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Acacia cineramis (Leguminosae: Mimosoideae), a new species endemic to south-eastern Australia, and an investigation of phyllode nervature in allied species

Harvey K. Orel[1,2], Daniel J. Murphy[2] & Neville G. Walsh[2]

[1] School of Biosciences, The University of Melbourne, Parkville, VIC 3010, Australia;
   e-mail: horel@student.unimelb.edu.au

[2] Royal Botanic Gardens Victoria, Birdwood Avenue, Melbourne, VIC 3004, Australia

Introduction

The genus *Acacia* Mill. (Leguminosae: Mimosoideae) is an iconic plant group of the Australian flora and, consisting of over 1050 currently recognised species (Maslin 2015), is also the largest angiosperm genus in the country. *Acacia rigens* A.Cunn. ex G.Don is a relatively widespread species found in all states except Tasmania, growing mainly on sandy and shaly soils in mallee scrub (Cowan 2001). It commonly extends from the Gawler Ranges, South Australia (S.A.), through north-western Victoria (Vic.) to Dubbo, New South Wales (N.S.W.). Occurrences of the species outside this range are scattered, with disjunct populations occurring in southern Western Australia (W.A.), the Northern Territory (N.T.) and southern Queensland (Qld) (Cowan 2001, 2018; AVH 2018). It is typically a spreading shrub to 3 m high or (occasionally) a tree to 6 m high, with straight to shallowly incurved, terete or sometimes flat plurinerved phyllodes 3–14 (–16) cm long and 0.8–3 mm in wide. Historically, the species has been regarded as a member of section *Plurinerves* (Pedley 1978); however, a more recent molecular study by Murphy *et al.* (2010)

Abstract

A morphometric multivariate analysis of *Acacia rigens* A.Cunn. ex G.Don and an entity previously recognised as *Acacia* sp. Gerang Gerung (M.G.Corrick 6451) was conducted, with the latter described here as a new species, *Acacia cineramis* H.K.Orel. This new species is considered endangered in accordance with IUCN classification, and known populations occur only in the Wimmera region of central-western Victoria. A preliminary assessment of phyllode anatomy, focusing on phyllode nervature, in *A. cineramis* and related species is also presented.

Keywords: Wimmera, Wattle, endangered, multivariate analysis, phyllode anatomy, vasculature

established that the sectional classification of the genus warranted revision, and that section Plurinerves was not monophyletic. As such, *A. rigens* was placed within a diverse group of taxa informally termed the 'p.u.b. clade' as a member of 'subclade E1' with six other taxa that mostly share narrow, sclerophyllous phyllodes and which occur in arid to semi-arid areas (Murphy *et al.* 2010). A larger phylogenetic study (Mishler *et al.* 2014) found a similar result, and resolved *A. rigens* in a clade sister to *A. enterocarpa* R.V.Sm. and *A. hexaneura* P.Lang & R.S.Cowan.

Prior to the current study, a number of specimens in the National Herbarium of Victoria (MEL), previously identified as *Acacia rigens,* had been noted to differ morphologically from typical *A. rigens* and were collectively given the informal name *Acacia* sp. aff. *rigens* (Gerang Gerung) (hereafter referred to as *A.* 'Gerang Gerung') in reference to the area from which several of the specimens were collected. The taxon is listed in APNI (CHAH 2006) as *Acacia* sp. Gerang Gerung (M.G.Corrick 6451) Vic. Herbarium. From herbarium records, the distribution of *A.* 'Gerang Gerung' extends approximately from the town of Dimboola, through Gerang Gerung and Nhill to just north of Kaniva near the South Australia-Victoria border. Specimens of *A.* 'Gerang Gerung' were principally distinguished from *A. rigens* by a number of morphological differences in characters of the branchlets, phyllodes, peduncles and inflorescences (VicFlora 2018). However, the degree of differentiation between these features was not thoroughly defined, and it was not certain that the distinguishing characters were appropriate to justify the recognition of *A.* 'Gerang Gerung' as a different taxon to *A. rigens*.

Accordingly, a morphometric analysis of specimens of *A. rigens* and *A.* 'Gerang Gerung' was undertaken in order to examine the morphological variation between the two putative entities and assess the taxonomic status of *A.* 'Gerang Gerung'. The analysis also presented an opportunity to conduct an examination of morphological variation in *A. rigens* across its range.

Phyllode nerve anatomy and classification in *Acacia*

Acacia phyllodes have an anatomical structure that is unique amongst angiosperms, in particular in the way vascular tissue (commonly referred to in *Acacia* as 'veins' or, as used in this study, 'nerves') is arranged (Gardner *et al.* 2008). It has long been recognised that the

arrangement and number of nerves is a key character for the taxonomy and classification of *Acacia* (Boughton 1986), and there have been a number of studies on the physiological function and development of phyllodes and these nerves (e.g. Boughton 1990; von Wartburg 1991; Brodribb and Hill 1993; Sommerville *et al.* 2012). However, there has been relatively little assessment of the phylogenetic implications of phyllode nervature beyond the broad categories of 'uninerved' versus 'plurinerved' that have been traditionally used to define sections in the classification of the genus. Given that there has been a recent overhaul of the phylogenetic understanding of *Acacia* in comparison to the traditional sectional classifications (Pedley 1978; Murphy *et al.* 2010), the understanding of phyllode anatomy is possibly highly significant, as differences in nerve arrangement among taxa thought to be closely related to *A.* 'Gerang Gerung' may assist in clarifying species boundaries, and help identify possible phylogenetic characters for use in assessing deeper phylogenetic relationships in *Acacia*. For this reason, a descriptive survey of phyllode anatomy was conducted, with an emphasis on recording any diagnosable differences in phyllode nervature between established and putative species.

