
Taxonomy and evolutionary studies on the genus *Psoralea* L. (Psoraleeae, Fabaceae)

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Thesis presented for the degree of Doctor of Philosophy in the Department of Biological Sciences, University of Cape Town

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December, 2015

DEDICATION

This study is dedicated to my late father, Alhaji Muhammadu Bello Sule who answered the call of his Lord on the cold morning of Tuesday, 2nd June, 2014 for his patience especially during his trying times on his sick bed when he was extremely in need of my presence but could not be with him as a result of this study. **My Lord! Bestow on him Your Mercy as he did bring me up when I was young.**

DECLARATION

I Abubakar Bello, hereby declare that the work on which this thesis is based is my original work (except where acknowledgements indicate otherwise) and that neither the whole nor any part of it has been, is being, or is to be submitted for another degree in this or any other university. I authorise the University of Cape Town to reproduce for the purpose of research either the whole or a portion of the content in any manner whatsoever.

This thesis includes three publications that were published in collaboration with my supervisors and research associates. My supervisors testified that I made substantial contributions to the conceptualisation and design of the papers; that I independently ran experiments and wrote the manuscripts (the first two manuscripts below), with their support in the form of comments and suggestions (see Appendix D).

The published papers:

- **Bello A.**, Daru B.H, Stirton C.H., Chimphango S.B.M, van der Bank M., Maurin O., and Muasya A.M. (2015). DNA barcodes reveal microevolutionary signals in fire response trait in two legume genera. AoB PLANTS, 7: plv124; doi:10.1093/aobpla/plv124. (Chapter 3).
- **Bello A.**, Stirton C.H., Chimphango S.B.M., Muasya A.M. (2015). *Psoralea diturnerae* and *P. vanberkelae* (Psoraleeae, Fabaceae): two new species restricted to the Core Cape Region of South Africa. PhytoKeys, 44: 97–107. doi: 10.3897/ phytokeys.44.8999. (Part of Chapter 4).
- Stirton C.H., Stajsic V., **Bello A.** (2015). Naturalised species of *Psoralea* (Fabaceae: Psoraleeae) in Australia. Muelleria, 33: 97−107. (Part of Chapter 4).

ACKNOWLEDGEMENTS

All thanks and glorifications are due to Allah (SWT) Who sustain my life to this day and endowed me with the strength and the ability to accomplish this study.

I thank Prof. A. Muthama Muasya for taking the risk of accepting me as a candidate for this study at a time when everyone is curious as to whom to employ as a good research candidate in a highly competitive academic arena. I also thank my Co-Supervisor Dr. S.B.M. Chimphango for his fatherly advice and critical challenges on my work, making sure that I did things in a right and orderly manner. I would also like to express my sincere gratitude to my external supervisor, Prof Charles H. Stirton for his mentorship, guidance, contributions and training both in the field, lab and in the write up.

The tuition and upkeep for this study were funded by Nigeria Tertiary Education Trust Fund (NTETF) and Umaru Musa Yarádua University Katsina, Nigeria and a top up from the J. W. Jagger Centenary Gift Scholarship from the University of Cape Town. The study was supported by grants from the South African National Research Foundation (NRF; AMM). Without the income provided by these funds, this study would not have been possible.

This thesis has had mentorship and various forms of support from numerous outstanding individuals both from within UCT and outside. I extend my heartfelt gratitude to these individuals for without them, this thesis would not have been completed. These individuals include: Dr Barnabas H. Daru, Meshack M. Dludlu, Jack Viljoen, Dr Tracey Nowell, Hassan Sadiq (UCT Statistics), Dr Benny Lemaire, Prof Michelle Van der Bank and Dr Olivier Maurin. I also thank Dr Ashley N. Egan for her tremendous help in the edit of the phylogeny chapter.

The fieldwork for this research was permitted by Cape Nature (permit 0028-AAA008-00035) and the Eastern Cape Department of Economic Development, Environmental Affairs and Tourism (No CRO. 153/13CR). I am indebted to the superb hospitality of Di and Bill Turner, over a number of visits, of the Outramps Group of the Custodians of Rare and Endangered Wild Flowers (CREW) from George, and Nicky van Berkel, without whose efforts, two of my new species might not have been discovered.

I thank the Curators and Staff of BOL, NBG and PRE herbaria particularly Terry Trinder-Smith, Dr Cornelia Klak, Dr Anthony R. Magee and Dr Dee Snijman. I also thank the Department of Biological Sciences of UCT and my fellow students for their company and support. I thank the staff and members of CREW (Custodians of Rare and Endangered Wildflowers (CREW) Programme, especially Mr. Ismail Ibrahim and Vathiswa Zikishe for field trip opportunities and friendship.

I want to truly express my sincere appreciation and gratitude to my parents, for my upbringing and prayers. There are no words to convey how much I love them. I would like to thank my wife, Mrs. Maryam Abdulkadir, for her patience, love and strength in taking over all the house activities and the training of the kids while I am undertaking this study for almost four years Away from home in Nigeria. For my kids, Fadila, Ahmad and Hafsat, I thank you very much for pressurising me to come back. This really gingered me towards putting more efforts and working harder to accomplish the mission. I thank my friend Buhari Yusuf Shinkafi for taking most of my responsibilities while I am away. There are no words to express my appreciations to him. I pray for Allah's guidance and assistance on your PhD journey.

Lastly to all those who helped in one way or the other whose names were not mentioned here due to shortage of space and time, I say a big thank you to all of you.

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LIST OF ACRONYMS AND ABBREVIATIONS

agnes	Agglomerative Nesting
AIC	Akaike information criterion
ANOVA	Analysis of variance
BBM	Bayesian Binary MCMC
BEAST	Bayesian Evolutionary Analysis Sampling Trees
BEAUti	Bayesian Evolutionary Analysis Utility
BI	Bayesian inference
BOL	Bolus herbarium
BOLD	Barcode of life Data Base
BS	Bootstrap
CA	Cluster analysis
CBOL	Consortium for the Barcode of Life
CCDB	Canadian Centre for DNA Barcoding
CCR	Core Cape Sub-Region
CFR	Cape Floristic Region
CI	Confidence interval
CTAB	Cetyltrimethylammonium Bromide
CV	Coefficient of variation
DNA	Deoxyribonucleic acid
diana	Divisive Analysis Clustering
EOO	Extent of occurrence
ETS	External transcribed spacer
fanny	Fuzzy Analysis Clustering
GCFR	Greater Cape Floristic Region
GTR	General time reversible
HPD	Highest posterior density
ITS	Internal transcribed spacer
IUCN	International Union for Conservation of Nature and Natural Resources
LC	Least concern
LPWG	Legume Phylogeny Working Group
ML	Maximum likelihood
MN	Mediterranean North
MOTU	Molecular operational taxonomic unit

MP	Marginal probabilities
MRCA	Most recent common ancestor
MS	Mediterranean South
MYA	Million years ago
MYO	Million years old
NAm	North American
NRF	National Research Foundation
NT	Near threatened
NTETF	Nigeria Tertiary Education Trust Fund
PAM	Partition around medoids
PCA	Principal component analysis
PCoA	Principal coordinates analysis
PCR	Polymerase chain reaction
PP	Posterior probability
RASP	Reconstruct Ancestral State Phylogeny
RAxML	Randomized Axelerated Maximum Likelihood
S	Succulent/Xeric shrublands
SAf	Southern African
SAm	South American
SD	Standard deviation
TG	Temperate Grassland
TF	Temperate Forest
UCT	University of Cape Town
UMYU	Umaru Musa Yarádua University
UPGMA	Unpaired group method of arithmetic averages
VU	Vulnerable

THESIS SUMMARY

Psoraleeae is a tribe of the papilionoid legumes in Fabaceae comprising ca. 223 species in nine genera. Members of Psoraleeae are distributed worldwide, though they mainly occur in the temperate biomes. Of these, ca. 60% of the species (mostly in *Otholobium* and *Psoralea*) are endemic to southern Africa predominantly in the Greater Cape Floristic Region (GCFR). The genus *Psoralea*, consisting of 75 species endemic to southern Africa, is the second most speciose legume in the GCFR after *Aspalathus* (280 species). This thesis, consisting of a literature review, three research chapters and synthesis, studies the taxonomy, evolutionary history and biogeography of Psoraleeae with an emphasis on *Psoralea*.

In Chapter 2, I infer phylogenetic relationships and reconstructed the biogeographical history of Psoraleeae using eight DNA markers (one nuclear and seven plastid). The objective was to determine the monophyly, phylogenetic positions, origins, divergence dates and evolution of characters in the Old World Psoraleeae, i.e. the African (*Cullen*, *Otholobium* and *Psoralea*) and European (*Bituminaria*), and the New World Psoraleeae, i.e. the American (*Hoita*, *Rupertia*, *Ladeania*, *Orbexilum*, *Pediomelum* and South American *Otholobium*) and Australian (*Cullen*). Datasets were analysed using maximum likelihood (RAxML) and Bayesian MCMC (MrBayes, BEAST and RASP) approaches. The monophyly of Psoraleeae and the phylogenetic position of African Psoraleeae (*Otholobium* and *Psoralea*) as sister to the rest of Psoraleeae are supported. The phylogenetic position of *Psoralea* as a genus is maintained due to a strong nodal support and its multiple synapomorphies. However, the monophyly of the North American Psoraleeae is not supported. The tribe Psoraleeae most likely evolved in the Mediterranean South biome of southern Africa (Marginal Probability = 53.4%) in the late Miocene at ca. 8.2 million years ago (mya) and then dispersed into the Succulent biome of the Americas, Mediterranean North biome of the Mediterranean region, Temperate Grassland biomes of Africa and Australia to the Temperate Forest biome of Asia (all key nodes have marginal and posterior probabilities for ancestral area and biome to be > 50% and 0.7 respectively). Within the GCFR, *Psoralea* is characterised by a rapid recent radiation, diversifying since 4 mya within the fynbos biome where it commonly occupies mountain seepages and exhibits both reseeding and resprouting life forms. Climate changes in the late Miocene, changes induced by glaciation during the Quaternary and environmental heterogeneity are proposed as the major drivers of speciation in *Psoralea* and Psoraleeae.

In Chapter 3, I evaluate the utility of DNA barcoding in identifying and detecting phylogenetic signals in fire response traits (reseeding and resprouting) at species and generic levels in the recently diverged *Otholobium* and *Psoralea* using the core DNA barcode markers (*matK* and *rbcL*). The dataset exhibited a barcoding gap, with a combination of *matK* and *rbcLa* (*matK* + *rbcLa*) being identified as a better barcode than the individual regions. Within the two genera, 100% of individuals were correctly identified to their respective genera, but identifying them correctly to species level was less accurate (<50%). A weak match (54%) was found between genetic species and morphologically-delimited species. I detected the presence of phylogenetic signals in fire response traits, which provides evidence for a convergent origin of the reseeding trait. The different lineages showed a weak but significant phylogenetic conservatism in their response to fire as reseeders or resprouters, with more clustering of resprouters than would be expected by chance. This chapter shows that DNA barcoding may be useful in identification of SAf Psoraleoid genera (*Otholobium* and *Psoralea*) and in inferring the impacts of recurrent fires on gene flow in resprouting and reseeding taxa in the Cape Floristic Region.

In Chapter 4, I revise the taxonomy of the *Psoralea pinnata* species complex using multivariate techniques to test for discrete clustering of samples that represented species recognised using one or a few characters. The objective was to determine whether the diversity that exists within the *P. pinnata* complex is a depiction of several local forms of a single variable species as proposed by Harvey (1862) or one which comprises several distinct species. The results support the recognition of 25 distinct species, of which 14 are described here as new: *P. azuroides*, *P. brilliantissima*, *P. elegans*, *P. floccosa*, *P. imminens*, *P. intonsa*, *P. ivumba*, *P. kougaensis*, *P. montana*, *P. muirii*, *P. rhizotoma*, *P. semota*, *P. sordida*, and *P. suaveolens*. The taxon *P. pinnata* var. *latifolia* Harv. is raised to species rank as *P. latifolia*.

This thesis contributes to science by generating new knowledge on the taxonomy, evolution and biogeography of the charismatic but poorly understood African representatives of the tribe Psoraleeae. The description of new species in South Africa and the report on a previously unrecorded invasive species in Australia adds to the understanding of biodiversity and provides valuable information for conservation managers and policy makers.

CHAPTER ONE: GENERAL INTRODUCTION AND OBJECTIVES

1.1. Introduction

Fabaceae, or Leguminosae (bean or pea family), is the third largest family of flowering plant in the world, with ca. 751 genera and 19 500 species (Legume Phylogeny Working Group (LPWG) 2013). Members of Fabaceae are of great economic importance and are widely distributed across all biomes of the world (Schrire *et al.* 2005a & b). Fabaceae consists of three subfamilies (Caesalpinioideae, Mimosoideae, and Papilionoideae or Faboideae) and most of its species (ca. 14 000) are in Papilionoideae. The phaseoloid clade represents one of the largest groups in Papilionoideae with 114 genera and ca. 2 000 species (Lewis *et al.* 2005; Stefanovic *et al.* 2009; Li *et al.* 2013). Psoraleeae, one of the phaseoloid lineages, is distributed worldwide, mainly in the temperate biomes (Stirton 2005). Psoraleeae has ca. 223 species in nine genera (*Otholobium* C.H.Stirt., *Psoralea* L., *Pediomelum* Rydberg, *Orbexilum* Raf., *Rupertia* Grimes, *Hoita* Rydberg, *Ladeania* A.N.Egan & Reveal, *Cullen* Medik. and *Bituminaria* C.H.Stirt.), of which 60% are in *Otholobium* C.H.Stirt. and *Psoralea* L. The genus *Psoralea*, consisting of 75 species, is the second most speciose legume in the Greater Cape Floristic Region (GCFR) of southern Africa after *Aspalathus* L. (Manning & Goldblatt 2012; Snijman 2013).

1.2. Evolution and biogeography of legumes

Legumes are thought to have evolved either in the humid tropics of western Gondwana in the late Cretaceous or more recently in northern Laurasia (Schrire *et al.* 2005a & b). Based on their distribution, the boreal hypothesis has suggested that legumes dispersed northwards from the Americas, through the boreotropics and down into Africa and Asia (Schrire *et al.* 2005a & b). After an extensive analysis of molecular data and some well characterised fossils, Lavin *et al.* (2005) dated the evolution of legumes to be ca. 60 mya. Using the distribution range of legumes across major world biomes, Schrire *et al.* (2005a & b) hypothesised that legumes originated in a shallow stretch of salt water, which split the two main groups of land masses in a region to the north of the Tethys Sea in a seasonally deciduous dry forest (succulent biome), and later dispersed into other biomes. Details on the evolution of Psoraleeae and *Psoralea* are discussed in Chapter 2.

Unravelling distribution patterns in plants has been one of the primary focuses of biogeographical studies. Extinction (range compression), vicariance (split into discontinuous parts) and long distance dispersal (extension of range due to geographic dispersal) have been known to influence intercontinental disjunctions (Mansion *et al.* 2008). Biogeographical reconstructions using phylogenetic approaches to demonstrate the evolutionary histories of organisms in space and time are currently gaining much attention among biologists. Model-based and event-based approaches have been used recently for biogeographical inferences.

1.3. Psoraleeae in the Greater Cape Floristic Region

The naming of taxa in the tribe Psoraleeae started with the description of the genus *Psoralea* by Linnaeus in the *Species Plantarum* (Linnaeus 1753). *Psoralea* was later re-classified into various genera in different parts of the world as more taxa were discovered and described in the subsequent 250 years. The currently circumscribed species of most southern African Psoraleeae (*Otholobium* and *Psoralea*) exhibit a wide diversity of growth forms that are rarely matched by other legume genera within the GCFR. Although the majority could be described as shrubs, some grow into big trees up to 10 m tall, while others are low creeping herbs. Psoraleeae represent most of the growth forms found in fynbos with the exception of succulent life-styles, although the sand dune species *Psoralea repens* Berg. often has quite fleshy leaves. The species richness of *Otholobium* and *Psoralea* is perhaps the most amazing given that the majority of the species are not wind-pollinated and the floral morphology is almost entirely the same across most of the papilionoid legumes (see Stirton *et al.* 2015). The majority of the species in fynbos have seeds that are either wind or ant-dispersed (Linder 1985). All Psoraleeae species have hard dry seeds, which indicate that they are dispersed by wind or water as most of the species occur along the riverine areas and wet mountains. Therefore, it reflects the patterns found in most other fynbos genera.

The GCFR extends in a broad L-shape from the mountains of Namaqualand in the north to Port Elizabeth, with some outliers in the hills towards Grahamstown, in the east. Five biomes are represented within this geographical area (Cowling 1992; Linder & Verboom 2015), but it is dominated by the fynbos biome, which has a distinct, sclerophyllous, heath-like vegetation (Manning & Goldblatt 2012; Linder & Verboom 2015) and which is almost entirely restricted to the GCFR. Fynbos vegetation is characterised by the presence of Restionaceae, Ericaceae and Proteaceae (Manning and Goldblatt 2012).

Otholobium and *Psoralea* species are found throughout the extent of the GCFR. They grow almost exclusively in fynbos, although some also occur in the transition zones between fynbos and other biomes. Within the fynbos biome, they are everywhere from coastal sand plains to the peaks of mountains (2325 m Seweweekspoort Mt.). Certain species require their roots to be constantly immersed in water while others exist in the rock crevices of mountains along the arid edge of the Karoo. The majority of *Psoralea* species are found in acidic, nutrient-poor, sandstone-derived soils characteristic of the fynbos or in the richer shale soils on mountain slopes. Outside the GCFR, *Psoralea* species are almost entirely restricted to the afromontane heathlands that span from the southern Drakensberg to as far north as St Helena Island in the Atlantic Ocean, although they occasionally occur on the low-altitude sandstones of the Eastern Cape and Kwazulu-Natal or the dolerites of the Great Escarpment. Two *Otholobium* species extend northwards to Zimbabwe, Malawi, Tanzania and Kenya, but the eastern African species are usually restricted to mountain peaks (Stirton 1989). Other legume genera co-occurring with *Otholobium* and *Psoralea* in the GCFR include: *Aspalathus* L., *Crotalaria* L., *Lotononis* (DC.) Eckl. and Zeyh., *Lebeckia* Thunb., *Calobota* Eckl. and Zeyh., *Wiborgia* Thunb. and *Wiborgiella* Boatwr. and B.-E. Van Wyk (Crotalarieae; Boatwright *et al.* 2008a & b, 2009), *Indigofera* L., (Indigofereae), *Amphithalea* Eckl. & Zeyh., *Podalyria* Willd., *Virgilia* Poir (Podalyrieae), *Lessertia* DC., (Astragaleae) *Argyrolobium* Eckl. & Zeyh. (Genisteae) and *Hypocalyptus* Thunb. (Hypocalypteae).

