 0 0 1 0 2 0 0 3 0 1 0	0 1 1 0 0 1 0 2 1 1	2 0 2 1 0 1 1 2 2 2 2 1 1 1 1 2 1 1 1 2
0 0 0 0 0 0 0 0 1 0 2 1 2 1 3	2 0 0 1 0 2 0 0 3 0 1 0	0 1 1 0 0 1 0 2 1 1	2 0 2 1 0 1 1 2 2 2 2 1 1 1 1 2 1 1 1 2
1 1 0 2 1 1 0 1 1 1 0 2 2 0 (	0 0 0 1 0 2 0 0 3 0 2 2	1 1 2 0 0 0 2 0 0 0	0 1 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 1 0 2 1 1 0 1 1 1 0 1 0 0	0 0 0 0 0 1 0 0 3 0 2 2	1 1 2 0 0 0 2 0 0 0	0 1 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 1 0 2 1 1 0 1 1 1 0 1 1 0 (	0 0 1 0 0 2 0 0 3 0 2 2	1 1 2 0 0 0 2 0 0 0	0 1 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 0 1 2 1 2 1 0 0 0 0 2 2 0 (	0 0 0 1 0 0 0 0 1 0 3 1	1 1 1 1 0 0 2 0 0 0	0 2 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 0 1 2 1 2 1 0 0 0 0 2 2 0 (	0 0 0 1 0 0 0 0 1 0 3 1	1 1 1 1 0 0 2 0 0 0	0 2 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 0 0 0 0 0 1 0 0 0 0 2 3 2	1 1 0 1 0 1 1 0 3 0 1 1	0 0 0 0 0 0 0 1 0 1	2 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 0 0 0 0 0 1 0 1 0 0 0 3 0	1 1 0 1 0 1 1 0 3 0 1 1	0 0 0 0 0 0 0 1 0 1	2 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 0 0 0 0 0 1 0 1 0 0 0 3 0	1 1 0 1 0 1 1 0 3 0 1 1	0 0 0 0 0 0 0 1 0 1	2 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 1 0 1 0 0 1 0 1 0 0 0 3 0	1 0 0 0 0 0 0 0 0 0 0 1	0 0 0 1 3 0 0 0 0 1	1 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 1 0 1 0 0 1 0 1 0 0 0 3 0	1 0 0 0 0 0 0 0 0 0 0 1	0 0 0 1 3 0 0 0 0 1	1 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
0 1 0 1 0 0 1 0 1 0 0 0 3 0	1 0 0 0 0 0 0 0 0 0 1	0 0 0 1 3 0 0 0 0 1	1 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0

A1 A2 A3 AA A5 A6 A7 A8 A0 50 51 52 53 5A 55 56 57 58 50 60 61 62 63 6A 65 66 67 68 60 70 71 72 73 7A 75 76 77 78 70 80 81 82 83 8A 85 86 87 88 80 00 01 02 03 0A 05 06





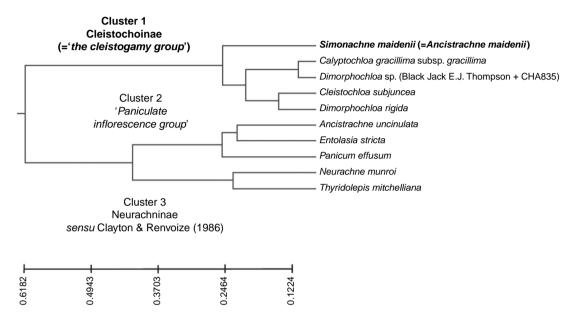
# Appendix 2. Topotypes for Ancistrachne uncinulata

PORT CURTIS DISTRICT: Fingerfield Rd Rules Beach – Private Holding, April 2003, Sankowsky and Sankowsky 1966; Yeppoon, ~2 km N of Township on road to Bayfield, June 1978, Sharpe 2366; between Lotus and Marlborough, June 1955, Beauglehole 3578; Pine Inlet, Percy Islands, 87 miles (~140 km) SE of Mackay, May 1956, Lazarides 5677A; Deep Water National Park, 40 km E of Miriam Vale, March 1990, Gibson TOI919.