Methods

Sampling

In total, 70 specimens were assessed in this study, with each specimen considered as a single operational taxonomic unit (OTU). Sixty herbarium specimens, housed in the National Herbarium of Victoria (MEL), were included from across the range of *Acacia rigens* (see Figure 1). Of these, 15 were previously identified as *A.* 'Gerang Gerung' and 45 were considered *A. rigens*. In addition, 10 fresh collections (eight of *A.* 'Gerang Gerung' and two *A. rigens*) were made to supplement the previously existing herbarium material and to enable comparison between fresh and dried specimens. Notably, these represent the first collections of *A.* 'Gerang Gerung' made in more than 30 years. All supplementary specimens have been lodged at MEL.

Morphometric characters

Ultimately, 26 morphological characters were used in this study (Table 1) based, in part, on characters utilised
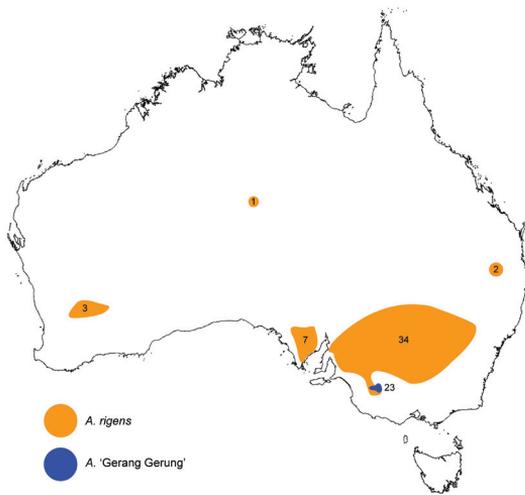
Figure 1. Map of Australia displaying the collection locations of *Acacia rigens* and *Acacia* 'Gerang Gerung' (now *Acacia cineramis*) specimens used in this study. Numbers inside or adjacent to coloured regions indicate the number of specimens from that region.

in identification keys for the delineation of *Acacia* species closely related to, or resembling *A. rigens*, as well as those characters informally employed to distinguish the subject taxa in the past (VicFlora 2018). Also included were some new characters observed to differ between the entities during the course of this study. The characters assessed can be segregated into four broad categories based on anatomy: branchlet, phyllode, inflorescence and pod characters. For each specimen, where possible, four measurements were taken for all phyllode, inflorescence and pod characters and used to calculate a mean value for each character which was utilised in our analysis. Due to the predominantly binary nature of the branchlet characters used, specimens were scored only once for each branchlet character. Seed characters were initially scored, however, it was ultimately considered that there were too many specimens with missing data for these characters, so they were excluded from the analysis. Reasons for the inclusion of the morphological characters are provided, with bracketed numbers corresponding to the character numbers listed in Table 1.

Branchlet characters

Several differences in features of the branchlet had previously been noted between *Acacia rigens* and *A.* 'Gerang Gerung'. Branchlet cross-sectional shape had

been principal in the differentiation of the two entities (1). *Acacia* 'Gerang Gerung' was described as having more or less terete branchlets and obscure ribs, whilst *A. rigens* was noted to have strongly ribbed branchlets (VicFlora 2018; Maslin 2018). Although less clear, the nature of the branchlet indumentum was also identified as a possible delimiting character (2, 3, 4, 5); *A. rigens* was thought to possess shorter hairs, with the indumentum absent on branchlet ribs (NGW pers. obs.; VicFlora 2018).

Phyllode characters

Noted features of difference in the phyllodes between *Acacia* 'Gerang Gerung' and *A. rigens* were centred mainly around nervature (12, 13) and features of the indumentum (14, 15). The former was described as possessing eight distinct main nerves (Maslin 2018), while the latter was described as possessing multistriate (many fine nerves) nervature (Cowan 2001, 2018). Additionally, it was observed that the main nerves of *A. rigens* were less raised (Maslin 2018), with appressed-puberulous hairs (Cowan 2018), whereas *A.* 'Gerang Gerung' was noted to have distinctly sub-appressed hairs (VicFlora 2018). To account for differences in plant and phyllode maturity, phyllodes were measured for length, diameter and angle (6.5 mm along the phyllode from the node) at the seventh node from the mature distal phyllode (7, 9 and 10 respectively).

Inflorescence characters

Features of the inflorescence have long been used for the division of groups and species in *Acacia* (Bentham 1875; Pedley 1986; Maslin 2014; Maslin & Barrett 2014). Both the nature of the peduncle indumentum (17, 18) and number of flowers per capitulum (19) had proven useful for the separation of *Acacia rigens* from *A.* 'Gerang Gerung' in the past (VicFlora 2018; Maslin 2018).

Multivariate analyses

Ordination and cluster analyses were conducted in the PATN for Windows software v.3.02 (Belbin & Collins 2004) using our morphological data as input to assess the overall similarities and differences of each OTU. Range-standardisation was performed using the '(Value-Minimum Value) Range of Value' setting prior to the creation of a distance matrix, produced using the Gower metric association measure (Gower 1971). Cluster analysis was performed using the flexible unweighted

pair-group method with arithmetic mean (UPGMA) to construct a dendrogram. A semi-strong hybrid and multidimensional scaling (SSH MDS) ordination was conducted on the dataset, employing 50 random starts and 200 maximum iterations for the examination of the relative positions of OTUs in three-dimensional space. Kruskal-Wallis (KW) values were calculated for assessment of the significance of individual characters in the analysis, where a higher KW value indicates a greater contribution to group differentiation.

Survey of phyllode anatomy

Phyllode material taken from herbarium specimens was rehydrated for one day in water with a small amount of detergent added. Transverse hand sections were made from the mid-point of the phyllodes length and the sections were mounted using a basic wet mount. In total, five species were included in the survey, selected primarily on the current understanding of phylogenetic relationships of *Acacia rigens* (Mishler *et al.* 2014). Species selected on this basis were: *A.* 'Gerang Gerung', *A. rigens*,

Table 1. Twenty-six morphological characters scored from herbarium specimens for the current study.