1.4. DNA barcoding and its applications

Identification, naming and classification of organisms are traditionally based on morphology (Linnaeus 1758, 1759). However, in an attempt to avoid the restrictions of relying soley on morphology, modern taxonomy uses molecular data, such as gene sequences, to make species identification fast, reliable and more accurate (Kress *et al.* 2014). DNA barcoding is the use of a short gene-sequence (DNA barcode) for species identification (Kress *et al.* 2014). The sequence of an unknown specimen is compared to barcodes in a sequence library of known species (Kress & Erickson 2007). DNA barcode sequences are mainly used for identification but are not intended for phylogenetic reconstruction (Kress & Erickson 2007) or as the only criterion in describing new species (Kress *et al.* 2014). DNA barcoding has many applications: enabling and facilitating identification when traditional methods are unrevealing; providing new technology that can be applied in the field to identify specimens; and providing evolutionary insights (Kress *et al.* 2014). DNA barcoding has been successfully implemented in phylogenetic reconstructions even though this was not its original purpose. Kress *et al.* (2009, 2014), for instance, reconstructed community- and species-level phylogenies using super-matrices of barcoding regions.

The Consortium for the Barcode of Life (CBOL 2009) Plant Working Group proposed the use of *rbcL* and *matK* as universal plant barcoding regions. However, due to a lack of universality, poor sequence quality and limited discriminatory power, a combination of the two regions (*rbcLa* and *matK*) was later on recommended (CBOL 2009). Poor resolution and identification success of the core barcoding regions in some plant groups has led to a new suggestion for the addition of the nuclear ribosomal DNA (ITS) region to the core barcoding regions (*rbcLa* and *matK*) to improve identification success (Hollingsworth 2011).

Species with recent divergence times such as those in the tribe Psoraleeae remain a big challenge for DNA barcoding despite its huge success in many plants groups (Bello *et al.* 2015a). Such species lack sufficient characters to separate them from their close relatives which is probably due to limited time to accumulate mutations among the nucleotide sequences. These species lack a barcode gap and have ambiguous barcode matches or a lack of barcode clusters on tree-based analyses. The utility of DNA barcoding in identifying and detecting phylogenetic signals in recently diverged *Otholobium* and *Psoralea* are explored in Chapter 3.

1.5. The genus *Psoralea* L.

The name *Psoralea*, first formulated by Van Royen (1740), is derived from the Greek name 'psoraleos' meaning 'scurfy' or 'scabby', in reference to the resinous, black glandular dots found on the foliage of most of the species. *Psoralea* is mostly constrained to wet mountains and lowland fynbos of the Core Cape Sub-Region (CCR) with outliers in eastern and Afromontane regions to Angola (Stirton & Schutte 2000, 2012). A few specialised species have adapted to drier regions (renosterveld, arid fynbos, and sand dunes) and can be found in the Sandveld, Klein Karoo and Namaqualand. The majority of the species are distributed on low nutrient soils derived from Table Mountain sandstone with a few habitat specialists that occur on shales, silcrete and limestone. A few species of *Psoralea* have invaded some parts of the world such as Australia, New Zealand and the United States (Stirton *et al.* 2015).

Psoralea pinnata L. is the type species of *Psoralea*. It is an erect, pinnately-leaved shrub or tree up to 5 m tall bearing either blue, mauve, purple, lilac or white flowers that bloom from October to April (Harvey 1862; Forbes 1930). All *Psoralea* species bearing (3)5–11(19)-foliolate leaves tend to be lumped together as the *Psoralea pinnata* species complex. This complex is thought to contain at least 28 species among which only 10 have been described formally. The complex includes all members of the genus *Psoralea* that fall within the broad concept of *P. pinnata* as described by Linnaeus (1753). Harvey (1862), who saw very few of the elements in the field and based his delimitation on herbarium specimens, proposed that the diversity that exists within the *P. pinnata* complex is a depiction of several local forms of a single variable species. Details on the problems surrounding the *P. pinnata* complex and reasons for making the complex a taxonomic focus are given in Chapter 4.

1.6. Statement of the problem

The current phylogenetic relationships within Psoraleeae are not clear. Egan & Crandall (2008a & b) placed southern African Psoraleeae as sister to the rest of Psoraleeae, while Dludlu *et al.* (2013) placed it as a sister to the North American genus *Hoita* in an unsupported relationship. Similarly, the generic and species boundaries between and within the southern African Psoraleeae (i.e. the genera, *Otholobium* and *Psoralea*) are not well resolved (Dludlu *et al.* 2013). The genus *Psoralea* is nested within the southern African *Otholobium* (Dludlu *et al.* 2013). In addition, the phylogenetic position of the African, Asian and European *Cullen* Medik. species has never been studied.

The taxonomy of the genus *Psoralea* was last critically revised nearly a century ago by Helena Forbes (Forbes 1930). Since then, many new species have been added to the genus making it very speciose, diverse and unwieldy. At the establishment of the genus *Psoralea sensu stricto* by Stirton (1981), 20 species were recognised, all of which occurred in South Africa, with a centre of diversity and endemism in the GCFR. As Stirton did more field work in an attempt to revise the genus, he discovered many more species. When he completed the revision of the genus *Otholobium* (Stirton 1989), he pointed out that the genus *Psoralea* had at least 47 species. In a later publication (Stirton 2005), he alluded to the possibility that there could be ca. 51 species of *Psoralea*. However, with the exception of the validly published species (Stirton 1983, 1984, 1995, 1998, Stirton *et al.* 2011, 2012, Bello *et al.* 2015b, Dludlu *et al.* 2015), the majority of the species are presently known by informal names in various herbaria and manuscripts that describe the flora of the region (e.g. Manning & Goldblatt 2012). The application of these names is always ambiguous and subjective due to a lack of clarity on species delimitation since such species have no formal description.

1.7. Aim and objectives of the thesis

The overall goal of this thesis is to investigate the processes that have led to the evolution and diversification of Psoraleeae with an emphasis on *Psoralea* by means of comparative analyses based on phylogenetic hypotheses. This involves studying the taxonomy, reconstructing phylogenetic histories and studying biogeographic distributions of the various genera of Psoraleeae within the GCFR and other parts of the world. In addition, the thesis evaluates the evolutionary pattern of the GCFR lineages for their affinity to fire both as reseeders or resprouters at species and generic levels, and finally revises the nomenclature and species delimitation within *Psoralea pinnata* L. species complex. The study has the following general objectives:

1. To infer the evolutionary and biogeographic histories in the tribe Psoraleeae.
2. To assess the efficacy of the universal plant DNA barcoding regions in species identification and evaluation of phylogenetic signals in fire response traits in recently diverged *Otholobium* and *Psoralea*.
3. To revise the taxonomy of the *Psoralea pinnata* complex.

1.8. Structure of the thesis

The research chapters in this thesis are presented as a series of papers for publication in peer reviewed journals. This first chapter has presented a general introduction and the objectives of the thesis. Chapter 2 addresses questions on the systematics of Psoraleeae, specifically the monophyly, phylogenetic position, origin, divergence dates and evolution of characters in the Old World Psoraleeae, i.e. African (*Cullen*, *Otholobium*, and *Psoralea*) and European (*Bituminaria*), and the New World Psoraleeae, i.e. American (*Hoita* Rydberg, *Rupertia* Grimes, *Ladeania* A.N.Egan & Reveal, *Orbexilum* Raf., *Pediomelum* Rydberg and South Amercian *Otholobium*) and Australian (*Cullen*). Chapter 3, already published as a journal paper (Bello *et al.* 2015a), explores the utility of the DNA barcoding technique as a modern tool for identifying biological specimens and managing species diversity and delimitation in the recently diverged southern African Psoraleeae (*Otholobium* and *Psoralea*). The chapter also assesses the role of ecological divergence on the lineages in relation to microevolutionary signals in fire response traits (reseeding and resprouting) in a fire prone CFR. Chapter 4 revises the taxonomy of an enigmatic complex in the genus *Psoralea*, the *Psoralea pinnata* complex. Two taxonomic papers have already been published (Bello *et al.* 2015b; Stirton *et al.* 2015) out of this chapter. The objective was to determine whether the diversity that exists within the *P. pinnata* complex is a depiction of several local forms of a single variable species, as proposed by Harvey (1862), or one which comprises several distinct species. Finally, Chapter 5 synthesises the whole thesis and provides a general conclusion.

CHAPTER TWO: PHYLOGENY, BIOGEOGRAPHY AND CHARACTER EVOLUTION IN PSORALEEAE (FABACEAE)

Abstract

Psoraleeae consists of ca. 223 species in nine genera, distributed worldwide but largely in the temperate biome. Here, I reconstruct the phylogenetic history of Psoraleeae using eight molecular markers (*rpl32-trnL*, *trnQ-rps16*, *trnS-trnG*, *matK*, *rbcLa*, *rpoB-trnC*, *trnL-trnF* and ITS) to test the monophyly and previous hypotheses on generic relationships and to reconstruct the biogeographical history (distribution pattern, potential ancestral area and biome). I found that the monophyly of Psoraleeae is supported. The African *Otholobium* and *Psoralea* formed a strongly supported clade that is sister to the rest of Psoraleeae. However, *Otholobium* is not monophyletic; the American taxa of the genus are resolved as closer to *Bituminaria*, whereas *Psoralea* is a clade derived from within the African *Otholobium*. Recognition of *Psoralea* as a genus in Psoraleeae is maintained due to a strong nodal support and its multiple unique synaphomorphies. An expanded sampling of *Cullen* revealed a basal Old World grade comprising Mediterranean (*C. americanum*), African (*C. obtusifolia* and *C. biflorum*) and Eurasian (*C. corylifolium* and *C. drupacea*) taxa while the Australian *Cullen* are a derived clade (BS = 95%, PP = 1.00). Ancestral character state reconstructions showed that ancestors of Psoraleeae had an herbaceous growth habit, compound leaves, and indehiscent fruits. Biogeographic reconstructions showed that Psoraleeae probably originated in the Mediterranean South biome of Africa in the Miocene, ca. 8.2 mya, and then dispersed into the Succulent biome of the Americas, Mediterranean North biome, Temperate Grasslands of Africa and Australia to the Temperate Forest of Asia. Climate changes in the late Miocene, changes induced by glaciation during the Quaternary and environmental heterogeneity were proposed as the major drivers of speciation in Psoraleeae.

Keywords: Legumes, long distance dispersal, New World, Old World, *Psoralea.*

2.1. Introduction

Psoraleeae is one of the 30 tribes in the Papilionoideae (Fabaceae) and consists of ca. 223 species in nine genera. Members of Psoraleeae are distributed worldwide (Table 1, Figure 1), mainly in the temperate biomes, with ca. 60% of the species endemic to the GCFR of southern Africa (Stirton 2005). The remaining species are distributed in the Americas (25%), Australia (13%), and Eurasia / Macaronesia (2%) (Table 1).

Table 1. Genera of the tribe Psoraleeae and their distributions

Currently recognised genera	**Number of species**	**Distribution**	**References to genera**
Otholobium C.H.Stirt.	63	Southern and eastern Africa; South America (ca. 8 spp.)	Stirton 1981. In R. M. Polhill & P. H. Raven, *Advances in Legume Systematics* 1: 341.
Psoralea L.	75	Southern Africa	Linnaeus 1753, Sp. Plant.
Pediomelum Rydberg	29	Canada, USA and Mexico	Rydberg 1919. N. Amer. Fl. 24: 18.
Orbexilum Raf.	9	Eastern USA	Rafinesque 1832. Atl. J. 1(4): 145.
Rupertia Grimes	3	Western USA (California Ranges)	Grimes 1990. Mem. New York Bot. Gard. 61: 1–113.
Hoita Rydberg	3	Western USA (California)	Rydberg 1919. N. Amer. Fl. 24: 18.
Ladeania A.N.Egan & Reveal	3	Western USA and Canada	A. N. Egan & Reveal 2009. *Novon* 19 (3): 310–314
Cullen Medik.	34	Mainly Australia, Eurasia (3) and Africa (4)	Medik. 1787 Vorles. Churpfälz. Phys.-Öcon. Ges. ii. 381.
Bituminaria C.H.Stirt.	5	Macaronesia, Mediterranean Europe, Caucasia	Stirton 1981. In R. M. Polhill & P. H. Raven, *Advances in Legume Systematics* 1: 341.

Historically, Psoraleeae began as the genus *Psoralea* first described by Linnaeus (1753) based on two South African species: *P. pinnata* L. and *P. aculeata* L. More species were later on described under *Psoralea*, with Bentham (1865) recognising ca. 100 species globally which were distributed in South Africa (40), North America (30) and Australia (11). Among the early works on Psoraleeae, Vail (1894) recognised 21 species in her revision of the American members of the genus *Psoralea*. In a subsequent revision, Rydberg (1928) restricted the name *Psoralea* sensu Bentham (1865), to only ca. 40 species occurring in southern Africa. He also re-classified the American species into a number of genera based on leaf and fruit characters including: *Amorpha* L., *Apoplanesia* C. Presl., *Aspalthium* Medik., *Cullen* Medik., *Eysenhardtia* Kunth., *Hoita* Rydb., *Kuhnistera* Lam., *Orbexilum* Raf., *Parosela* Cav., *Parryella* Torr. & A. Gray, *Pediomelum* Rydb., *Petalostemon* Michx., *Psoralidium* Rydb., *Psorodendron* Rydb., *Psorothamnus* Rydb., *Rhytidomene* Rydb. and *Thornbera* Rydb. However, many subsequent works (e.g. Guthrie 1939, Isely 1958, Ockendon 1965) ignored the generic classification by Rydberg (1928) and preferred to retain the name *Psoralea* for all species. Ockendon (1965) asserted that it was difficult to divide *Psoralea* into natural groupings, highlighting the lack of correlation in the features used by Rydberg, which were mostly pod and leaf features. The splitting of *Psoralea* into different genera became an accepted classification only after Stirton's (1981) classification based on flowers, fruits, seeds, leaf arrangement and leaf morphology.

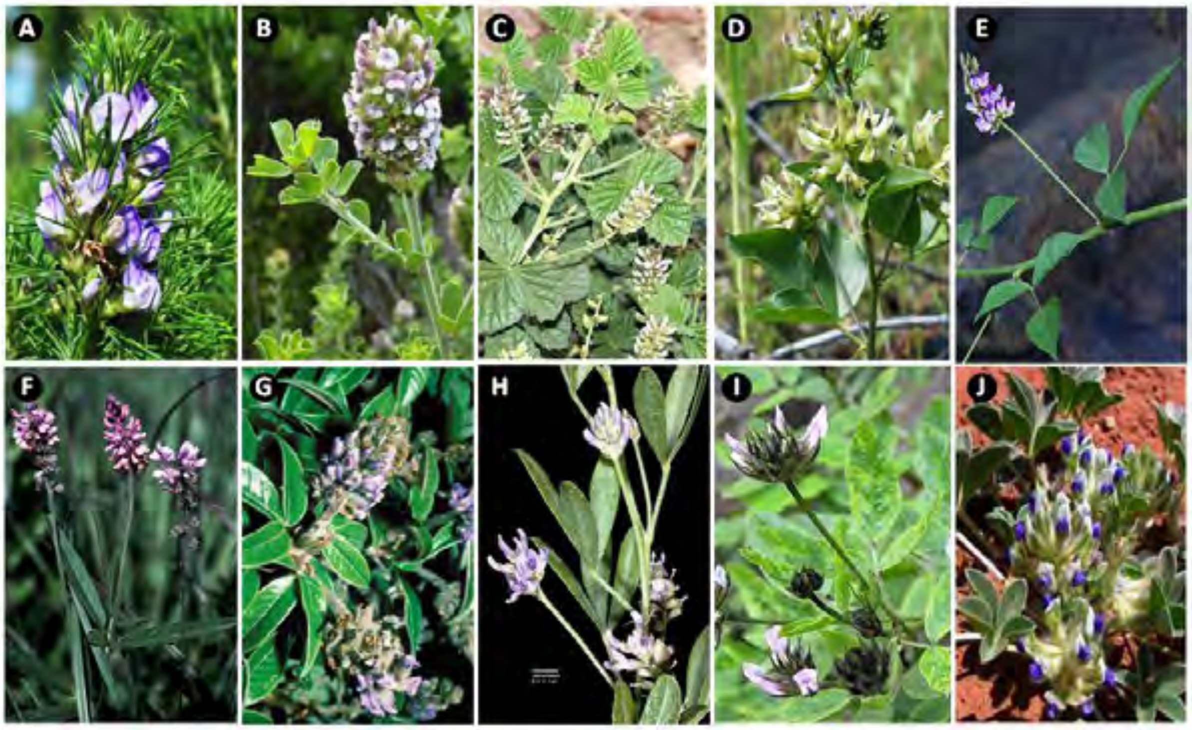


Figure 1. Representatives of Psoraleeae showing morphological variations in the genera. **A**. *Psoralea* **B**. *Otholobium* **C**. *Cullen* **D**. *Rupertia* **E**. *Hoita* **F**. *Orbexilum* **G**. South American *Otholobium* **H**. *Ladeania* **I**. *Bituminaria* **J**. *Pediomelum*. Photographs: A. Bello, C.H.S. and Muasya.