SOUTH KENNEDY DISTRICT: Cape Hillsborough National Park, near Hidden Valley, July 1994, *Bvatianoff & Dillewaard 9407140*; Shoalwater Bay Training Area, Pine Mountain sector, western slope of Pine Mountain, April 2011, *Halford & Bean QM329*; Calder Island, 60 km NE of Mackay, May 1992, *Halford & Crombie Q1346*.

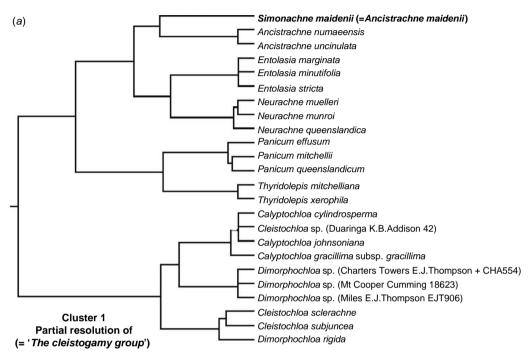
# Appendix 3. Dendrogram generated from analysis of dataset composed of 10 samples corresponding to the 10 putative genera

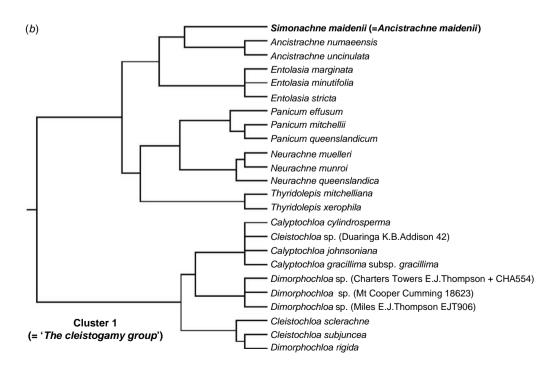
Dendrogram generated from PATN analysis using GOWER association measure and a subset of 10 samples and 161 morphological characters. Three main clusters, subtribes Cleistochloinae and Neurachninae sensu Clayton & Renvoize (1986) and the 'paniculate inflorescence group', congruent with the result using the baseline dataset comprising 24 samples and 161 characters. Classification strategy set at flexible UPGMA agglomerative hierarchical fusion technique with Beta = -0.10.

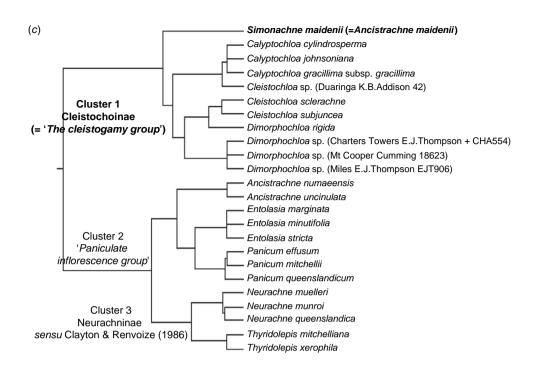


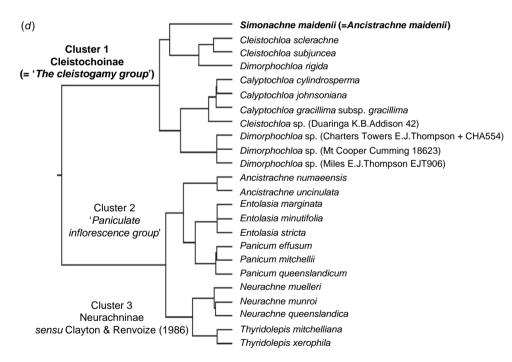
# Appendix 4. Dendrograms generated from PATN analyses using various datasets and analyses run using GOWER association measure

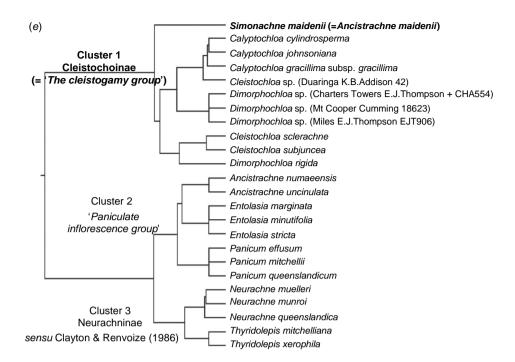
Datasets comprise 24 samples and sets of most discriminating characters based on Kruskal–Wallis values obtained from the analysis of the baseline dataset of 24 samples and 161 morphological characters. (a) Top 10–20 characters. (b) Top 30 characters. (c) Top 40–60 characters. (d) Top 70 characters. (e) Top 80–161 characters. Topologies consistent with the baseline for datasets with 80 or more characters. Classification strategy set at flexible UPGMA agglomerative hierarchical fusion technique with Beta = -0.10.