	Branchlet characters
1.	Slightly ribbed (0) or strongly ribbed (1)
2.	Indumentum covering ribs (0) or between ribs (1)
3.	Indumentum density: moderate (0) or dense (1)
4.	Hair length minimum (mm)
5.	Hair length maximum (mm)
	Phyllode characters
6.	Pulvinus length (mm)
7.	Length of phyllode at 7th node (mm)
8.	Length of longest phyllode (mm)
9.	Diameter of phyllode (mm)
10.	Phyllode angle from stem at 6.5mm from node (degrees)
11.	Shape in cross-section: flat to compressed (0) compressed to subterete (2) subterete (3) subterete to terete (4) terete (5)
12.	Nerve number
13.	Nerves equal (in size, width and raising) (0) or some less prominent (1) (viewed under stereo microscope)
14.	Internerve indumentum: absent (0) sparse (1) moderate (3) dense (4)
15.	Primary nerve indumentum: glabrous (0) sparse (1) sparse to moderate (2) moderate (3)
	Inflorescence characters
16.	Peduncle length at anthesis (mm)
17.	Peduncle indumentum density: glabrous (0) glabrous to sparse (1) sparse (2) sparse to moderate (3) moderate (4) moderate to dense (5) dense (6)
18.	Peduncle indumentum: appressed (0) subappressed (1) hirsute (2)
19.	Number of flowers per capitulum
20.	Capitulum diameter at anthesis (mm)
21.	Bud shape: galeiform (0) elliptic (1) elliptic to ogive (2) ogive (3)
	Pod characters
22.	Length (mm)
23.	Width at narrowest point (mm)
24.	Width at widest point (mm)
25.	Distance between seeds (mm)
26.	Shape: submoniliform and curved (0) or submoniliform and curved to coiled (1)

A. enterocarpa and *A. calcicola*. Along with these species, *A. havilandiorum* was included to provide outgroup comparison and because of its superficial morphological similarity to *A.* 'Gerang Gerung' and *A. rigens*.

Results

Multivariate analyses

The dendrogram produced by cluster analysis resolved two clear groups consistent with our *a priori* hypothesis of the classification specimens into *A.* 'Gerang Gerung' and *A. rigens* (Figure 2). The separation of these groups was consistent in our ordination and was resolved with a stress value of 0.0885. For this method of ordination, any stress value less than 0.1 corresponds to a good representation of the data in ordination space, with no real prospect of drawing false inferences (Clarke 1993). Groups in the ordination were definitively isolated from one another across every dimension in multidimensional space (Figure 3).

Throughout the course of this study it was observed that a broad, flat phyllode variant of *A. rigens* exists from the Eyre Peninsula, S.A. Along with this, specimens from this region were noted to possess a greater number of nerves per phyllode. These individuals formed a group in the dendrogram (MEL2365638A, MEL0500676A, MEL0682135A, MEL1582539A, MEL0500677A) clustered within *A. rigens*. Further investigation of this group was beyond the scope of the present study, but a study incorporating more specimens from this area is proposed.

Survey of phyllode anatomy

Based on the characters illustrated from phyllode transverse sections (Figure 4), *Acacia* 'Gerang Gerung' bears most similarity to *A. enterocarpa,* and not *A. rigens* with which it was previously included. *Acacia rigens* possesses subsidiary nerves which, along with main and minor nerves, bring its nerve number to over 30. *Acacia* 'Gerang Gerung' and *A. enterocarpa* display 15 and 16 nerves respectively (including subsidiary nerves). These subsidiary nerves
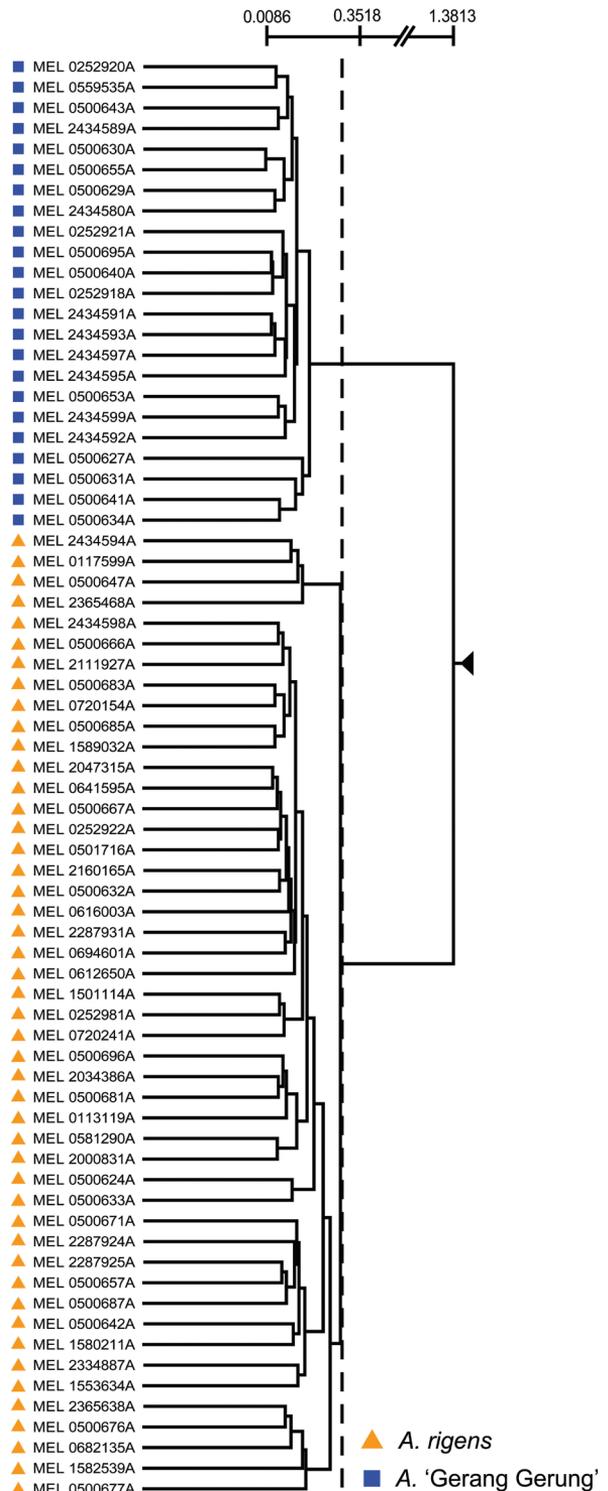