Stirton (1981) separated *Psoralea sensu lato* into six genera: *Psoralea*, *Hallia* Thunb., *Cullen, Bituminaria* Heist ex. Fabr., *Orbexilum*, and *Otholobium* C.H.Stirt. a new genus cleaved out of the African *Psoralea.* He re-restricted the name *Psoralea* to ca. 20 species endemic to the GCFR and with some outliers stretching due north up to Swaziland and Mozambique. However, Stirton (1981) did not commit to sub-dividing the North and South American Psoraleeae recognised under the name *Orbexilum* until a monographic revision was made. This was done in Grimes's (1990) taxonomic revision of the American Psoraleeae, based on 20 morphological characters. He recognised a monophyletic North American (NAm) group of five genera: *Hoita*, *Orbexilum, Pediomelum*, *Psoralidium*, and *Rupertia*. He also recognised four Old World genera: *Cullen*, *Bituminaria*, *Psoralea*, and *Otholobium*. Grimes (1990) placed eight South American species into the genus *Otholobium*, albeit reluctantly, noting that these species shared certain characteristics in the calyx and fruit with *O. caffrum*, the type species of the genus, but which did not fit cleanly into the genus circumscription of *Otholobium* as outlined by Stirton (1981) e.g, "recurved-mucronate leaflets".

Grimes (1990) hypothesised that *Otholobium* and *Psoralea* were sister genera and formed a clade sister to the rest of Psoraleeae, with *Bituminaria* and *Cullen* included in the latter clade. However, Grimes's (1990) hypothesis on relationships between the genera in Psoraleeae is not supported by the molecular phylogenetic studies of Egan & Crandall (2008a & b) or Dludlu *et al.* (2013). For example, the Dludlu *et al.* (2013) study does not support the sister relationship between African *Otholobium* and *Psoralea* and the Egan & Crandall (2008a & b) study resolved the genus *Rupertia* as sister to *Ladeania*, a clade then sister to *Pediomelum*, while *Cullen* and the southern African (SAf) *Otholobium* were sister to the rest of the American Psoraleeae.

Apart from the phylogenetic position of *Psoralea*, the phylogenetic relationships within *Otholobium* and between *Otholobium* and the other genera of Psoraleeae are yet to be resolved. Grimes's (1990) placement of the South American (SAm) species of tribe Psoraleeae into *Otholobium* has not been supported by molecular evidence; the polyphyly of *Otholobium* was confirmed by a phylogenetic study of the American Psoraleeae by Egan & Crandall (2008a) based on eight DNA markers and a recent study by Dludlu *et al.* (2013), which used three of the markers employed by Egan & Crandall (2008a). Despite the inclusion of only three African taxa, Egan & Crandall (2008a) found that the SAf *Otholobium* species formed a clade sister to the rest of the American Psoraleeae, while the SAm *Otholobium* species were nested within the American clade as sister to the genus *Orbexilum*. Dludlu *et al.* (2013) built on Egan & Crandall's (2008a) taxonomic sampling by increasing the sampling with nearly 50% of the African Psoraleeae and found that *Psoralea* and the SAf *Otholobium* formed a well-supported clade derived within the tribe and sister to the genus *Hoita* (relationship not well supported), and the two SAm *Otholobium* species included were resolved as a clade sister to *Bituminaria*. Therefore, *Otholobium* sensu Grimes (1990) is not monophyletic, and these two studies suggested that *Otholobium* should be broken into two groups based on geography; South America and South Africa. However, Egan & Crandall's (2008a) analyses were not completely congruent with respect to the position of SAm *Otholobium* species in relation to other Psoraleeae genera. Consequently, Egan & Crandall (2008a) did not make any taxonomic changes, calling for further sampling across both geographic regions before making such a decision. Dludlu *et al.* (2013) on the other hand could not conclusively resolve the issue due to low support at the backbone of their phylogeny, and the sister relationship of the strongly supported SAf clade could not be clearly established. Dludlu *et al.* (2013) therefore suggested the need to sample more DNA loci in addition to the three already sampled in order to test the hypothesis of the sister relationship between *Otholobium* and *Psoralea* as proposed by Grimes (1990) and to determine their phylogenetic position relative to the other genera in the tribe.

Furthermore, the monophyly and phylogenetic position of the genus *Cullen* was not conclusive as previous studies did not include the vast majority of the species in the genus. As Egan & Crandall (2008a & b) and Dludlu *et al.* (2013) included only five species in their studies, the monophyly of the genus was not unequivocally ascertained, and the phylogenetic position of the Old World *Cullen* species has never been studied. Although Grimes (1997) carried out a taxonomic revision of *Cullen* based on morphology, his revision is largely restricted to the Australian species. Despite the fact that Kroiss *et al.* (2009) deposited some sequences of *Cullen* species on GenBank, studies have not utilised them yet. There is therefore a need to study *Cullen* species along with the rest of Psoraleeae in order to test the monophyly and establish the true phylogenetic position of the genus.

Studies on the tribe Psoraleeae suggest a rapid evolution or diversification of its lineages relative to other legume clades (Lavin *et al.* 2005; Egan & Crandall 2008b). The age of the divergence between two transcontinental clades of Psoraleeae genera (*Cullen* of Australia, and *Rupertia* of North America) is estimated at ca. 6.3 mya (Lavin *et al.* 2005). This age estimate is an indication of recent diversification in this group, especially as the NAm clade of Psoraleeae diversified after the transcontinental split.

Psoraleeae is predominantly temperate to montane tropical in distribution, with 60% of species in the Temperate South biome and 17% in the Temperate North biome (Schrire *et al.* 2005a & b). Schrire *et al.* (2005b) also reported that the out-groups for Psoraleeae, *Glycine* L. and *Amphicarpaea* Elliott (Egan & Crandall 2008a & b), might be distributed in the

Temperate North biome. The majority of the species of Psoraleeae are predominantly distributed in the fynbos and succulent Karoo biomes of southern Africa. Schrire *et al.* (2005b) hypothesised that dispersal explains the distribution of Psoraleeae, arguing that the recent age of Psoraleeae diversification precludes Tethyan vicariance.

This chapter builds on the previous research detailed above by comprehensively sampling all genera and building a robust phylogeny based on eight DNA regions to test the monophyly of and previous hypotheses on generic relationships. I also aim to reconstruct the biogeographical history of Psoraleeae both at global and local scales. The specific objectives are:

1. To test the Egan & Crandall (2008a & b) and Dludlu *et al.* (2013) hypotheses on generic relationships within Psoraleeae based on expanded sampling.
2. To evaluate the utility of four morphological diagnostic characters used by Stirton (1981) and Grimes (1990) in delimiting the sections and genera of Psoraleeae.
3. To reconstruct the biogeographical history (distribution pattern, potential ancestral area and biome) of Psoraleeae and its genera.

2.2. Materials and methods

2.2.1. Taxon sampling

This chapter includes extensive sampling of the tribe Psoraleeae (Appendix A), building on previous studies (Egan & Crandall 2008a; Kroiss *et al.* 2009; Dludlu *et al.* 2013). New species/samples added here were ca. 50/70 (see appendix A). For the southern African genera, 55 species (including the unrecognised species) of *Otholobium* representing 99% of the total species were included. For the genus *Psoralea*, 63 species representing the recognised and the unrecognised species were sampled for the phylogenetic analysis. For *Cullen*, 61% (20) of the species were sampled, including African (3 species), European (1) and Australian species (sequences deposited by Kroiss *et al.* 2009). Sequences for the remaining Psoraleeae genera not in southern Africa (i.e. *Pediomelum*, *Cullen*, *Hoita*, *Rupertia*, *Orbexilum*, *Ladeania* and *Bituminaria*) were downloaded from GenBank. For field-collected samples sourced specifically for this study, tissue from young, fresh leaves (or shoots in the case of leafless species) was dried in silica gel for DNA extraction, and voucher specimens were deposited in the Bolus Herbarium (BOL) at the University of Cape Town. The out-groups used by Egan & Crandall (2008a & b) and Dludlu *et al.* (2013) were maintained in this study. According to Egan & Crandall (2008a & b), the out-group taxa were chosen on the basis of a recent Legume family phylogeny based on *matK* chloroplast DNA sequences (Wojciechowski *et al*. 2004) that show *Glycine* as the putative sister of Psoraleeae. These included species in the tribes (all author citation were given in Appendix A): Abreae (*Abrus precatorius*); Desmodieae (*Desmodium floridanum*); Phaseoleae (*Apios americana*); and Phaseoleae, subtribe Glycininae (*Amphicarpaea bracteata*, *Cologania pallida*, *Glycine canescens*, *Glycine microphylla*).

2.2.2. DNA extraction, amplification and sequencing

Total genomic DNA was extracted from the dried material (20 mg) using a modified Cetyltrimethylammonium Bromide (CTAB) technique from Doyle & Doyle (1987) and Gawel & Jarret (1991). To do this, all the samples were ground into fine powder using a Retsch MM400 mixer mill (Retsch GmbH, Haan, Germany) for 15 min at 30 Hz.

Several markers were screened for successful amplification, sequencing quality and the amount of sequence variation between species. These were: *ETS* (Chandler *et al.* 2001); *ITS* (White *et al.* 1990); *waxy* (Egan & Crandall, 2008a); the intergenic spacer *psbA-trnH* (Sang *et al.* 1997); and *trnL-F, trnS-trnG, rpoB-trnC*, *trnQ-rps16*, *ndhF-rp132, rpl32-trnL*, *rps16*, *ycf1*and *trnD-trnT* (Shaw *et al.* 2005, Shaw *et al.* 2007). The markers were chosen based on previous studies on Psoraleeae (Egan & Crandall 2008a & b, Dludlu *et al.* 2013) or their application at family level (Fabaceae) phylogeny. The screening was done with at least 10 samples of *Otholobium* and *Psoralea* and two to three samples of *Cullen* and *Bituminaria* species. The DNA regions that were eventually used were ITS, *rpl32-trnL, trnQ-rps16, trnS-trnG*, *rpoB-trnC*, *trnL-trnF* plus the DNA barcodes (*matK* and *rbcL*). The DNA regions were chosen because of their ability to provide successful amplification, good sequencing and a considerable amount of sequence variation between species. The details of the primer sequences and the corresponding references are shown in Table 2.

Table 2. Primers and their corresponding sequences that were used for amplification and sequencing of the different DNA regions. F/R= forward and reverse respectively.

Region	**Name**	**F/R**	**Sequence (5′-3′)**	**Reference**
ITS	ITS5	F	GGAAGTAAAAGTCGTAACAAGG	White *et al.* (1990)
	ITS4	R	TCCTCCGCTTATTGATATGC	White *et al.* (1990)
matK	MatK-1RKIM-f	F	ACCCAGTCCATCTGGAAATCTTGGTTC	CCDB Primer set www.ccdb.ca
	MatK-3FKIM-r	R	CGTACAGTACTTTTGTGTTTACGAG	CCDB Primer set www.ccdb.ca
rbcL	rbcLa-F	F	ATGTCACCACAAACAGAGACTAAAGC	Levin (2003)
	rbcLa-R	R	GTAAAATCAAGTCCACCRCG	Kress & Erickson (2007)
rpl32-trnL	trnL$^{(UAG)}$	F	CTGCTTCCTAAGAGCAGCGT	Shaw *et al.* (2007)
	rpL32-F	R	CAGTTCCAAAAAAACGTACTTC	Shaw *et al.* (2007)
trnL-trnF	trnL^{UAA}	F	CGAAATCGGTAGACGCTACG	Taberlet *et al.* (1991)
	TrnF^{GAA}	R	ATTTGAACTGGTGACACGAG	Taberlet *et al.* (1991)
trnQ-5'rps16	trnQ$^{(UUG)}$	F	GCGTGGCCAAGYGGTAAGGC	Shaw *et al.* (2007)
	rpS16x1	R	GTTGCTTTYTACCAC ATCGTTT	Shaw *et al.* (2007)
trnS-trnG	trnS-F	F	GCCGCTTTAGTCCACTCAGC	Hamilton (1999)
	trnG-R	R	GAACGAATCACACTTTTACCAC	Hamilton (1999)
rpoB-trnC	trnC^{GCA}R	F	CACCCRGATTYGAACTGGGG	Shaw *et al.* (2005)
	rpoB	R	CKACAAAAYCCYTCRAATTG	Shaw *et al.* (2005)

Polymerase chain reactions (PCR) were prepared using a total volume of 30 µl per sample for all the regions. These were made up of 18.6 µl of sterile distilled water; 3 µl buffer; 3 µl $MgCl_2$; 1.2 µl dNTPs; 1 µl forward and reverse primers; 2 µl of template DNA; and 0.2 µl *Taq* polymerase. For the ITS, *trnL-trnF*, and *rpoB-trnC* markers, the process involved an initial denaturation phase of 5 minutes at 94 ºC followed by 35 cycles of 1 minute at 94 ºC: 1 minute at 54 ºC (annealing); 1.5 minutes at 72 ºC (extension); and a final extension phase of 7 minutes at 72 ºC. For the *rpl32-trnL*, *trnQ-rps16*, and *trnS-trnG* markers, the reaction was run for an initial denaturation phase of 3 minutes at 94 ºC followed by 35 cycles of: 45

seconds at 94 ºC; 45 seconds at 52 ºC (annealing); 1 minutes at 72 ºC (extension); and a final extension phase of 7 minutes at 72 ºC. All the PCRs were run on an Applied Biosystems GeneAmp 2700 thermal cycler (Applied Biosystems, Foster City, CA, USA). The PCR products were loaded (3 µl of each sample) into wells on a 1% agarose gel stained with ethidium bromide and ran in an electrophoresis tank containing 0.5 × TBE at 100 V for 15 minutes. The gel was then visualised under UV light, a photo of the gel was taken, and successful amplifications were used for the sequencing step. Amplified PCR products were sent to MacroGen Netherlands (http://www.macrogen.com) or the University of Stellenbosch DNA sequencing facility for sequencing using the primers from the PCR step. For the DNA barcode markers, dry tissue was sent to the CBOL laboratory in Canada where amplification and sequencing were performed.

2.2.4. Ancestral state reconstructions

Four characters used by Stirton (1981) and Grimes (1990) to distinguish between the sections and genera of Psoraleeae were coded (Table 3). Taxa, in which a particular character was not applicable, were excluded from the analysis. This was only applicable for the members of the *Psoralea aphylla* complex, which have no leaves, and were excluded for the character "leaf type". Ancestral state reconstruction was done in Reconstruct Ancestral State Phylogeny (RASP) using a model-based approach, Bayesian Binary MCMC (BBM) (Yu *et al.* 2011). The topological constraint corresponding to each node was enforced using a Bayesian tree, and then ancestral states for each node were reported. One thousand random post-burn-in trees from the Bayesian analysis of the dataset were loaded in RASP and a BBM analysis was conducted with 50 000 generations, 10 runs and a sample frequency of 100. Relative frequencies of ancestral areas were plotted on a condensed tree. A condensed (consensus) tree is a majority rule consensus tree with compatible groups with less than 50% support allowed. The phylogeny was condensed using a TreeAnnotator (Drummond *et al.* 2012), setting the posterior probabililty limit to 0.5 and burn in to 2500.

Table 3. Characters used for ancestral state reconstruction for *Otholobium* and *Psoralea*.

Character	**State A**	**State B**
Habit	Herbs (without woody stems)	Shrubs/trees (woody stem)
Leaf type	Compound	Simple
Cupulum	Absent	Present
Fruits	Indehiscent	Dehiscent

2.2.5. Estimation of divergence dates

Only the combined dataset (nrDNA + cpDNA) was used for the dating analysis. The calibration points used were: the most recent common ancestor (MRCA) of *Cullen* and *Rupertia* with a mean date of 6.3 ± 0.9 mya (Lavin *et al.* 2005); the MRCA of *Glycine* and Psoraleeae with 14.90 ± 2.18 mya; and the MRCA of *Pediomelum* with 3.28 ± 1.48 mya (Egan & Crandall 2008b). These divergence ages were modelled as a normal distribution of which respective node age is equal to the mean and the upper and lower bounds are standard deviations.

2.2.6. Dating the tree

The program BEAST (Bayesian Evolutionary Analysis Sampling Trees) version 1.8.2 (Drummond *et al.* 2012) was used for the dating analysis. All analyses were carried out on CIPRES web portal 3.1 (http://www.phylog.org) (Miller *et al.* 2010) employing a relaxed clock model with log-normally distributed, uncorrelated rates of substitution between branches. No topological constraints were employed so as to allow the topological uncertainties to be taken into account. The tree prior was modelled under the Yule speciation process. Other than the normally distributed priors on the calibration points, all other priors were set as default values in the program BEAUti version 1.8.2 (which comes as part of the BEAST package; Drummond *et al.* (2012)) in which the input files for BEAST were created. The MCMC settings consisted of six parallel runs of 100 million generations sampled every 10 000 generations. The program LogCombiner (Drummond *et al.* 2012) was used to combine the log files upon completion. The program Tracer version 1.6 (Rambaut *et al.* 2014) was used to confirm likelihood stationarity, adequate mixing of the MCMC chains, and whether the six separate runs had converged as well as the burnin. The trees for each run were then combined after discarding the first 2 500 trees. TreeAnnotator (Drummond *et al.*

2012) was used to produce a best-supported summary tree, annotated with the mean ages of nodes and posterior clade probabilities for each node. FigTree v1.4.2 (http://tree.bio.ed.ac.uk/software/figtree/) was used to display the estimated tree with node ages and their 95% confidence intervals.

2.2.7. Biogeographic analysis

The distribution range of Psoraleeae was divided into five broad biomes corresponding to six areas with one or more endemic species, as shown in Figure 2. The biomes were coded as Succulent/Xeric shrublands (S), Mediterranean North (MN), Mediterranean South (MS), Temperate Grassland (TG) and Temperate Forest (TF). The areas were coded as A = Africa, B = Australia, C = North America, D = South America, E = Eurasia/Mediterranean, and F = Asia (Figure 2).

A model-based approach, BBM (Yu *et al.* 2011), was used to infer the biogeographic histories of Psoraleeae. A topological constraint corresponding to each node was enforced using the BEAST tree obtained from the analysis of the combined dataset, and then, ancestral states for each node were reported. One thousand random post-burn-in trees from the BEAST analysis of the combined dataset (see above) were loaded in RASP and a BBM analysis was conducted with 10 runs, a sample frequency of 100 for 50 000 generations. The maximum number of ancestral areas was set to two, three and four in three independent runs to allow for widespread ancestors. Relative frequencies of ancestral areas were plotted on a condensed tree.