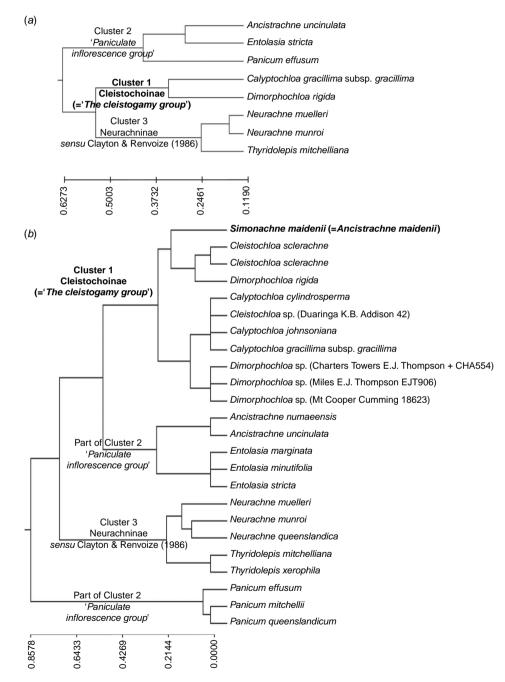






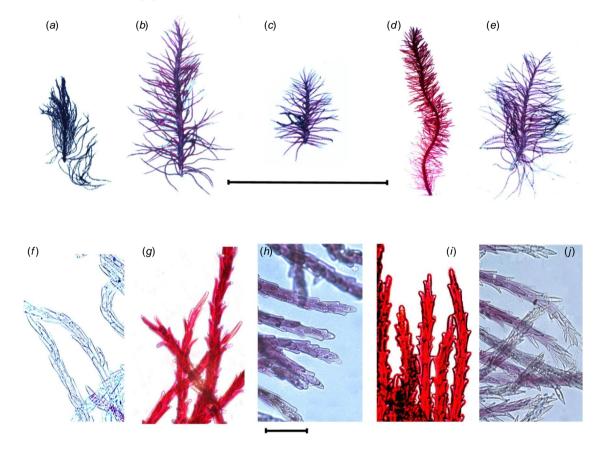
# Appendix 5. Dendrograms generated from analysis of datasets comprising predetermined sample composition and character set as used by Morrone et al. (2012)

Dendrograms generated from PATN analyses using GOWER association measure and two subsets of data based on 15 characters used Morrone *et al.* (2012). (a) Overlapping samples from Neurachninae *sensu* Morrone *et al.* (2012) using Australian species of *Panicum s.s.* and *Neurachne munroi* substituted for *Neurachne alopecuroidea*. Three main clusters congruent with result from baseline dataset but similarity relationships different. (b) All 24 samples. The topology is not congruent with the result from the baseline dataset. Classification strategy set at flexible UPGMA agglomerative hierarchical fusion technique with Beta = -0.10.



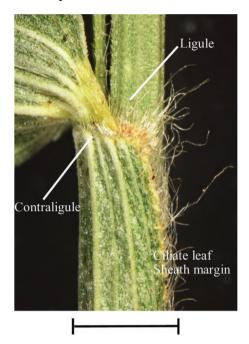
## Appendix 6. Stigmas of some of the species from rehydrated and fresh samples

(a, f) Simonachne maidenii (rehydrated) from Thompson EJT936 (BRI). (b, g) Ancistrachne uncinulata from Thompson EJT1018 (BRI). (c, h) Panicum effusum from Thompson EJT916 (BRI). (d, i) Neurachne muelleri from Thompson GAL354 (BRI). (e, j) Dimorphochloa sp. (Miles E.J. Thompson EJT906) from Thompson + EJT906 (BRI). Images: E. J. Thompson. Scale bar: 2 mm for a-e and  $50 \text{ }\mu\text{m}$  for f-j.