Figure 2. Dendrogram produced from analysis of *Acacia* 'Gerang Gerung' (*Acacia cineramis*) and *Acacia rigens* based on 26 characters. Scale of relative distance has been adjusted for publication (indicated by // on scale bar). MEL numbers provided as per AVH database entry.
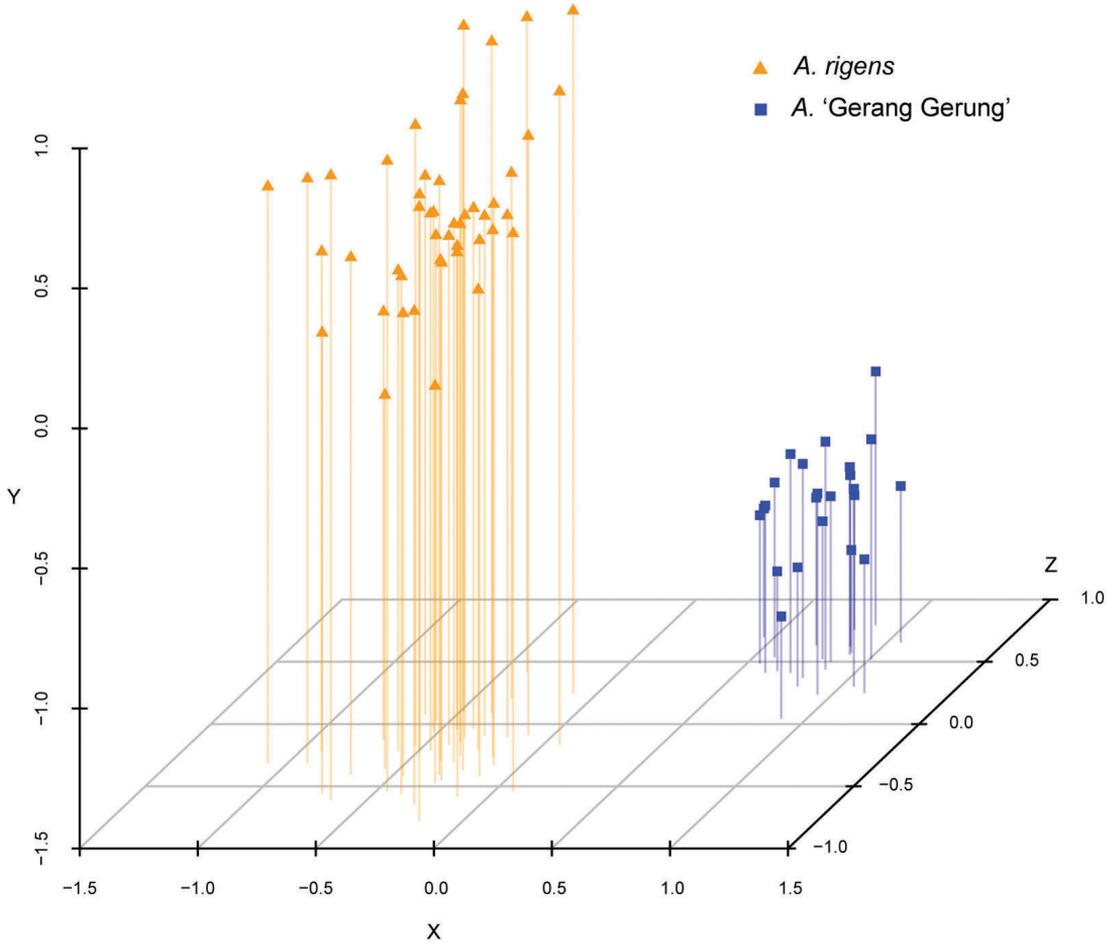
Figure 3. Three-dimensional ordination of all specimens included in study. Vertical drop lines are included to provide greater visualisation of spacing on the X-Z plane.
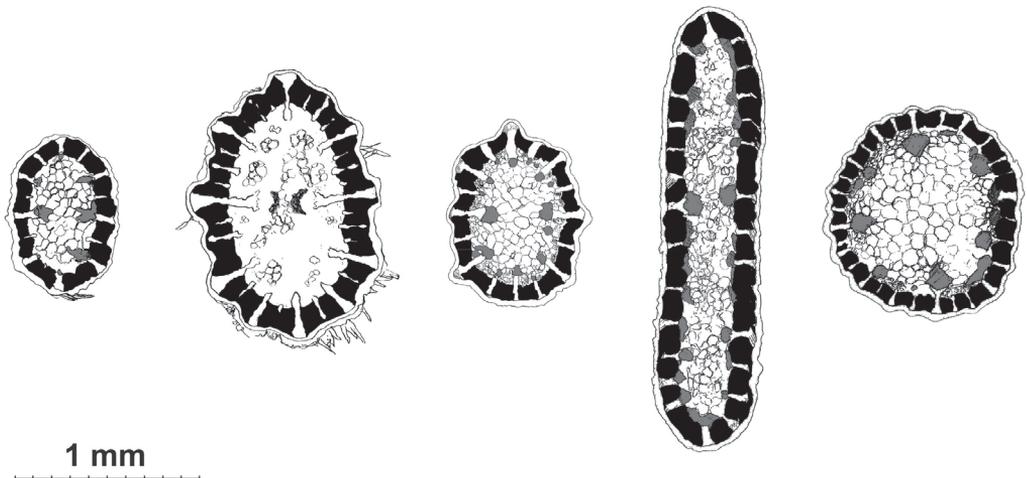