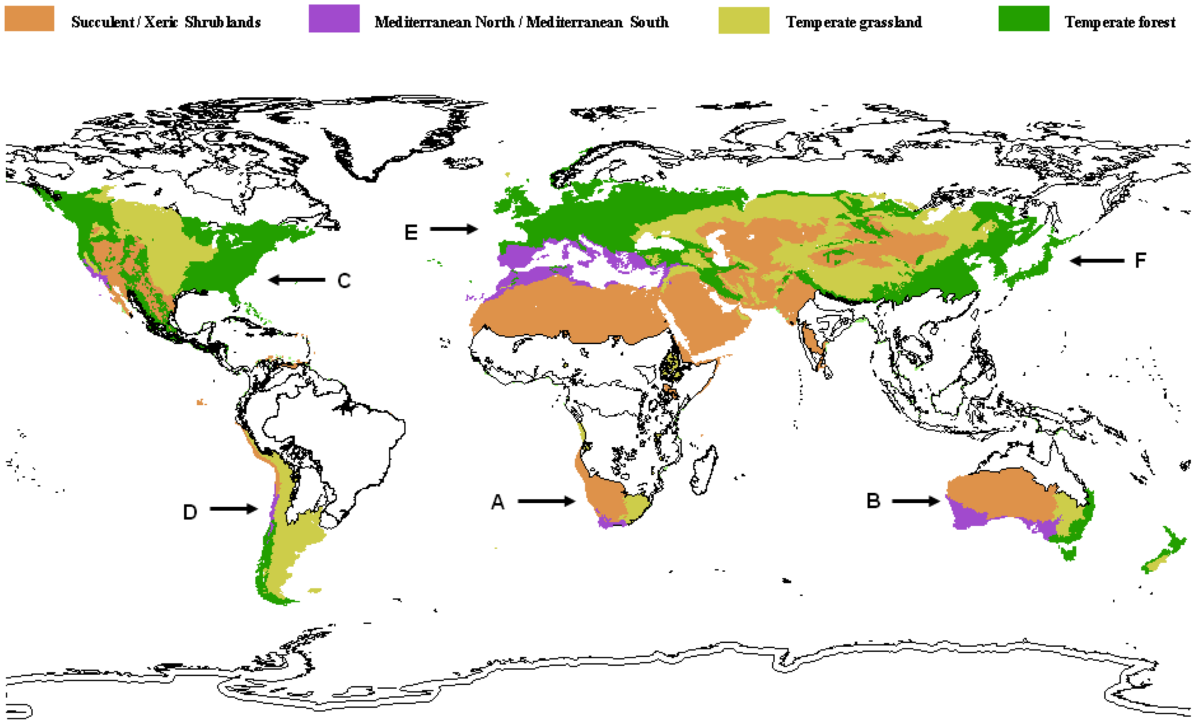


Figure 2. Biogeographical regions: **A**. Africa, **B**. Australia, **C**. North America, **D**. South America, **E**. Eurasia/Mediterranean, and **F**. Asia.

2.3. Results

2.3.1. Data matrices

The aligned matrices for the eight gene regions had 202 taxa, out of which seven were from the out-group. Each of the matrices contained 812, 775, 553, 404, 1584, 1171, 557 and 996 characters for ITS, *matK*, *rbcL*, *rpl32-trnL*, *rpoB-trnC*, *trnL-F*, *trnQ-rps16* and *trnS-trnG* respectively, and the combined matrix, i.e. the nrDNA and cpDNA, consisted of 6845 characters. The percentage coverage for each DNA region was: ITS = 16%; *trnS-trnG* = 14.6%; *rpoB-trnC* = 14.1%; *trnL-trnF* = 13.8%; *matK* = 12.4%; *rpl32-trnL* = 10.9%; *trnQ-rps16* = 10.3; and *rbcL* = 7.9%.

2.3.2. Generic relationships within Psoraleeae

The trees from the ML and BI analyses of the individual gene regions showed little resolution, and most of the branches formed polytomies (Appendix B1–18). However, most of the trees were of similar topology across the DNA regions. The nrDNA resulted in a tree nearly identical to the tree obtained from the cpDNA dataset for both ML and BI but with minor exceptions. For the BI of the nrDNA, *Hoita* was not supported (Appendix B15), while for the cpDNA, none of the individual genera were actually supported, even though the tree is tree was mostly congruent with the nrDNA tree (Appendix B17). For the ML, the clade comprising the NAm Psoraleeae, SAm *Otholobium*, *Bituminaria* and *Cullen* is weakly supported in the nrDNA tree (Appendix B16), while the cpDNA tree showed a long basal polytomy (Appendix B18). Partition homogeneity tests of the individual partitions (nrDNA and the seven cpDNA regions) found all the datasets to be congruent (P= 0.998), and these were henceforth analysed as a combined dataset. The combined analysis of the eight gene regions as total evidence resulted in a more resolved tree in both BI (Figure 3a & b) and ML (Figure 4a & b) analyses than those from the individual gene regions. The BI topology showed better support for the backbone relationships among the genera of Psoraleeae (Figure 3a & b), with the ML tree (Figure 4a) showing no support for the backbone of the clade comprising the NAm Psoraleeae, SAm *Otholobium*, *Bituminaria* and *Cullen*, and instead supporting the individual genera.

The monophyly of Psoraleeae was strongly supported (Bootstrap (BS) = 100%, Posterior probability (PP) = 1.00). The SAf Psoraleeae (SAf *Otholobium* and *Psoralea*) were strongly supported (PP = 1.00) as sister to the rest of Psoraleeae (Figure 3a & b) in BI, but weakly supported (BS = 67%) in ML (Figure 4a & b). *Psoralea* is nested as a clade within *Otholobium* (BS = 70%, PP = 1). Furthermore, relationships among species within *Psoralea* and among African *Otholobium* were poorly supported, yielding a polytomy (Figure 3b and 4b). The two SAm species of *Otholobium* were resolved as a well-supported clade (BS = 100%, PP = 1.00) sister to the genus *Bituminaria* (BS = 71%, PP = 1.00) (Figure 3a and 4a). The genus *Orbexilum* is resolved as sister to the genus *Hoita* and the genus *Cullen* with 0.97 PP. The Australian, South African and Eurasian *Cullen* species formed a strongly supported clade (PP = 1.00) (Figure 3a). *Ladeania* and *Rupertia* were resolved as sisters (BS = 88%, PP = 1.00) and form a clade sister with *Pediomelum* (BS = 87%, PP = 1.00) (Figure 3a and 4a).

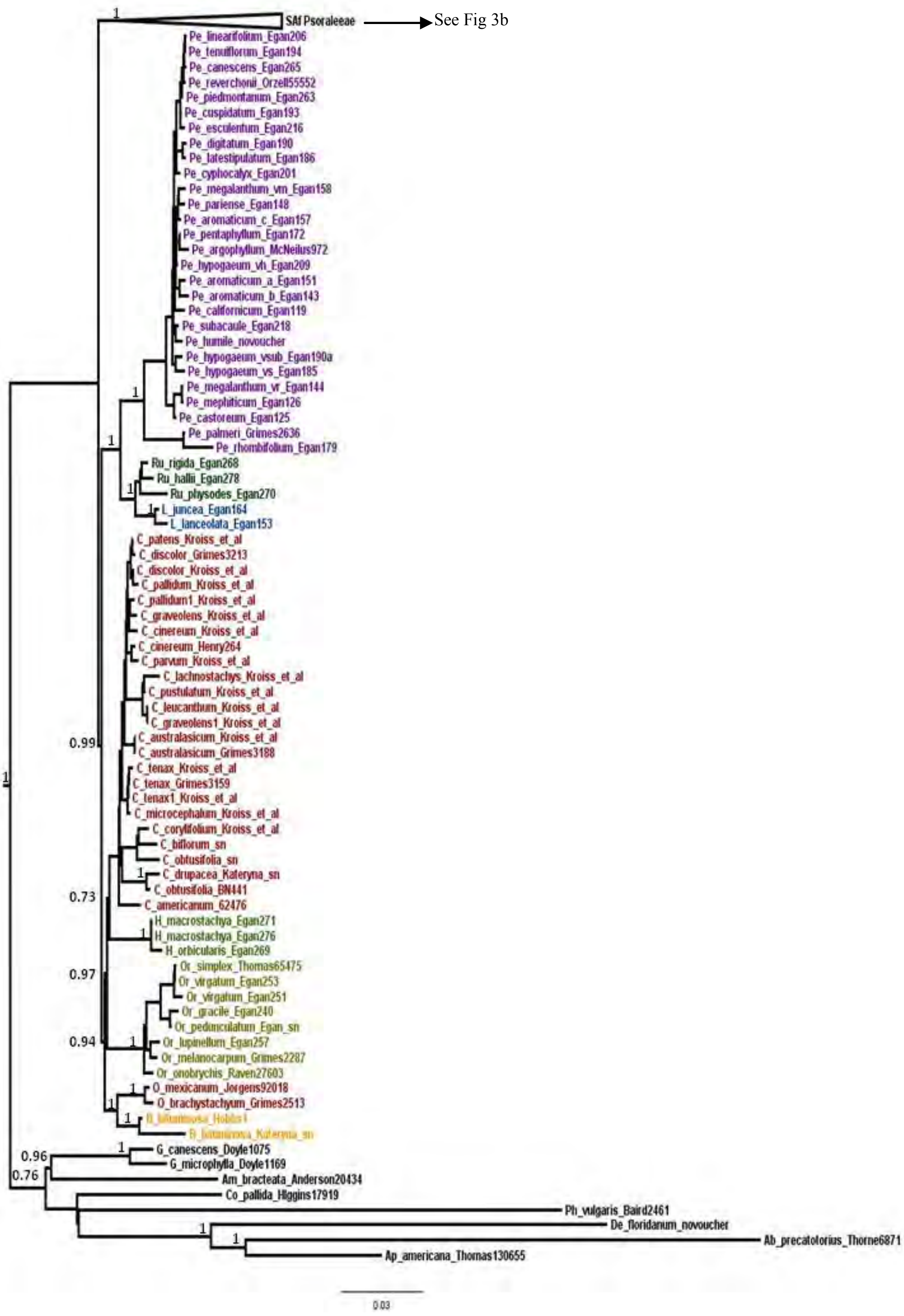


Figure 3a. Phylogenetic relationships of Psoraleeae (202 taxa) based on Bayesian Inference analysis of the combined DNA dataset. Numbers above branches are posterior probability (PP) values greater than 0.7. The names of the genera are abbreviated as follows: Ap = *Apios*, Ab = *Abrus*, Am = *Amphicarpaea*, B = *Bituminaria*, Co = *Cologania*, C = *Cullen*, De = *Desmodium*, G = *Glycine*, H = *Hoita*, O = *Otholobium*, Or = *Orbexilum*, P = *Psoralea*, Pe = *Pediomelum*, L = *Ladeania* and Ru = *Rupertia*.

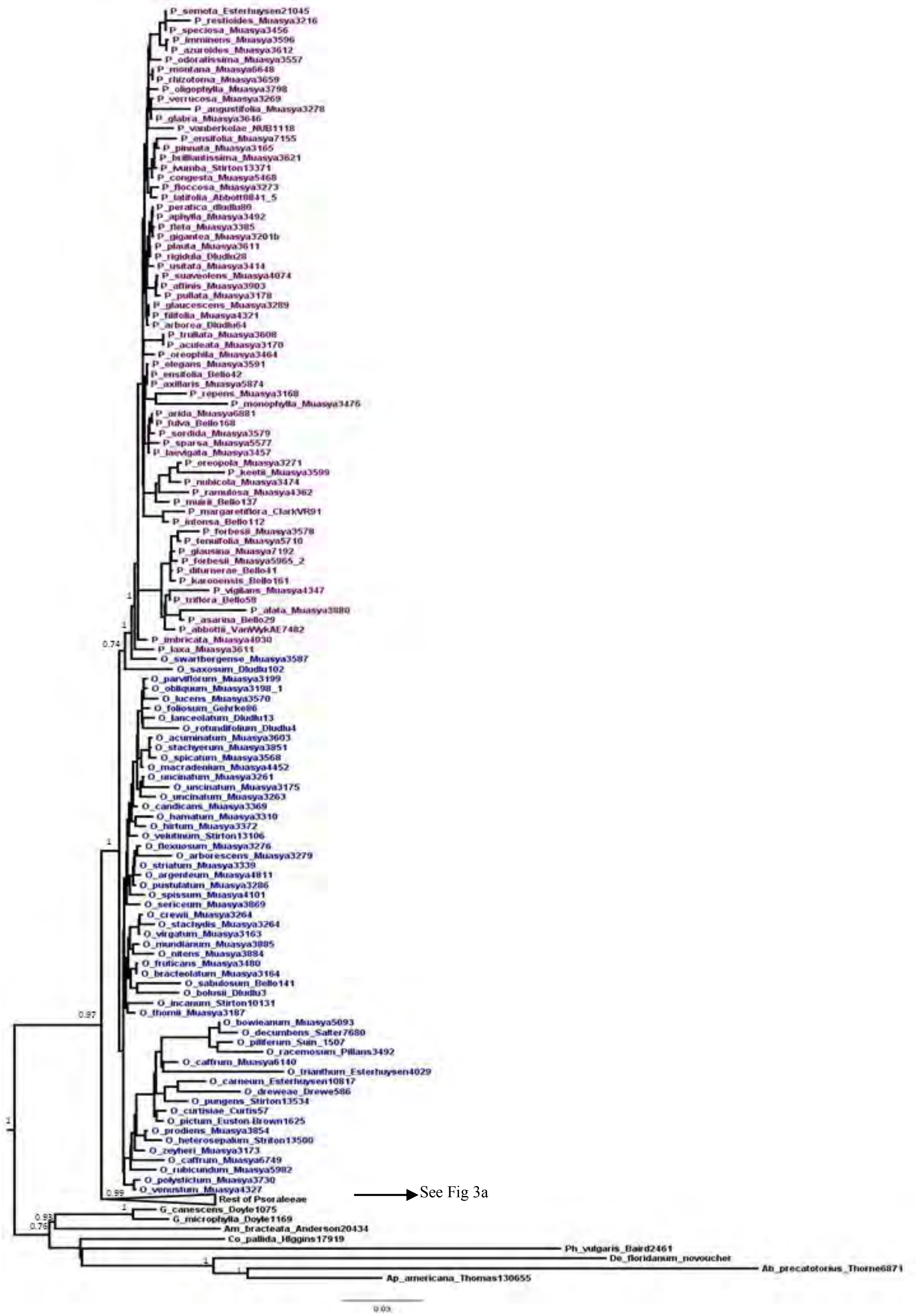


Figure 3b. Phylogenetic relationships of Psoraleeae (202 taxa) based on Bayesian Inference analysis of the combined DNA dataset. Numbers above branches are posterior probability (PP) values greater than 0.7. The names of the genera are abbreviated as follows: Ap = *Apios*, Ab = *Abrus*, Am = *Amphicarpaea*, B = *Bituminaria*, Co = *Cologania*, C = *Cullen*, De = *Desmodium*, G = *Glycine*, H = *Hoita*, O = *Otholobium*, Or = *Orbexilum*, P = *Psoralea*, Pe = *Pediomelum*, L = *Ladeania* and Ru = *Rupertia*.

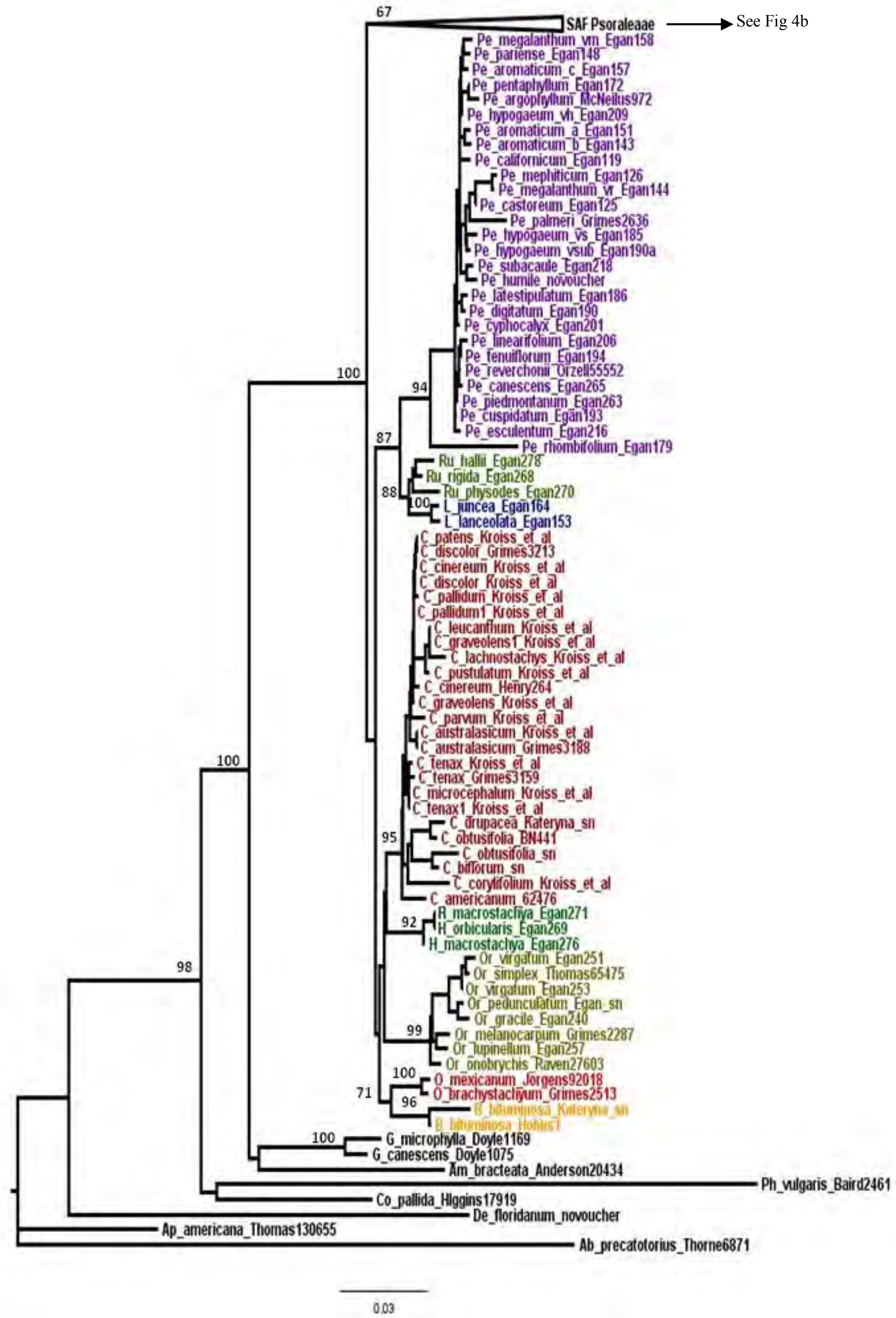


Figure 4a. Phylogenetic relationships of Psoraleeae (202 taxa) based on RAx-ML analysis of the combined DNA dataset. Numbers above branches are bootstrap support values (%) greater than 50%. The names of the genera are abbreviated as follows: Ap = *Apios*, Ab = *Abrus*, Am = *Amphicarpaea*, B = *Bituminaria*, Co = *Cologania*, C = *Cullen*, De = *Desmodium*, G = *Glycine*, H = *Hoita*, O = *Otholobium*, Or = *Orbexilum*, P = *Psoralea*, Pe = *Pediomelum*, L = *Ladeania* and Ru = *Rupertia*.