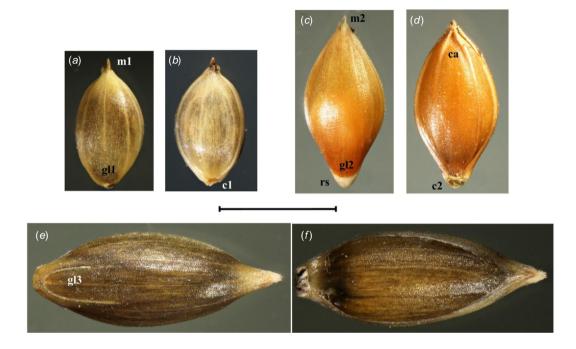
# Appendix 7. Contraligule of Simonachne maidenii

From Thompson EJT936 (BRI). Photo: E. J. Thompson. Scale bar: 1 mm.



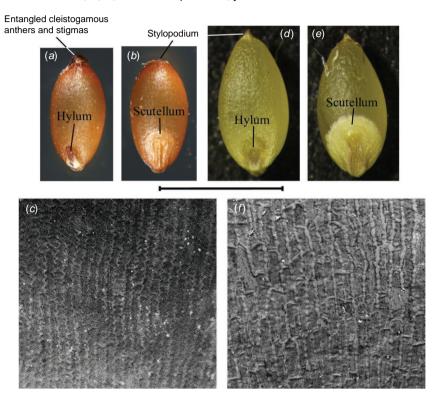
## Appendix 8. Lemmas and paleas of upper florets

(a, b) Cleistogamous spikelet from axillary inflorescence of Simonachne maidenii. (a) Upper lemma. (b) Upper palea. (c, d) Ancistrachne uncinulata. (c) Upper lemma. (d) Upper palea. (e) Cleistogamous spikelet from axillary inflorescence of Cleistochloa subjuncea, upper lemma. (f) Upper palea. (a, b) From Thompson EJT936 (BRI), (c, d) from Thompson EJT1018 (BRI) and (e, f) from Thompson HUG815 & Simon (BRI). Photos: E. J. Thompson. c1, transverse callus; ca, creased apex; c2, oblique callus; g11, germination lid with inconspicuous crescent-shaped depression between raised veins; g12, lid inconspicuous; g13, lid with crescent-shaped depression between raised veins; m1, mucronate to shortly awned; m2, mucronate; pb, proximal beak. Scale bar: 2 mm.



## Appendix 9. Caryopses of Simonachne maidenii and Ancistrachne uncinulata

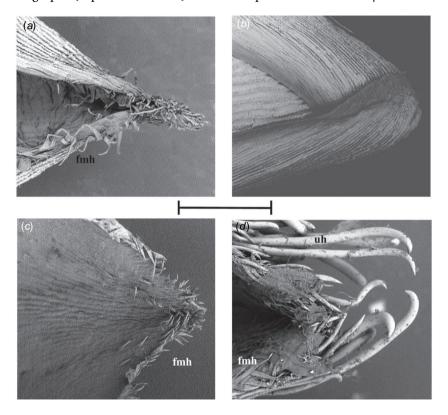
(a–c) Simonachne maidenii. (a) View of hylum. (b) View of scutellum. (c) Scanning electron micrograph of endosperm. (a–f) Ancistrachne uncinulata. (d) View of hylum. (e) View of scutellum. (f) Scanning electron micrograph of endosperm. (a–c) From Thompson EJT936 (BRI) and (d–f) from Thompson EJT1018 (BRI). Photos and micrographs (captured at  $500 \times$ ): E. J. Thompson. Scale bar: 2 mm for a, b, d, e and  $200 \, \mu m$  for c, f.