1 mm

Figure 4. Illustrations of transverse section of phyllodes showing palisade mesophyll (black) and vascular bundles (grey). Species are arranged by relatedness according to Mishler *et al*. (2014); (left to right) *Acacia cineramis*, *A. rigens*, *A. enterocarpa*, *A. calcicola*, *A. havilandiorum*.

appear undeveloped, are unobservable by eye or stereo microscope and do not connect to vascular bundles located in the spongy mesophyll (see *A.* 'Gerang Gerung' under *A. cineramis* in Figure 4). Aside from being significantly larger and more developed, nerves that are visible by eye and stereomicroscope generally abut vascular bundles within the spongy mesophyll.

Discussion

Given the consistency and extent of the separation between *Acacia rigens* and *A.* 'Gerang Gerung' in our analyses, as well as the long-standing acknowledgement of morphological discrepancies between the two entities, we consider it appropriate to propose the formal taxonomic recognition of *A.* 'Gerang Gerung' as a new species. This treatment is reflective of the level of morphological differentiation between *A.* 'Gerang Gerung' and *A. rigens* which is consistent with, if not greater than, the degree of morphological variation that has justified the designation of other new *Acacia* species (e.g. Maslin & Thomson 1992; Maslin 2014).

Kruskal-Wallis scores indicated that the most important characters for the delineation of groups were largely in line with the differentiating characters identified prior to this study, however, several novel characters have been discovered as useful (Table 2). Of these, phyllode nerve equality (character 13, KW: 69) was revealed as a key character in distinguishing the two

Table 2. Ten highest Kruskal-Wallis (KW) values across all scored characters used in the multivariate analyses. 'Used historically' denotes whether the character was used to distinguish *Acacia* 'Gerang Gerung' (*Acacia cineramis*) from *Acacia rigens* prior to this study.

Character	KW value	Used historically
13.	69.0	No
1.	69.0	Yes
2.	65.3	Yes
15.	63.6	Yes
14.	58.4	Yes
18.	51.7	Yes
12.	48.2	Yes
8.	43.8	No
7.	42.9	No
21.	31.1	No

species. All of the included pod characters produced statistically insignificant KW values, suggesting little difference in the fruits of the species. However, this result may be attributed to the limited availability of pods in the specimens used for analysis; only 26 out of 70 specimens included enough material for measuring pod characters. It is worth noting that some of the best characters for separating the species in the field received lower than expected KW scores due to small amounts of missing data. For example, character 19 (number of flowers per capitulum) was measured as discontinuous for both species (*A.* 'Gerang Gerung' possesses a maximum of 14 flowers per capitulum whilst *A. rigens* possesses a minimum of 20), however, the significance of this character in terms of KW value was lessened (KW: 26.5) by the fact that the character could not be measured for 22 out of 76 specimens. For this reason, KW values should not be taken as a definitive indicator of the usefulness of a character, and pod characters should not be dismissed as entirely uninformative; the collection of more pod data may reveal further differences between the species. Additionally, considering cases where *Acacia* species have been resolved as polyphyletic in molecular analyses (Brown *et al.* 2010), the relatively high level of morphological differentiation between *A.* 'Gerang Gerung' and *A. rigens* presents an appealing case for a molecular comparison of the two species in the future, as we hypothesise that these two taxa may not be sister species but rather each may be more closely related to other taxa.

The new species previously referred to as *Acacia* 'Gerang Gerung' is hereafter referred to as *Acacia cineramis*. The description of this new species is based on the measurements and scoring used for multivariate analyses in this study. Outlying measurements are denoted by brackets.

Phyllode nervature

Much of what is known about nerve development in *Acacia* phyllodes has been derived through morphometric taxonomic investigations and consists predominantly of observed trends in small subject groups (Gardner *et al.* 2005). This makes the application of these hypotheses across wider groups difficult, especially given the extent of variability and likely homoplasy identified in a large genus such as *Acacia* (Murphy *et al.* 2010). One hypothesised correlation

highlighted by Gardner *et al.* (2005) is between phyllode breadth and longitudinal nerve number in plurinerved species, where broader phyllodes possess a greater number of nerves (Boke 1940; Pedley 1978). Upon initial observation this relationship appears consistent with the variation observed during this study in *Acacia rigens* on Eyre Peninsula, however, further study is required to confirm if this is indeed the case.