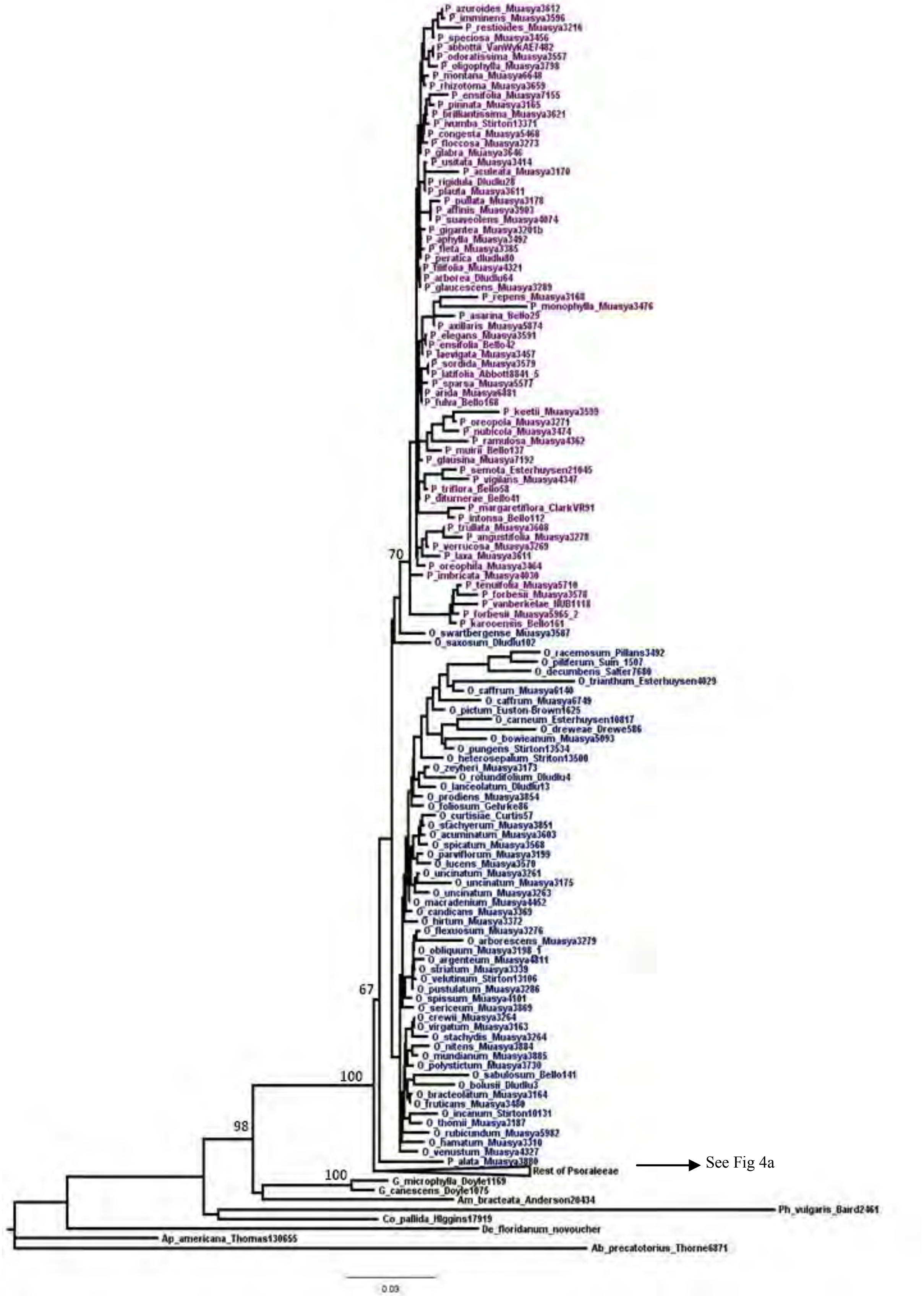


Figure 4b. Phylogenetic relationships of Psoraleeae (202 taxa) based on RAx-ML analysis of the combined DNA dataset. Numbers above branches are bootstrap support values (%) greater than 50%. The names of the genera are abbreviated as follows: Ap = *Apios*, Ab = *Abrus*, Am = *Amphicarpaea*, B = *Bituminaria*, Co = *Cologania*, C = *Cullen*, De = *Desmodium*, G = *Glycine*, H = *Hoita*, O = *Otholobium*, Or = *Orbexilum*, P = *Psoralea*, Pe = *Pediomelum*, L = *Ladeania* and Ru = *Rupertia*.

2.3.3. Ancestral state reconstructions

The ancestral state reconstruction based on the BI tree showed that the ancestral state of growth habit in Psoraleeae is herbaceous (Figure 5a). The woody growth habit has evolved multiple independent times within Psoraleeae, largely within the SAf clade and in *Cullen*. The possession of compound leaves is ancestral to Psoraleeae, while having a simple leaf type is a derived state observed in African Psoraleeae (Figure 5b). Within the *Psoralea* clade, there are multiple, independent loss of leaves (white tip nodes on Figure 5b), observed in the *Psoralea aphylla* complex. This character state is another derived state that evolved only in the genus *Psoralea.* The presence of a cupulum is a derived state that evolved independently, again in *Psoralea*, and the absence of it is the ancestral state in the tribe (Figure 5c). The occurrence of indehiscent fruits is ancestral for Psoraleeae, with dehiscence evolving only in *Pediomelum* (Figure 5d).

2.3.4. Divergence dates estimates

The maximum clade credibility tree obtained from the analysis of the combined dataset (nrDNA + cpDNA) was largely congruent with the phylogeny obtained from the BI analysis (Figure 6). The monophyly of Psoraleeae and its various genera was recovered (Figure 6). The mean ages and 95% highest posterior density (HPD) intervals (upper and lower bound) for the key nodes are shown in Table 4. The mean age of the crown node of the Psoraleeae was estimated to be ca. 8.22 million years old (myo), with a 95% confidence interval (CI) of 5.90–11.01 mya, with the African clade sister to a clade comprising the rest of Psoraleeae (see Table 4 and Figure 6 for the ages and CI of the various genera). The African clade began diversifying ca. 6.47 mya (CI = 4.30–8.78), showing a grade comprising *Otholobium* and origin of *Psoralea* at ca. 4.06 mya (CI = 2.58–5.63) (Figure 6b). The second major clade split into two subclades. The subclade consisting of *Bituminaria*, SAm *Otholobium*, *Orbexilum*, *Hoita*, and *Cullen* species diversified ca. 6.71 mya (CI = 5.15–8.26) (Figure 6a). The other subclade began diversifying ca. 5.19 mya and consists of the MRCA of *Pediomelum* (3.82 mya, CI = 2.54–5.24), *Rupertia* (2.13 mya, CI = 0.75–3.72) and *Ladeania* (1.12 mya, CI = 0.15–2.43) (Figure 6a).

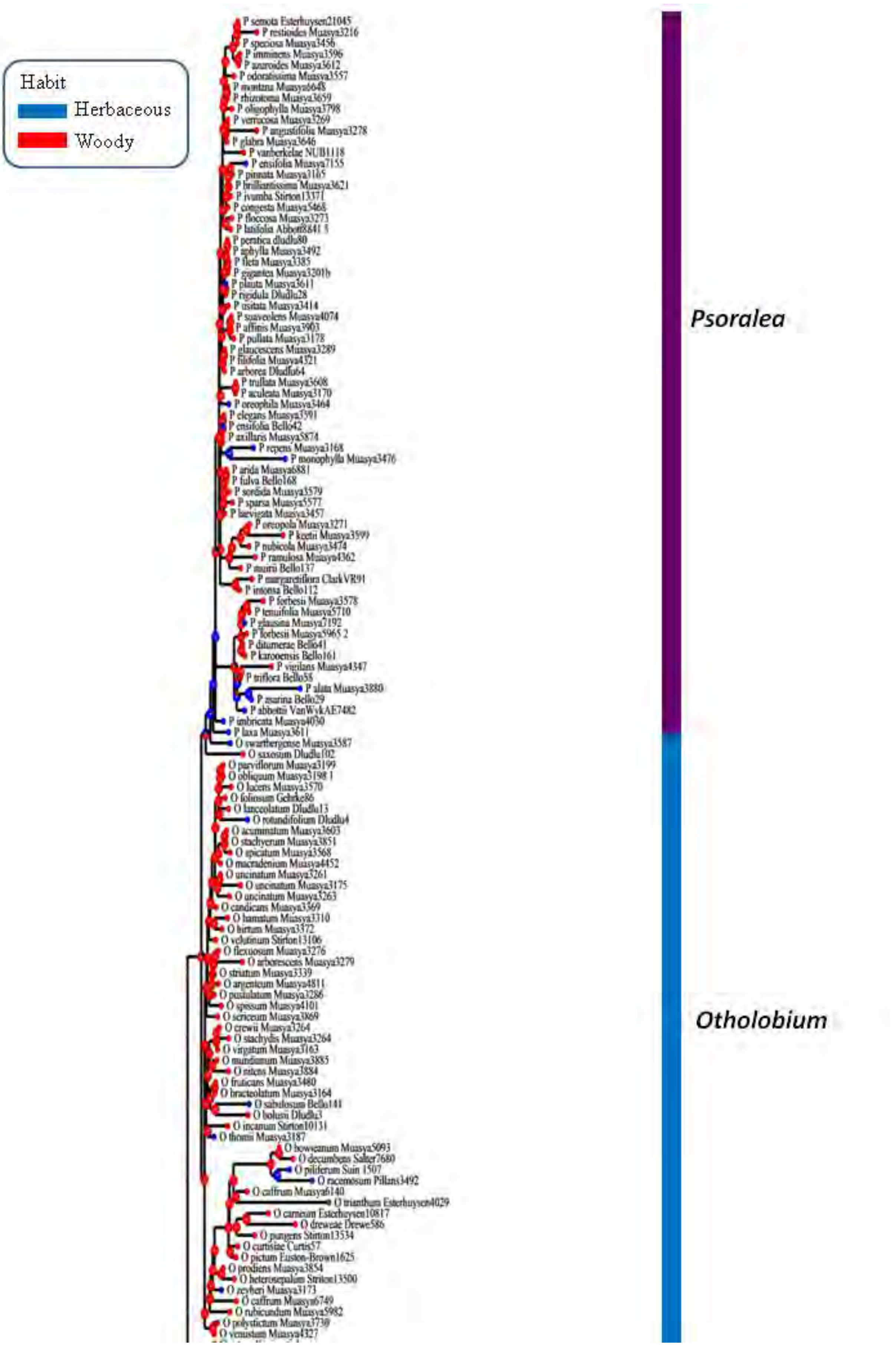


Figure 5A. Ancestral state reconstructions based on Bayesian Inference tree for growth habit (herbaceous i.e., without secondary growth or woody i.e., with secondary growth).

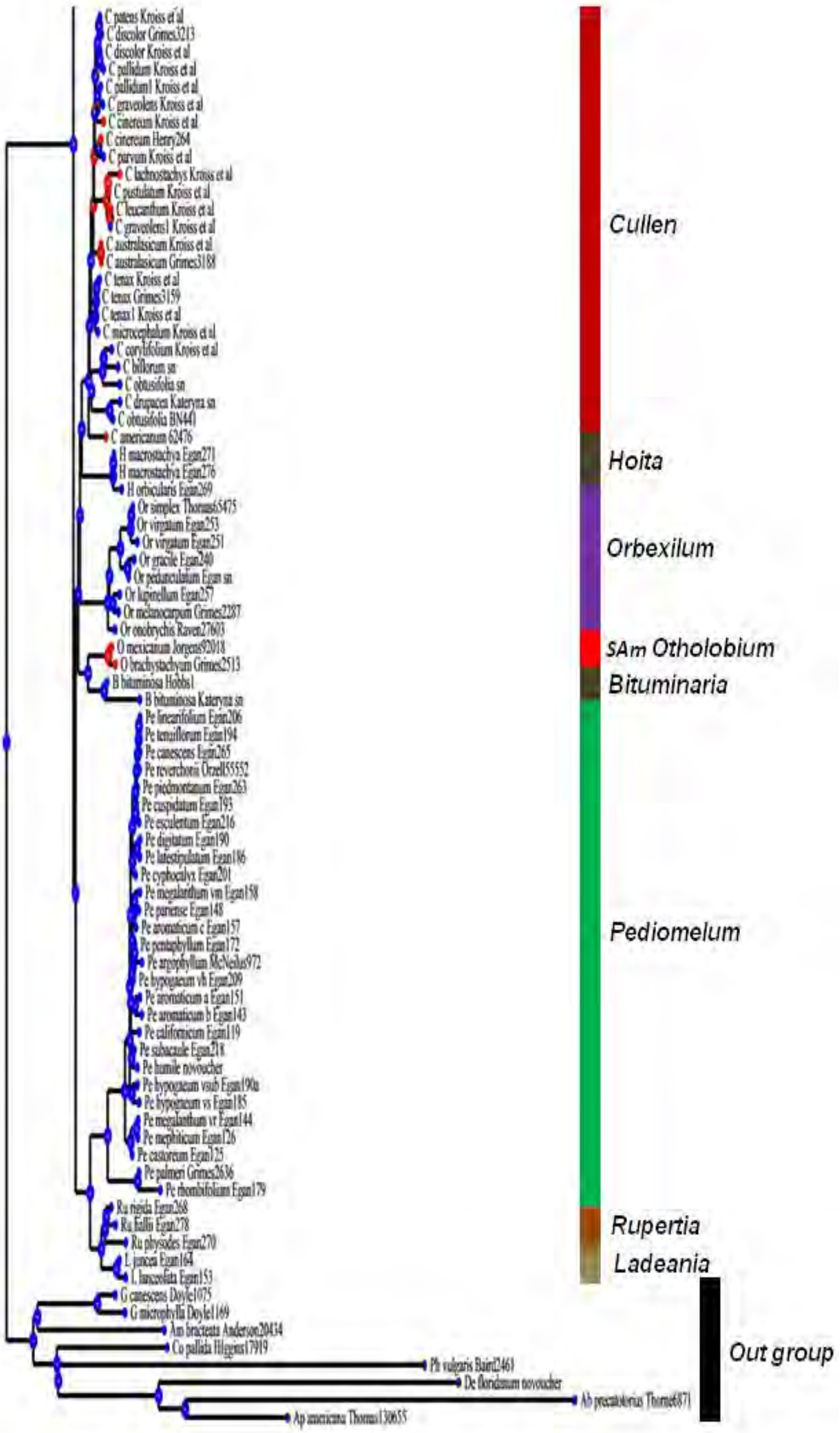


Figure 5A. Continued

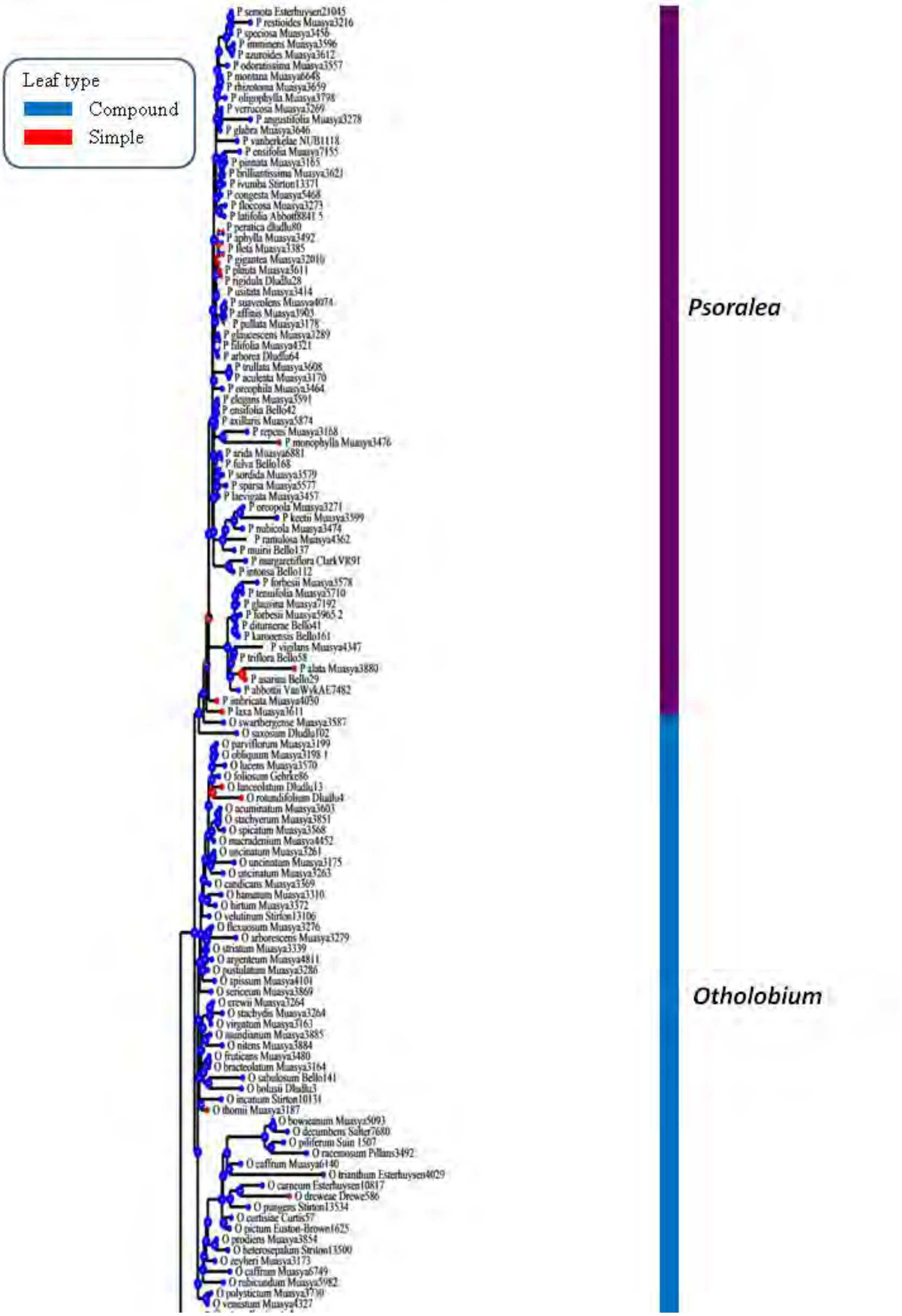


Figure 5B. Ancestral state reconstructions based on Bayesian Inference tree for leaf type i.e., whether compound (multi-foliolate) or simple (uni-foliolate).