# Appendix 10. Scanning electron micrographs of surface of upper lemmas

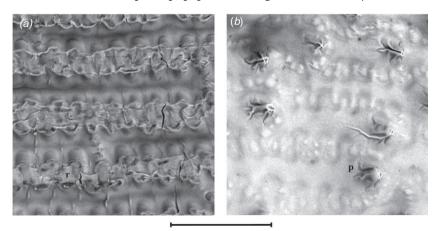
(a) Simonachne maidenii, from terminal spikelet. (b) S. maidenii from axillary spikelet. (c) Ancistrachne uncinulata. (d) Ancistrachne numaeensis. (a, b) From Thompson EJT936 (BRI), (c) from Thompson EJT1018 (BRI) and (d) from

van Drot 567 (NOU). Micrographs (captured at 500×): E. J. Thompson. Scale bar: 200 μm.



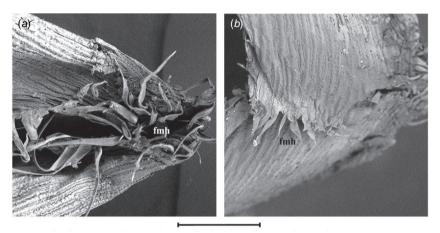
# Appendix 11. Scanning electron micrographs of surface of upper lemmas

(a) Simonachne maidenii, longitudinal ridges, papillae absent, elongated U-shaped anticlinal walls of long cells. (b) Ancistrachne uncinulata, lacking ridges, papillae with beak-like apex to nipple-shaped, irregularly spaced in longitudinal rows, U-shaped anticlinal walls of long cells. (a) From Thompson EJT936 (BRI) and (b) from Thompson EJT1018 (BRI). Micrographs (captured at 4000×): E. J. Thompson. p, papillae, r, ridge. Scale bar: 30 μm.



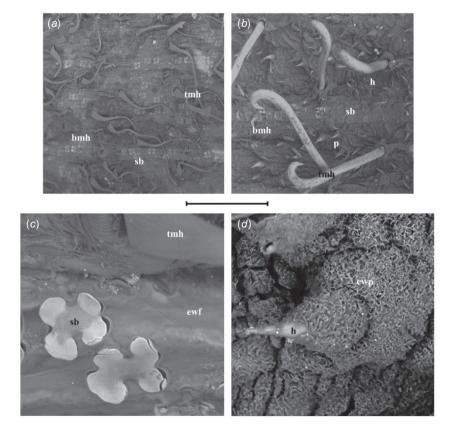
## Appendix 12. Scanning electron micrographs of surface of upper paleas

(a) From chasmogamous spikelet of *Simonachne maidenii*. (b) From spikelet of *Ancistrachne uncinulata*. (a) From *Thompson EJT936* (BRI), (b) from *Thompson EJT1018* (BRI). Micrographs (captured at  $1000 \times$ ): E. J. Thompson. fmh flattened microhairs on margin. Scale bar:  $50 \, \mu m$ .



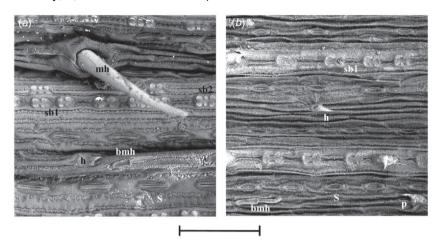
# Appendix 13. Scanning electron micrographs of apex of lower lemmas

(a, c) Simonachne maidenii, silica bodies with narrow waist and indented ends. (b, d) Ancistrachne uncinulata, silica bodies with broad waist and rounded ends. (a, c) From Thompson EJT936 (BRI) and (b, d) from Thompson EJT1018 (BRI). Micrographs (captured at  $1000 \times \& 5000 \times$ ): E. J. Thompson. bmh, bicellular microhair; ewf, film of epicuticular wax; ewp, platelets of epicuticular wax; h, hook; p, prickle; sb, silica body; tmh, tuberculate-based macrohair. Scale bars:  $200 \, \mu m$  for a, b and  $20 \, \mu m$  for c, d.



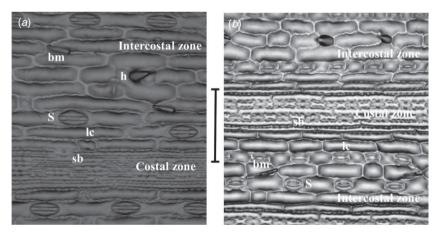
## Appendix 14. Scanning electron micrographs of abaxial surface of leaf blade

(a) Simonachne maidenii. (b) Ancistrachne uncinulata. (a) From Thompson EJT936 (BRI); (b) from Thompson EJT1018 (BRI). Micrographs: E. J. Thompson, captured at  $1000 \times$ . bmh, bicellular microhair; h, hook; mh, macrohair; sb1, bilobate silica body; Sb2, polylobate silica body; S, stoma. Scale bar:  $100 \, \mu m$ .