Potentially the most notable observation made among species examined was the presence of undeveloped subsidiary nerves between developed nerves in *Acacia cineramis*, *A. rigens* and *A. enterocarpa* (Figure 4) which are unobservable by eye or under stereomicroscope. For example, *A. enterocarpa* displays 16 nerves in transverse section despite displaying only 10–12 visible nerves under stereomicroscope (as described in Cowan & Maslin 2018). These findings may be the result of within-species variation, however, it is more likely that there is an overall disconnect between 'visible nerve number' (i.e. the total number of nerves countable by eye or with a stereomicroscope) and 'actual nerve number' (i.e. the total number of nerves countable from phyllode transverse sections viewed under a compound microscope) for these species. Visible nerve number has long been used as a character for distinguishing various groups in *Acacia* with varying degrees of success. These results suggest that, as visible and actual nerve number do not necessarily align, considering only visible nerve number in the assessment of taxonomic groups and evolutionary relationships in *Acacia* may be insufficient and provide misleading taxonomic signals. Instead, both of these nerve number characters should be used in conjunction to provide a more accurate assessment of phyllode nervature in comparisons between groups, especially where the application of visible nerve number on its own is limited. In particular, consideration of subsidiary nerves may allow greater understanding of phylogenetic groupings of species in the 'plurinerved' clades of *Acacia* and perhaps provide additional morphological characters useful for distinguishing these clades, which to date have had limited morphological synapomorphies identified (Murphy *et al.* 2010). The mapping of both actual nerve number and visible nerve number onto molecular phylogenies at a broad scale has the potential to reveal possible patterns in phyllode nervature that have so far been unidentified, which may

in turn assist in further resolving relationships in the genus.

The shared similarity in nervature between *Acacia cineramis* and *A. enterocarpa* suggests a closer relationship between these two species than was originally thought and is further supported by the similarity of their stem and indumentum morphology, where *A. cineramis* is more similar to *A. enterocarpa* than to *A. rigens*, although the very stiff, conspicuously pungent phyllodes and the very distinctive flexuose fruit of *A. enterocarpa* clearly separate it from both *A. cineramis* and *A. rigens*. A molecular study of the relationships of these three species would enable the assessment of the hypothesis that *A. cineramis* is more closely related to *A. enterocarpa* than to *A. rigens* and, in turn, prove useful in assessing 'actual nerve number' as a character and concept. It is notable that in Victoria *A. enterocarpa*, like *A. cineramis*, is restricted to the Wimmera, with some populations being within 2 km of each other near Sandsmere.

Patterns in phyllode nervature possess remarkable potential for contributing to the morphological delineation of groups within *Acacia*, yet remain poorly understood due to the evolutionary complexity of the genus and the lack of a comprehensive body of work dedicated to the study of phyllode nerves. Our investigation into phyllode nervature should be considered preliminary in nature and serves to act as an expansion on the existing literature that focuses on the connection between phyllode nervature and taxonomy (e.g. Boke 1940; Boughton 1986; Gardner *et al.* 2005, 2008). It is hoped that this work may assist in informing future studies of phyllode nervature, such as anatomical surveys, which hold significant promise for *Acacia* taxonomy.

Taxonomy

Acacia cineramis H.K.Orel, **sp. nov.**

Acacia sp. aff. *rigens* (Gerang Gerung), Ross & Walsh (2003)

Acacia sp. Gerang Gerung (M.G.Corrick 6451) Vic. Herbarium, CHAH (2006)

***Type*:** **AUSTRALIA**. Victoria. Gerang Gerung, north of railway line. Small bushland reserve surrounded

by farmland, 5 October 1979, *M.G. Corrick 6451* (holo: MEL559535, iso: PERTH 00681482)

Spreading, sometimes erect single or multi-stemmed *shrub* 1–2 m high, 0.8–4 m wide; *branchlets* resinous, ±terete, slightly ribbed by excurrent ridges from phyllode bases (typically 3 ridges per phyllode), covered uniformly by an indumentum of white, appressed hairs 0.1–0.4 mm long. *Phyllodes* widely spreading, subterete to terete, shallowly incurved, (1–)2–4.5(–6.5) cm long, (0.6–) 0.8–1 mm wide, slaty-green (new growth bronze-green), apex acute, but hardly pungent, usually slightly asymmetric; distinctly 8-*nerved* (visible by eye or stereo microscope), nerves equal and raised (more so when dry) with sparse to moderate subappressed hairs like those of branchlets, the distinct nerves interspersed with indistinct subsidiary nerves (visible only under magnification in transverse section); *internerves* longitudinally grooved (more so when dry), mostly glabrous; gland basal, indistinct; *pulvinus* c. 1.2 mm long. *Inflorescences* simple, (1–)2(–4) per axil; *peduncles* 1.5–2(–3) mm long, indumentum of dense subappressed to spreading white hairs; *basal bracts* spathulate, indumentum subappressed-puberulous; *heads* globular, 2–5 mm diam., 10–14-flowered, yellow. *Buds* resinous, galeiform to ellipsoid. *Flowers* 5-merous; petals ovate, 1.3–1.5 mm long, glabrous, free; sepals obovate, 0.7–0.8 mm long, fimbriate distally, sometimes with a few appressed hairs, very shortly fused at base (for <0.05 mm). *Pods* submoniliform, curved to loosely coiled, 2–4 cm long, 0.7–2 mm wide, chartaceous to coriaceous, indumentum of sparse to moderately dense, appressed, white hairs. *Seeds* longitudinal, elliptic, 1.8–3 mm long, 1–1.6 mm wide, glossy, brown; *aril* white, apical, conical, 0.6–2 mm long.