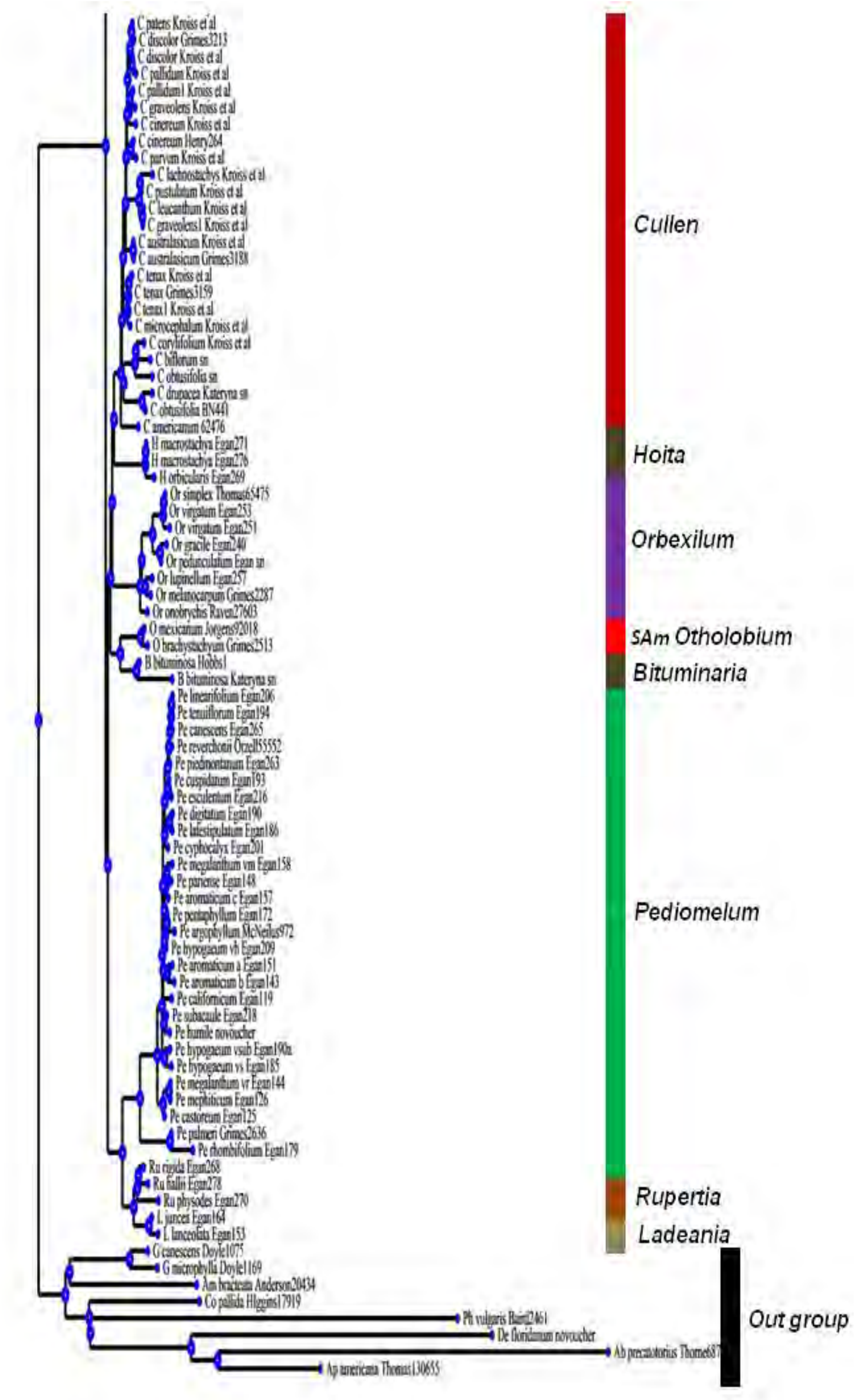


Figure 5B. Continued

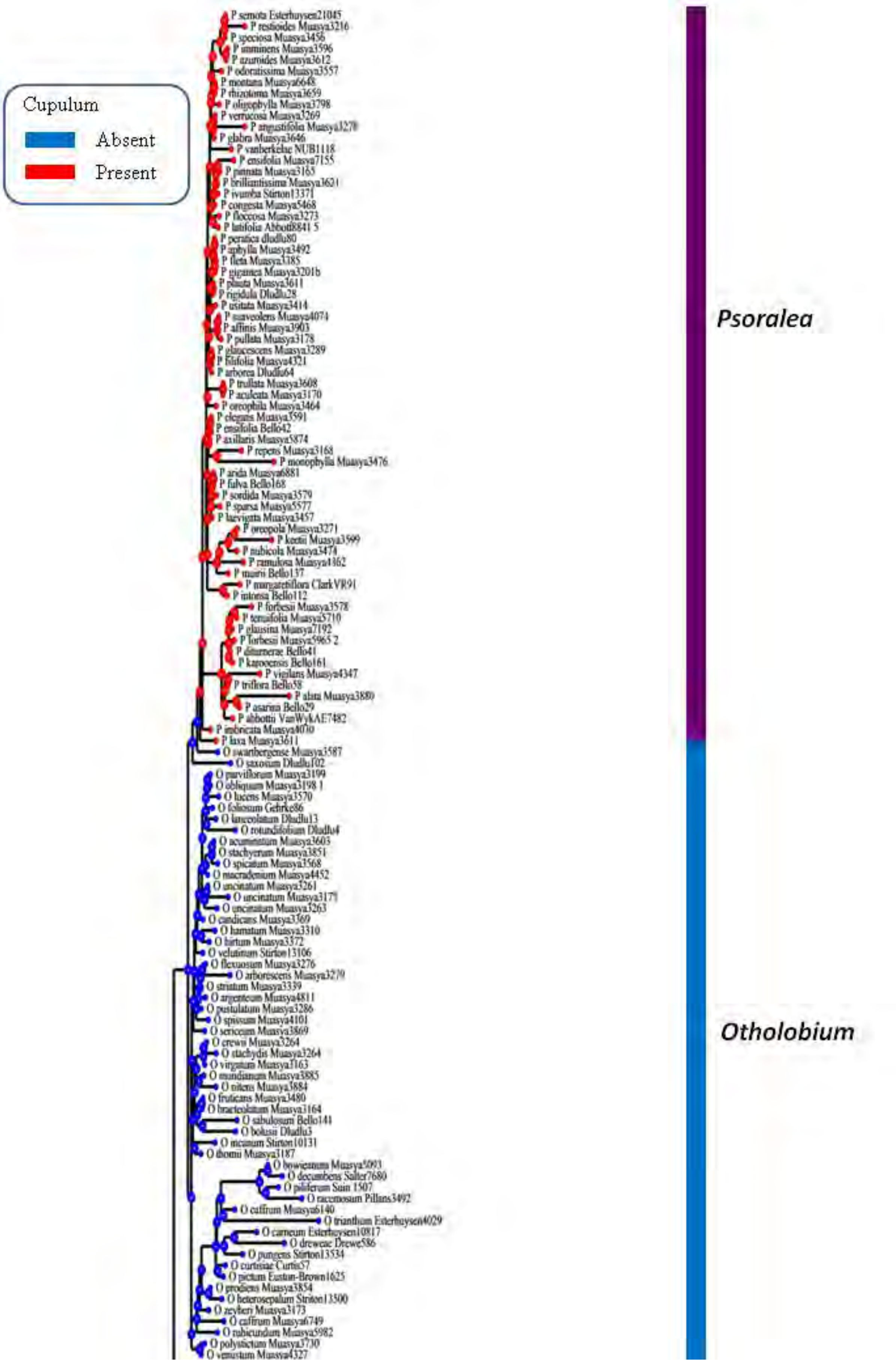


Figure 5C. Ancestral state reconstructions based on Bayesian Inference tree for presence or absence of the cupulum (a protective structure around the calyx during development).

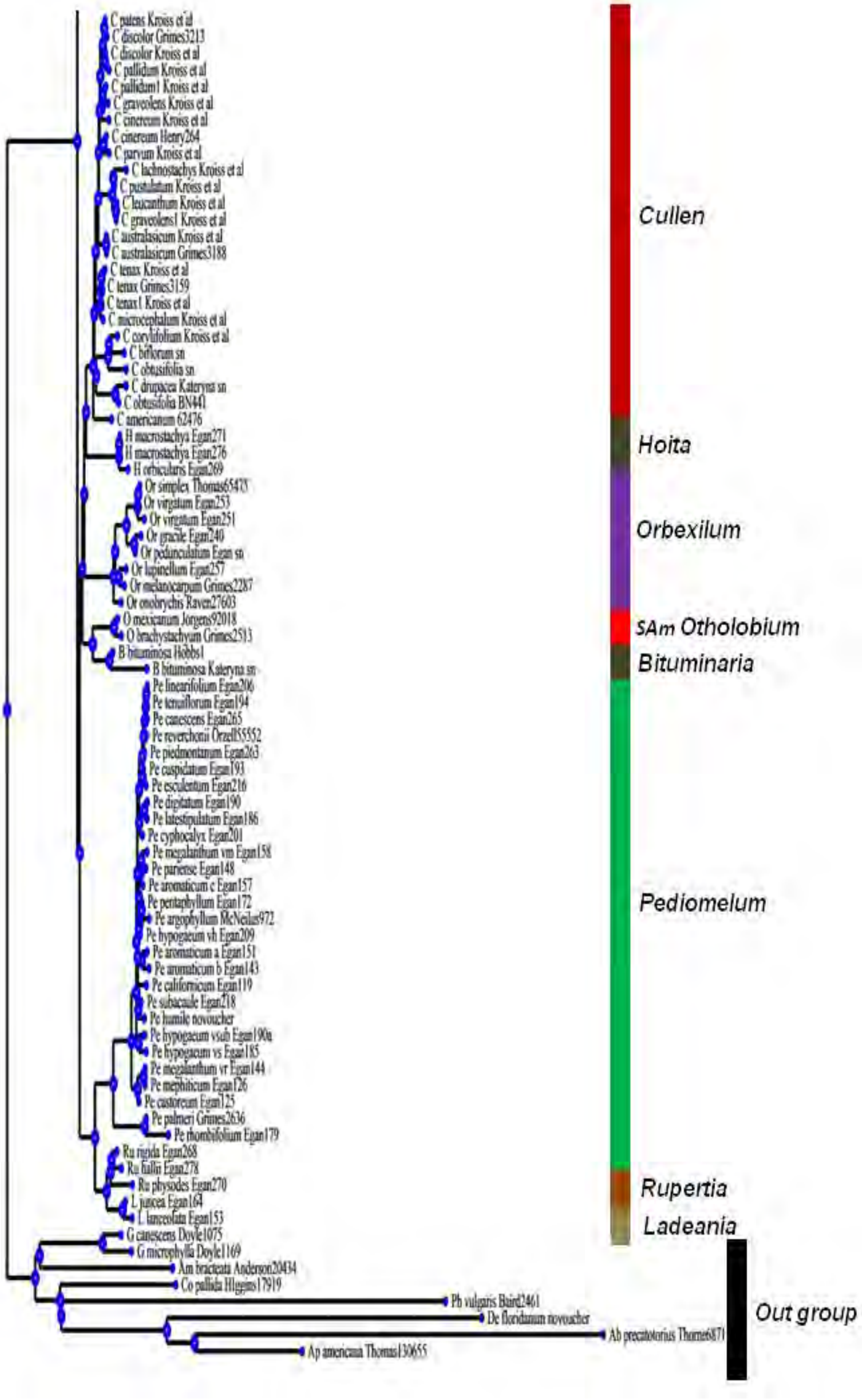


Figure 5C. Continued

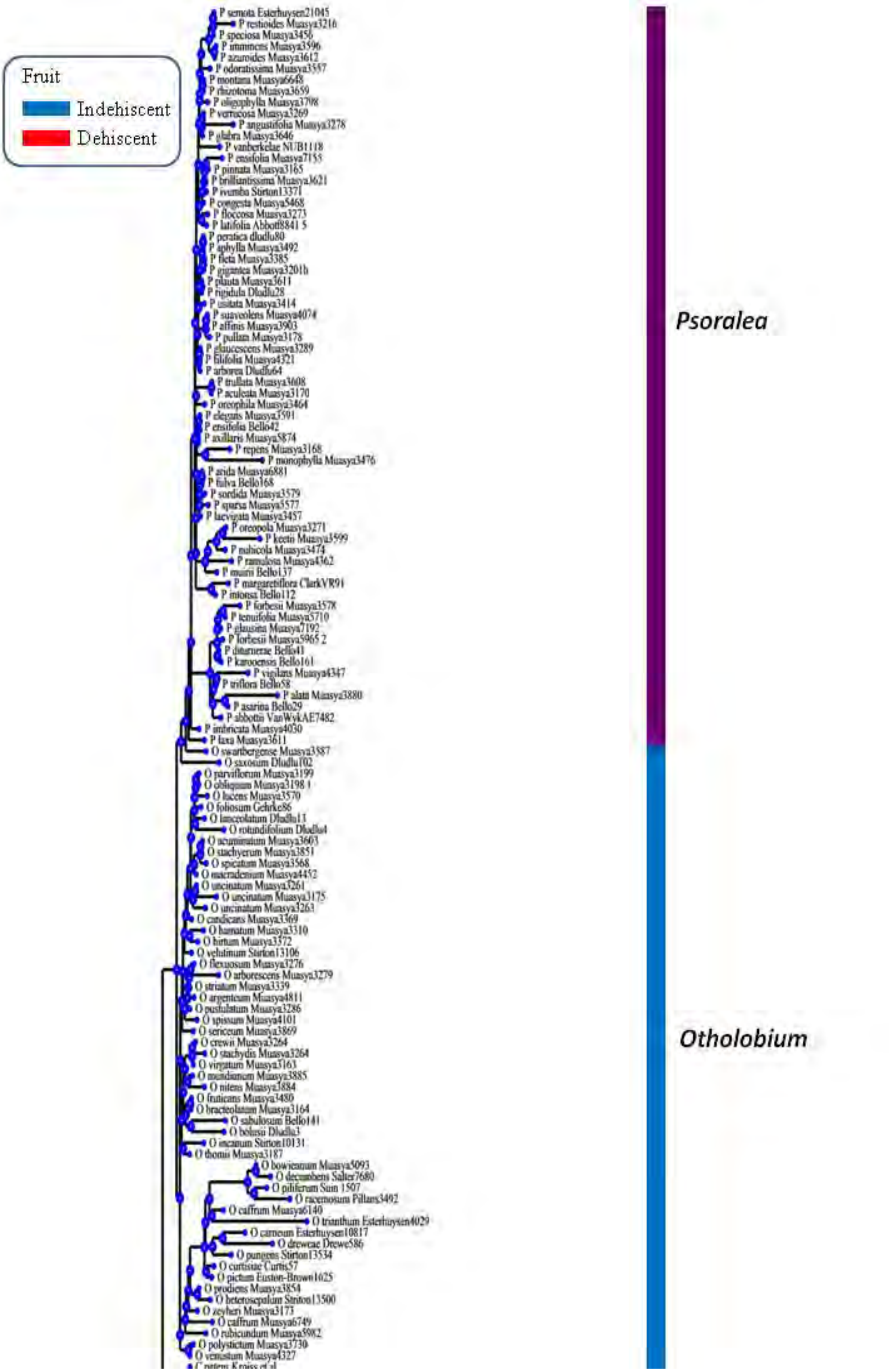


Figure 5D. Ancestral state reconstructions based on Bayesian Inference tree for fruit type (dehiscent or indehiscent).