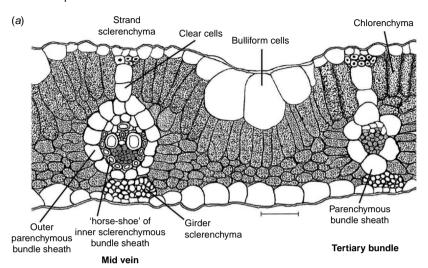
# Appendix 15. Replicas of abaxial surface of fresh leaf blade

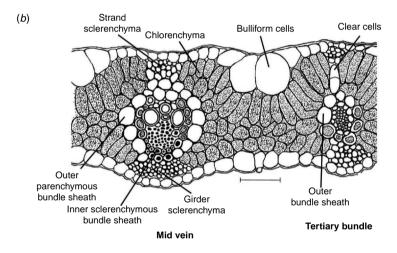
(a) Simonachne maidenii. (b) Ancistrachne uncinulata. (a) From Thompson EJT936 (BRI); (b) from Thompson EJT1018 (BRI). Photos: E. J. Thompson. bm, bicellular microhair; h, hook; lc, long cell; sb, silica body; S, stoma. Scale bar: 100 µm.



## Appendix 16. Transverse section of fresh leaf at mid-vein

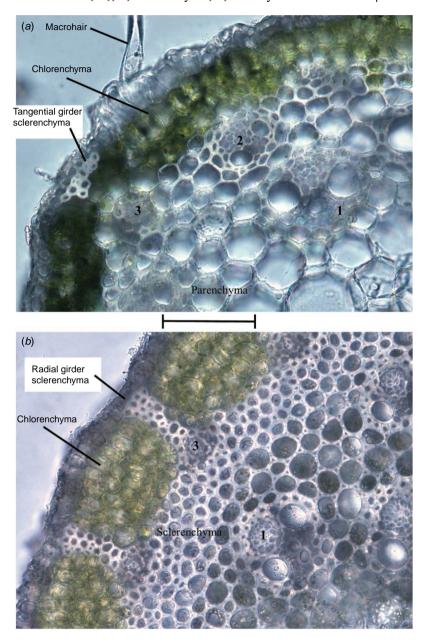
(a) Simonachne maidenii. (b) Ancistrachne uncinulata. (a) From Thompson EJT936 (BRI); (b) from Thompson EJT1018 (BRI). Del. E. J. Thompson. Scale bar: 50 µm.





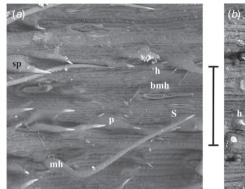
# Appendix 17. Transverse section of fresh terminal inflorescence culm

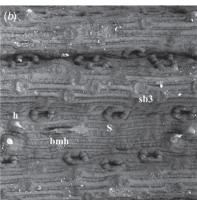
(a) Simonachne maidenii, vascular bundles with parenchymous sheath. (b) Ancistrachne uncinulata, example with vascular bundles surrounded by sclerenchyma. (a) From Thompson EJT936 (BRI); (b) from Thompson EJT1018 (BRI). Photos: E. J. Thompson. 1, primary vascular bundle (vb); 2, secondary vb; 3, tertiary vb. Scale bar: 50 µm.



# Appendix 18. Scanning electron micrograph of surface of terminal inflorescence culm

(a) Simonachne maidenii. (b) Ancistrachne uncinulata. (a) From Thompson EJT936 (BRI); (b) from Thompson EJT1018 (BRI). Micrographs: E. J. Thompson captured at  $1000 \times$ . bmh, bicellular microhair; h, hook; mh, macrohair; p, prickle; sb3, saddle-shaped silica body; sp, spicule; S, stoma. Scale bar:  $100 \, \mu m$ .





Appendix 19. Holotype of Eriochloa maidenii (=Simonachne maidenii) (A.A. Hamilton s.n. (NSW 52514))