Specimens examined: **VICTORIA**. Near Coker Dam, 29.ix.1895, *F.M. Reader s.n.* (MEL500655); Near Coker Dam, 29.xii.1895, *F.M. Reader s.n.* (MEL500641); Near Coker Dam, 29.xii.1895, *F.M. Reader s.n.* (MEL500630); Mallee District, x.1899, *St. E. D'Alton s.n.* (MEL500629); Dimboola, Wimmera, 12.xi.1899, *St. E. D'Alton 4* (MEL, AD); Dimboola, xi.1905, *St. E. D'Alton 255* (MEL); Jeparit-Lorquon, ix.1945, *A.C. Beauglehole 39851* (MEL); Big Desert, Parish of Murrawong, West Wimmera, 27.ix.1957, *A.J. Hicks s.n.* (MEL500634); Near Gerang [Gerung], 5.x.1957, collector unknown (MEL500640); Sandsmere Roadside Reserve, *A.C. Beauglehole 84095* (MEL); Cattle Dam Bushland Reserve, 16.ix.1986, *A.C. Beauglehole 84494* (MEL); Crown land adjacent to allotment 51, Parish Woraigworn, 10 km S of Gerang

[Gerung], 13.ix.1987, *C. Brownsea s.n.* (MEL252920); Sandsmere, 19.xii.2018, *H.K. Orel 1* (MEL); Sandsmere, 19.xii.2018, *H.K. Orel 3* (MEL); North of Little Desert, *H.K. Orel 5* (MEL); Little Desert NP northern edge, 20.xii.2018, *H.K. Orel 7* (MEL); Gerang Gerung, 20.xii.2018, *H.K. Orel 9* (MEL); Glenlee, 20.xii.2018, *H.K. Orel 11* (MEL); **SOUTH AUSTRALIA**. Murray Bridge, 17.x.1953, *R.I.M. Humphrey s.n.* (AD 98534178).

Distribution and ecology: Known only from extant populations in the Wimmera district of central-western Victoria. Historically recorded from Dimboola as far north as Lake Hindmarsh, with one outlying specimen recorded from Murray Bridge in South Australia (*R.I.M. Humphrey s.n.*, 1953, AD98534178). It is possible that the species' range extends, or once extended, that far west, however, the taxon was propagated at Wail nursery in the early 1950s (VicFlora 2018) and it is equally likely that this specimen was planted. Another outlying specimen recorded from the Big Desert Wilderness Area (*A.J. Hicks s.n.*, 1957, MEL500634) has not been confirmed by any recent collections and the record may be questionable. Surveys undertaken as part of this study were conducted in Victoria and have recorded the species from seven sites across an area of approximately 1145 kilometres bounded by the Little Desert to the south, Kaniva to the east, Gerang Gerung to the west and Glenlee to the north. The largest known population (~2500 individuals) occurs just inside the Eastern Block of Little Desert National Park while another significant population (>300 individuals) occurs within Glenlee Flora and Fauna Reserve. Population numbers in the five other sites from which it is currently known are very low (fewer than 15 individuals across all sites).

Acacia cineramis favours sandy clay loam soils overlying Tertiary Loxton Sand and is a co-dominant species in the two communities where its population is significant. From its current distribution and ecology, it seems likely that the species was distributed throughout the relatively fertile soils of the Wimmera and its range was bound in the north and south by the Big Desert and Little Desert respectively. Due to heavy clearing for cereal cropping throughout the Wimmera, it is likely that most of the species' habitat has been lost and the few remaining populations exist at or near the margins of cropping land. As such, the largest remaining population (occurring within the Little Desert Eastern Block) may not be fully representative of the 'preferred' habitat for the species. The Little Desert population is situated in

heathy mallee scrub and on slightly sandier soil than other occurrences. Associated species are *Eucalyptus phenax* Brooker & Slee, *Melaleuca uncinata* R.Br., *Daviesia pectinata* Lindl., *Acacia rigens* and *Eremophila gibbifolia* F.Muell. Another co-occurring wattle is *Acacia glandulicarpa* Reader, which is considered vulnerable (EPBC 1999). Glenlee Flora and Fauna Reserve may better represent the 'preferred' habitat of the species. Here it occurs in scrubby mallee woodland along with *Eucalyptus leucoxylon* F.Muell., *E. phenax* Brooker & Slee, *Allocasuarina luehmannii* (R.T.Baker) L.A.S.Johnson, *Acacia pycnantha* Benth., *A. acinacea* Lindl., *Dodonaea bursariifolia* F.Muell., *Ozothamnus retusus* Sond. & F.Muell. and *Lepidosperma viscidum* R.Br.