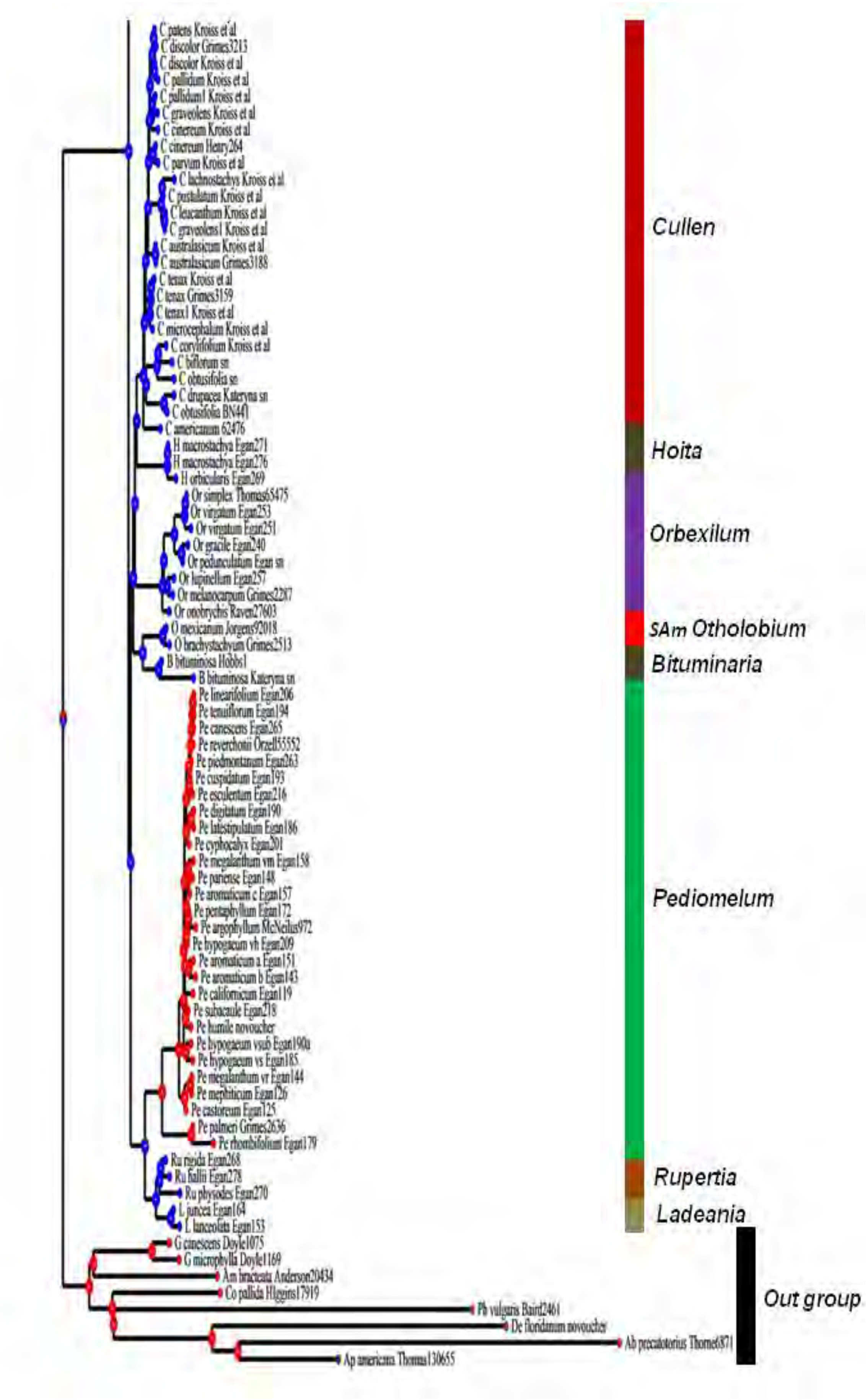


Figure 5D. Continued

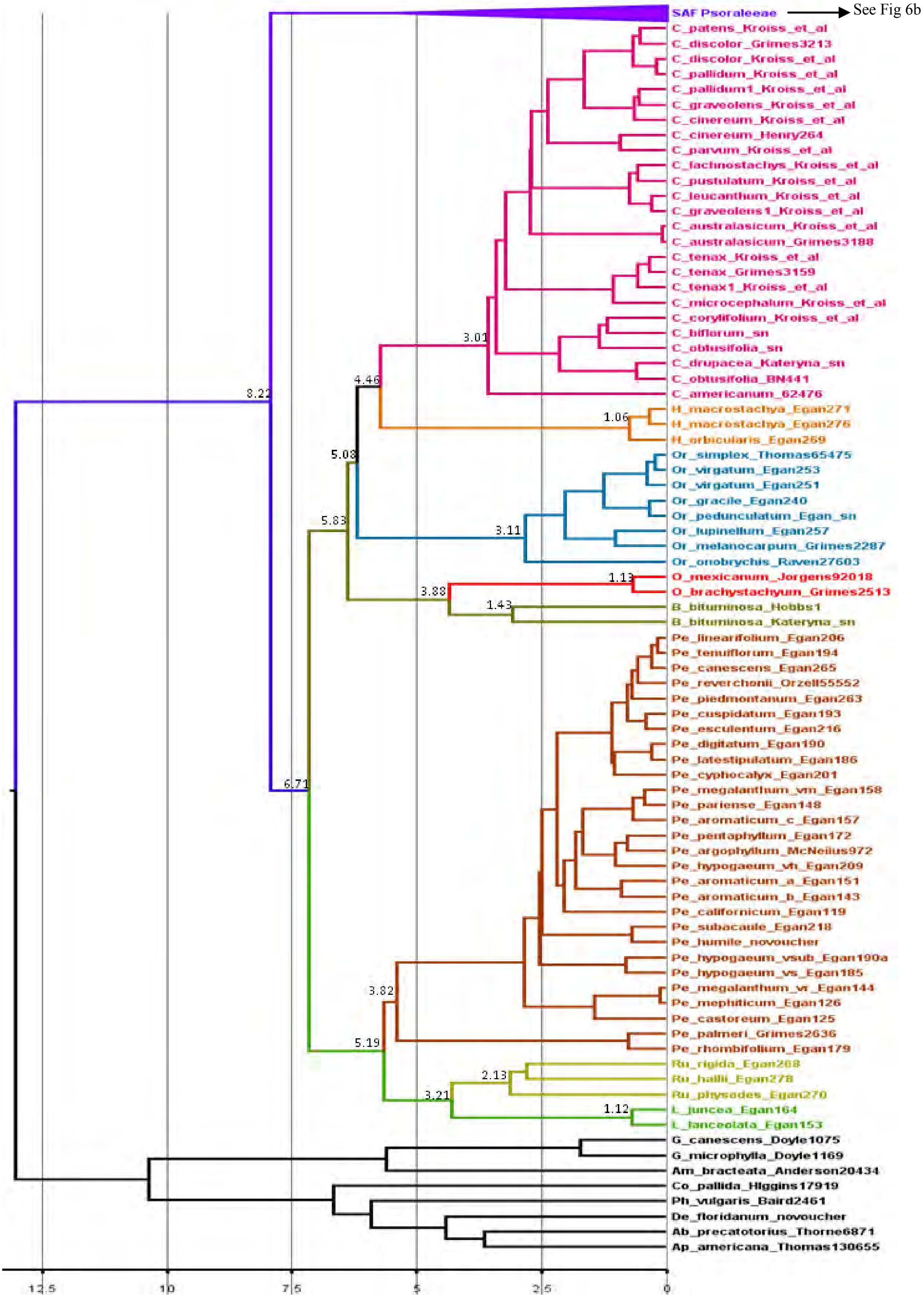


Figure 6a. Maximum clade credibility tree from the BEAST estimation of divergence dates of Psoraleeae. Numbers above branches represent mean age in millions of years. The names of the genera are abbreviated as follows: Ap = *Apios*, Ab = *Abrus*, Am = *Amphicarpaea*, B = *Bituminaria*, Co = *Cologania*, C = *Cullen*, De = *Desmodium*, G = *Glycine*, H = *Hoita*, O = *Otholobium*, Or = *Orbexilum*, P = *Psoralea*, Pe = *Pediomelum*, L = *Ladeania* and Ru = *Rupertia*.

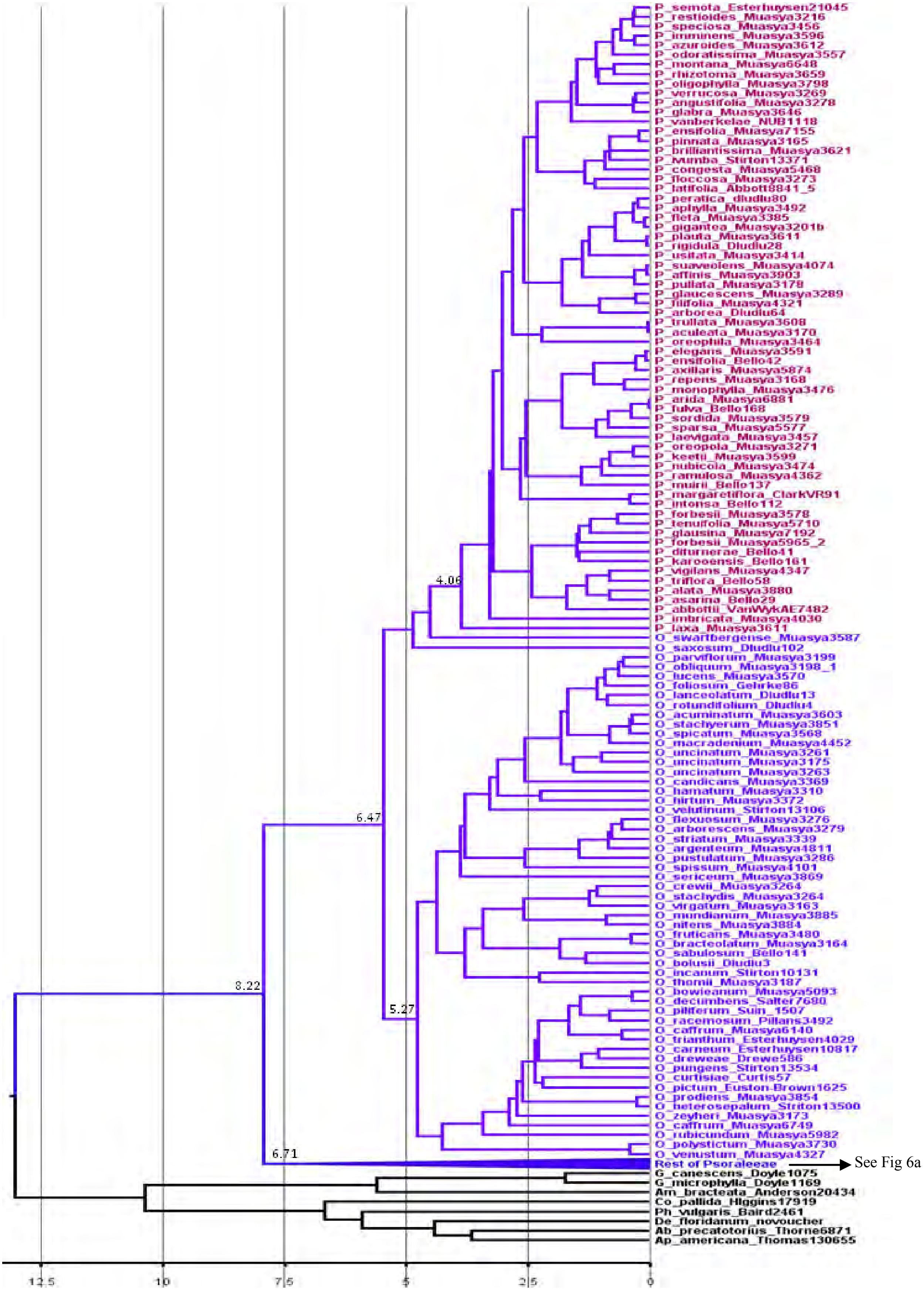


Figure 6b. Maximum clade credibility tree from the BEAST estimation of divergence dates of Psoraleeae. Numbers above branches represent mean age in millions of years. The names of the genera are abbreviated as follows: Ap = *Apios*, Ab = *Abrus*, Am = *Amphicarpaea*, B = *Bituminaria*, Co = *Cologania*, C = *Cullen*, De = *Desmodium*, G = *Glycine*, H = *Hoita*, O = *Otholobium*, Or = *Orbexilum*, P = *Psoralea*, Pe = *Pediomelum*, L = *Ladeania* and Ru = *Rupertia*.

2.3.5. Ancestral area and biome reconstruction

The biogeographic reconstructions obtained by the BBM (Figure 7) indicate the possible ancestral ranges at each node in the phylogeny of Psoraleeae. The summary of the ancestral area and biome ranges with marginal probabilities (MP) for the key nodes are indicated in Table 4. Dispersal was favoured to explain the current disjunct distribution pattern in Psoraleeae because the ages of the clades presented here are too recent to be the result of a vicariance event. Psoraleeae originated in the MS biome (MP = 53.2%, Figure 7b) of southern Africa (MP = 53.4%, Figure 7a) during the Miocene, ca. 8.22 mya (PP = 1.00, Figure 7, Table 4). Following an in situ diversification in Africa, it dispersed into the S biome of North America (MP = 96.7%) ca. 6.7 mya. This was followed by a series of both local and transoceanic dispersal and speciation events. Among these were dispersal to the S biome (MP = 99.5%) in South America (MP = 98.2), as seen in SAm *Otholobium*, and MN biome (MP = 97.5%) in Europe and the Mediterranean (MP = 97.7%), as seen in *Bituminaria* (Figure 7, Table 4). There was also dispersal to the MS biome of Africa, as seen in *Cullen*, followed by a local dispersal to TG and transcontinental dispersal to the TF biome of Australia (Figure 7, Table 4). *Cullen* then profusely dispersed and speciated into the TG and S biomes within Australia before it dispersed across the continent to the TF of Asia (Figure 7).

In summary, the present global distribution pattern in Psoraleeae can be explained by 27 dispersals events (Figure 7a). The highest speciation event within a geographical area occurred in Africa with ca. 124 speciation events (Figure 7a). This was followed by North America with 44, Australia with 18, and one each in South America and Mediterranean regions based on the current data (Figure 7a). The results showed ca. 126 global dispersal events across the biomes (Figure 7b). The highest speciation event of 111 occurred within the Mediterranean South biome followed by Succulent/Xeric shrublands 28, Temperate Grassland 15, Temperate Forest 11 and Mediterranean North 9 (Figure 7b).

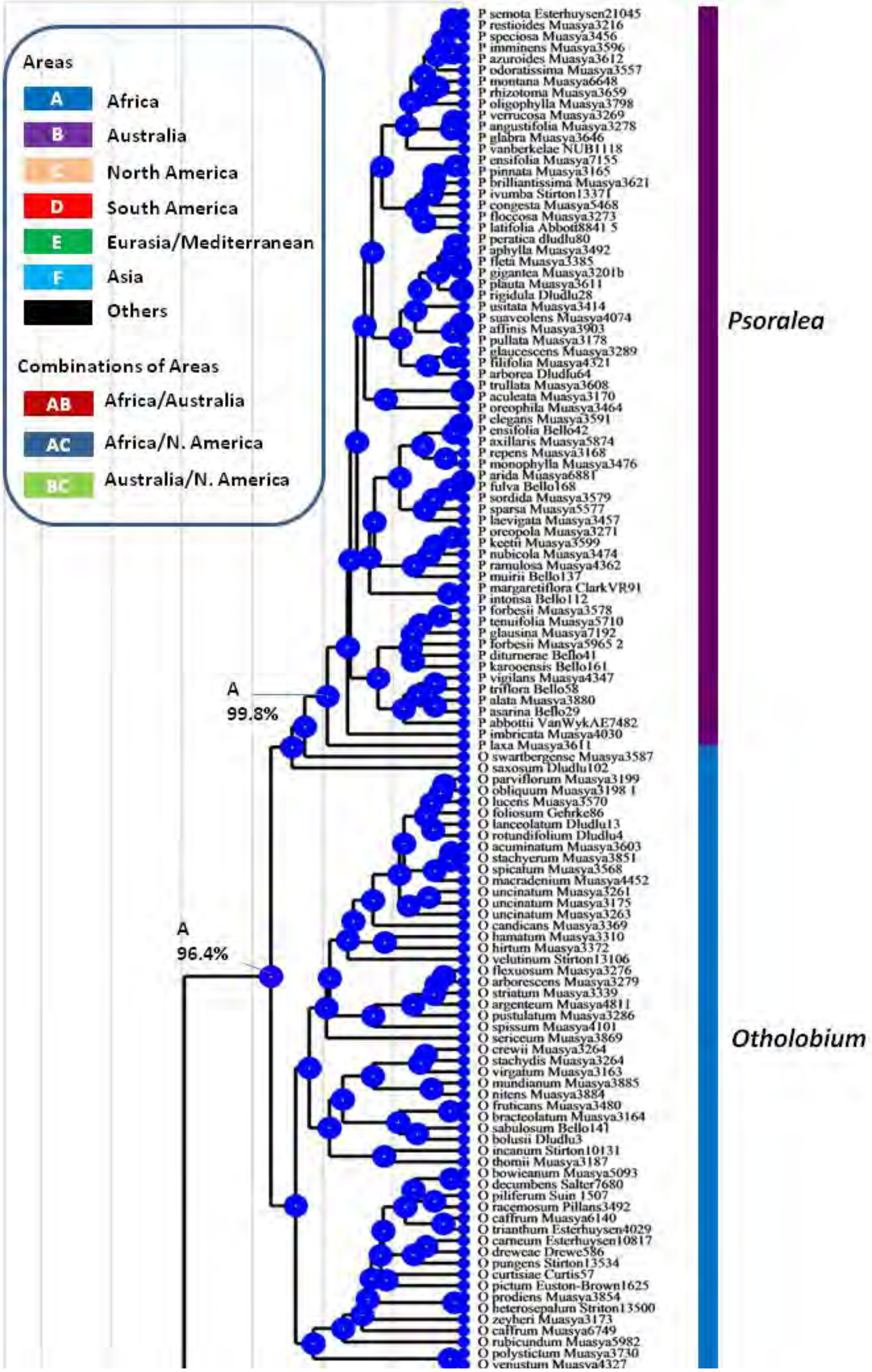


Figure 7a. Output of the Bayesian binary MCMC (BBM) optimization of ancestral states of Psoraleeae on a maximum clade credibility tree generated by BEAST representing ancestral area. The names of the genera are abbreviated as in Figure 3.