Conservation status: This species has been conservatively assessed as endangered (EN) according to IUCN (2012) criteria. This classification is based on the species' extent of occurrence (EoO; *ca.* 1145 km^2), area of occupancy (AoO; *ca.* 5 ha) and number of locations (as defined by IUCN 2012 – 1 across the species EoO). Also contributing to the classification is the estimation of continuing decline in EoO, AoO and quality of habitat. It is conceivable that a very large proportion (90% or more) of the original extent of the species has been lost to agriculture, but there is insufficient evidence in the form of herbarium specimens to support this. If this extent of loss of occurrence were included in the assessment, a risk code of critically endangered (CR) *sensu* IUCN (2012), could be supported. Five of the seven known occurrences of the species are along degraded roadside vegetation remnants adjacent to crops, and without intervention, the future of these populations (some of only a single, old plant) is insecure. Approximately 50% of the largest population (Little Desert) was infertile when visited in December 2018. This may be a seasonal response after a very dry spring, however, it may indicate that the species does not produce abundant seed and that the soil seed bank, if insubstantial, may render the
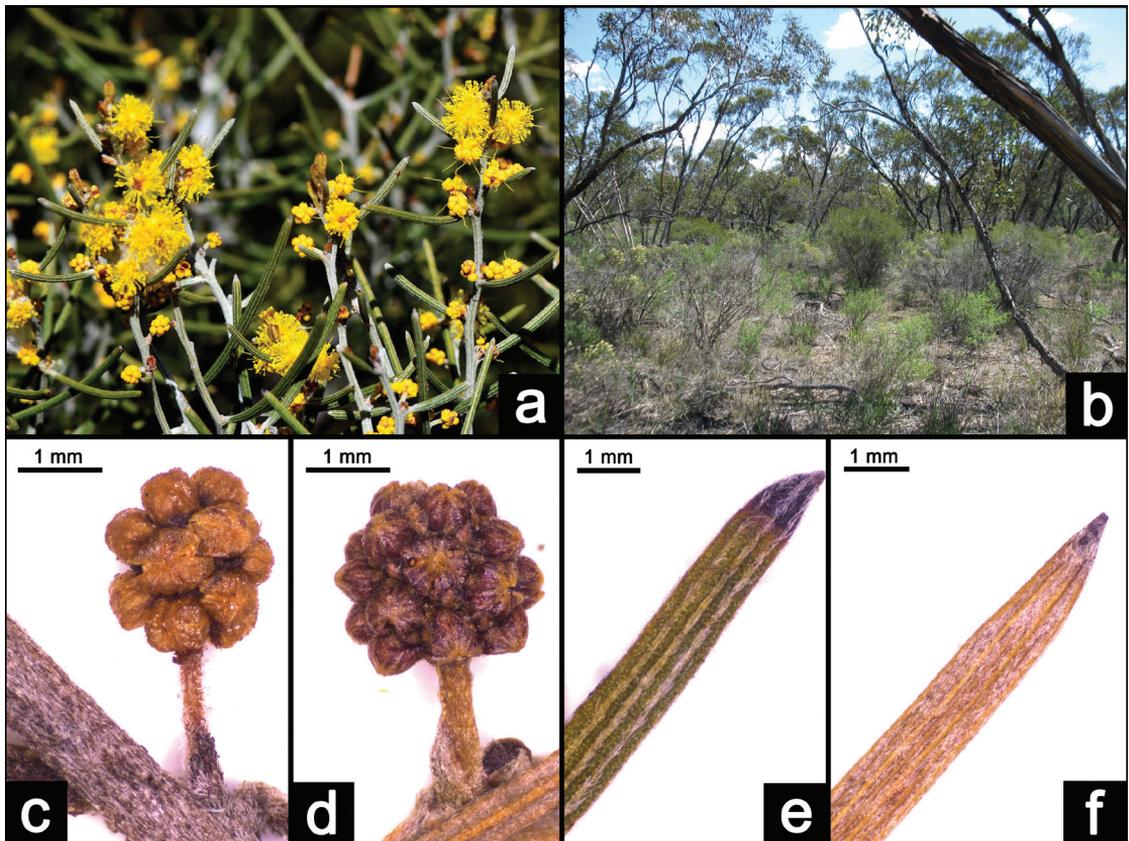

Figure 5. *Acacia cineramis* and *A. rigens*: a. Branchlets of *A. cineramis* with phyllodes and inflorescences (Photo: G. and M. Goods); b. *A. cineramis in situ*; c. Immature inflorescence of *A. cineramis*; d. Immature inflorescence of *A. rigens*; e. Phyllode of *A. cineramis*; f. Phyllode of *A. rigens*.

species vulnerable to repeated burning (a common management strategy in the Little Desert). Furthermore, some populations were observed to be heavily galled as a result of infection by a gall fungus, *Uromycladium* sp. This pathogen was observed affecting other *Acacia* species in the region and represents a threat to the species (Doungsa-ard *et al.* 2018), particularly when acting on smaller, more vulnerable populations in conjunction with other environmental stresses.

Etymology: The species name is compounded and derived from the Latin words *cinereus* (ash-coloured) and *ramus* (branch) in allusion to the appearance of the branchlets of the species – one of the more obvious characters that distinguishes the plant from *Acacia rigens*.

Notes: *Acacia cineramis* is distinguished from *Acacia rigens* by a number of distinctive features. In the field, *A. cineramis* is recognised by its slaty-green foliage and bronze-green new growth. The phyllodes of *A. rigens* are generally a brighter green but the new growth is of a similar colour to that of *A. cineramis* (Figure 6). While not retrieved as a primary character in the analysis, the orientation of the phyllodes on live plants may be a useful character: those of *A. cineramis* being widely spreading from the stems and those of *A. rigens* being very acutely inclined to stems in areas where the species co-occur. The branchlets of *A. cineramis* are very indistinctly ribbed and uniformly covered by a dense indumentum of closely appressed white hairs, whereas *A. rigens* displays strong yellow ribs that are more or less glabrous, with the branchlet indumentum occurring only between the ribs. As a consequence, the branchlets of *A. cineramis* are significantly whiter or greyer in colour than those of *A. rigens*. Perhaps the most reliable character in distinguishing between these species from dry specimens is the difference in phyllode nervature; *A. cineramis* is characterised by having eight prominent, raised nerves, often with a glabrous longitudinal groove between each nerve and subappressed hairs on the nerves (Figure 5e). The phyllodes of *A. rigens* possess significantly more nerves (from 14 to 29) which are less prominent and most commonly have subappressed hairs between the nerves (Figure 5f). Phyllodes of *A. cineramis* are generally shorter than those of *A. rigens* and reach a maximum length of 6.5 cm versus a maximum recorded length of 16 cm for the latter species. Differences in the inflorescence are also conspicuous, most notably in the number of flowers per capitulum (*A. cineramis* is 10–14-flowered, *A. rigens* is 20–33-flowered), and in the shape of the mature buds of individual flowers; *A. cineramis* possesses rounded, galeiform buds, whereas *A. rigens* displays buds that

Figure 6. Habit of *Acacia rigens* (left) and *A. cineramis* (right) in the field.

most commonly taper apically to a point (i.e. ogive-shaped). This latter feature results in the immature capitula of *A. rigens* generally appearing stellate or spiky, while those of *A. cineramis* appear more bulbous (Figure 5c & Figure 5d). Several other diagnostic features of *A. cineramis* are apparent, however their use may be limited in the field given their nature as microscopic characters. These include differences in the peduncle indumentum (which is dense and subappressed to spreading), branchlet hair length (hairs 0.1–0.4 mm long), and phyllode nerve equality (where in *A. cineramis* all visible nerves are equal).

Acknowledgements

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