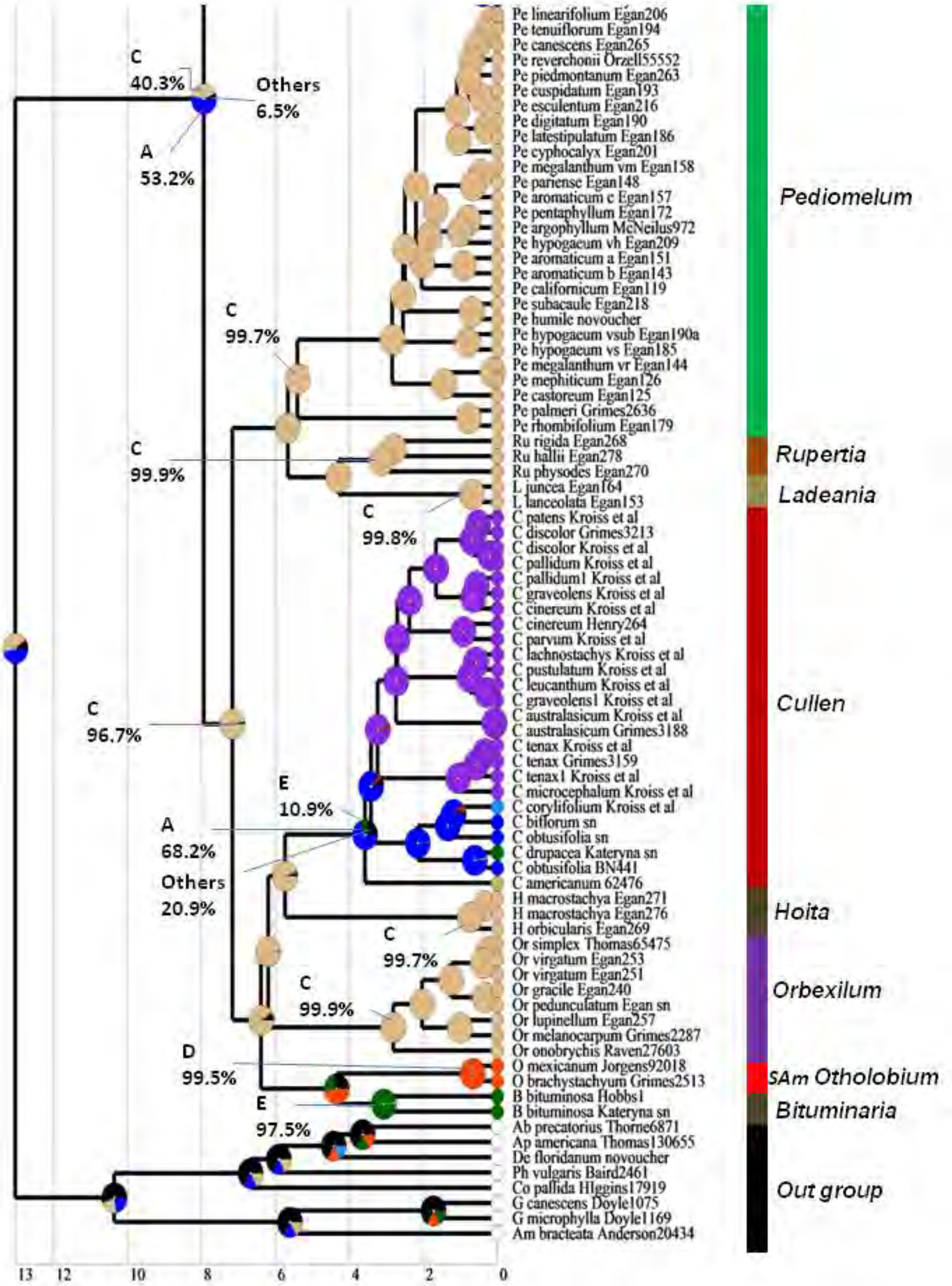


Figure 7a. Continued

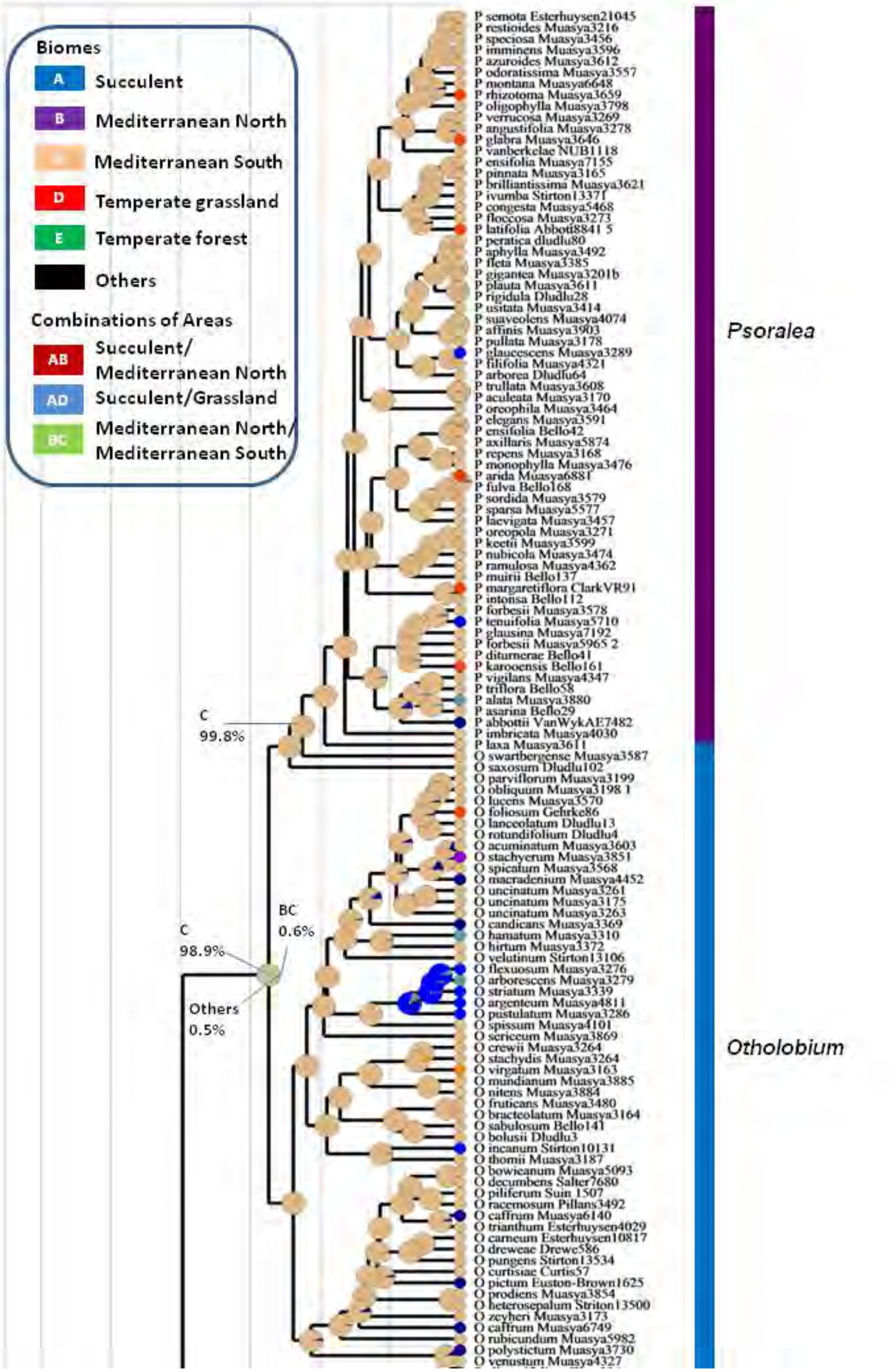


Figure 7b. Output of the Bayesian binary MCMC (BBM) optimization of ancestral states of Psoraleeae on a maximum clade credibility tree generated by BEAST representing ancestral biome. The names of the genera are abbreviated as in Figure 3.

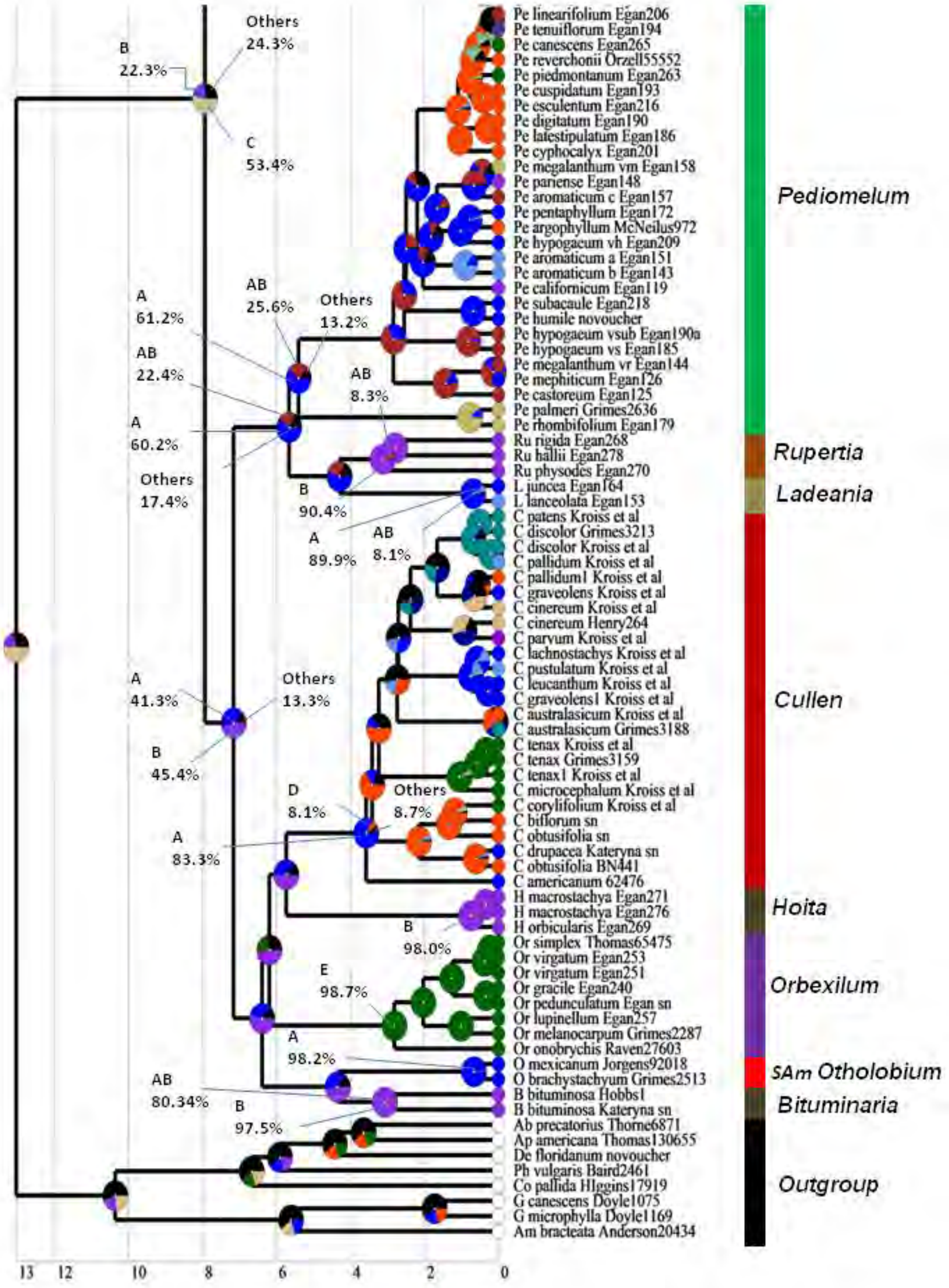


Figure 7b. Continued

Table 4: Bayesian age estimation (mean, 95% highest posterior density (HPD) lower and upper bound) with posterior probability values (PP) and estimation of ancestral areas and biomes with marginal probabilities for the key nodes of Psoraleeae generated in BEAST and RASP.

Age estimation (Mya)				**Biogeography**			
MRCA of	**Mean Age**	**95% HPD Lower–Upper**	**Posterior Probabilities**	**Ancestral Area**	**BBM Marginal probabilities (%)**	**Ancestral Biome**	**BBM Marginal probabilities (%)**
Psoraleeae	8.22	5.90–11.01	0.97	Africa	53.2	Mediterranean South	53.4
SAf Psoraleeae	6.47	4.30–8.78	1.00	Africa	96.4	Mediterranean South	99.0
Otholobium	6.47	4.30–8.78	1.00	Africa	96.4	Mediterranean South	99.0
Psoralea	4.06	2.58–5.63	1.00	Africa	99.8	Mediterranean South	99.9
Ladeania/Rupertia & Pediomelum	5.19	3.58–6.92	1.00	North America	99.7	Succulent / Xeric Shrublands	60.2
Pediomelum	3.82	2.54–5.24	1.00	North America	99.7	Succulent / Xeric Shrublands	61.2
Ladeania & Rupertia	3.21	1.43–5.10	0.96	North America	99.7	Succulent / Xeric Shrublands	55.5
Ladeania	1.12	0.15–2.43	1.00	North America	99.8	Succulent / Xeric Shrublands	89.8
Rupertia	2.13	0.75–3.72	1.00	North America	99.9	Mediterranean North	99.9
SAm Otholobium & Bituminaria	3.88	1.77–6.03	1.00	South America	48.0	Mediterranean North	48.2
SAm Otholobium	1.13	0.13–2.51	0.96	South America	99.5	Succulent / Xeric Shrublands	98.2
Bituminaria	1.43	0.21–2.91	1.00	Eurasia/Mediterranean	97.5	Mediterranean North	97.7
Orbexilum/Hoita & Cullen	5.83	4.07–7.48	1.00	North America	98.8	Mediterranean North	48.1
Orbexilum	3.11	1.69–4.69	1.00	North America	99.9	Temperate forest	98.7
Hoita	1.06	0.12–2.36	1.00	North America	99.7	Mediterranean North	50.6
Cullen	3.32	1.87–4.70	1.00	Africa	68.2	Mediterranean South	83.3

2.4. Discussion

2.4.1. Monophyly of the tribe Psoraleeae

The main objective of this study was to test previous hypotheses on generic relationships within Psoraleeae. The phylogeny inferred from the present study, including all the genera of the tribe Psoraleeae and out-groups from several legume tribes, shows that Psoraleeae genera together form a well-supported clade (Figure 3), confirming the findings of previous works (Egan & Crandall 2008a & b; Dludlu *et al.* 2013). Based on these results and that of Egan & Crandall (2008a) and Dludlu *et al.* (2013), it is unlikely that sampling more Phaseoleae will change the monophyly of Psoraleeae. However, previous studies at a tribal level have shown that Psoraleeae are embedded within the tribe Phaseoleae as sister to the subtribe Glycininae (Kajita *et al.* 2001; Wojciechowski *et al.* 2004; Stefanovic *et al.* 2009; LPWG 2013), thereby questioning the recognition of Psoraleeae as a tribe rather than a subtribe (Grimes 1990; Schrire 2005; LPWG, 2013). The results of this study cannot support the recognition of Psoraleeae as a subtribe of the Phaseoleae (Schrire 2005; Dludlu *et al.* 2013; LPWG, 2013) until more sampling of the various subtribes of Phaseoleae is achieved.

2.4.2. Generic relationships

Egan & Crandall (2008a) previously attempted to address generic relationships by sampling nearly all genera of Psoraleeae with the exception of the genus *Psoralea* but with a strong focus on North American Psoraleeae. Dludlu *et al.* (2013) employed three of Egan & Crandall's genetic markers and built on their dataset by strengthening taxonomic inclusion of the southern African genera, particularly *Otholobium* and *Psoralea*. Although Dludlu *et al.* (2013) attempted to sample the whole of Psoraleeae, they could not resolve whether the hypothesis of a sister relationship between *Otholobium* and *Psoralea* as proposed by Grimes (1990) should be accepted or not due to the poor resolution and support across their phylogeny, based on three markers. Here, the focus was on the whole of Psoraleeae by capitalising on the strength of Egan & Crandall's (2008a) genetic markers, employing five of their eight markers, and the strength of taxonomic sampling by Dludlu *et al.* (2013). I also included an unpublished data set of *Cullen* species placed on GenBank by Lori Kroiss *et al.* in 2009, a dataset which adds two more markers. To this foundational matrix, I added the *rbcLa* gene region and expanded the sampling of the Old World Psoraleeae (*Bituminaria*, *Cullen*, *Otholobium*, and *Psoralea*). The current data indicate that the two southern African genera (*Otholobium* and *Psoralea*) formed a clade, with *Otholobium* being paraphyletic with respect to *Psoralea* (Figure 3), a similar finding to that of Dludlu *et al.* (2013). *Otholobium* is also polyphyletic, with the SAm *Otholobium* resolved as sister to the genus *Bituminaria* and the *Psoralea* species resolved as a clade nested within the southern African *Otholobium* (Figure 3).

Egan & Crandall (2008a) also discovered the polyphyletic nature of *Otholobium* and proposed the splitting of *Otholobium* into two by geography. However, they did not revise the genus at that time, instead citing the need for further taxonomic inclusion of both South American and southern African species. Although the data from Dludlu *et al.* (2013) indicate that the South African *Otholobium* species do not form a clade with the South American species, they did not make any taxonomic changes in that regard, also citing the need for a more robust phylogeny, including all eight South American species, to be reconstructed. In this study, my results strongly support the Egan & Crandall (2008a) proposal to split *Otholobium* into two, as originally recognised by Stirton (1981), based on geography and the presence of unique morphological characters specific to each of the two taxa (Table 5). Although the current study did not improve the sampling of the SAm *Otholobium*, it is unlikely that doing so would change the position and the monophyly of both the SAf *Otholobium* and the SAm *Otholobium*. I therefore propose a new genus to accommodate all the eight species of the SAm *Otholobium*. This new genus will be described formally and separately after the inclusion of the remaining species.

Egan & Crandall (2008a) reported the monophyly of the New World Psoraleeae. However, the results of this study did not support the monophyly of the New World Psoraleeae. Instead, they formed a strongly supported clade together with the Mediterranean and European *Bituminaria* sister to the SAf *Otholobium* and *Psoralea*. The monophyly of the New World Psoraleeae could not be established by Dludlu *et al.* (2013) either. On the other hand, the phylogenetic position of the southern African *Otholobium* and *Psoralea* relative to the rest of Psoraleeae could not be definitively established by Dludlu *et al.* (2013) as their phylogeny resolved them (*Otholobium* and *Psoralea*) as sister to the genus *Hoita* but with no support. In this study, there is strong support for the SAf Psoraleeae (*Otholobium* and *Psoralea*) as a clade sister to the rest of Psoraleeae (Figure 3). It is worth mentioning here that neither the New World nor the Old World Psoraleeae are supported as a monophyletic group in this study as the African *Cullen* species were resolved as a basal grade in the clade comprising the rest of the *Cullen* species.

Table 5. Comparison of morphological characters of Psoraleeae genera. A= character state occurs in all species of the genus, B= character states occurs in some species of the genus, and X = character absent in all the species of the genus.

Character/Genera	***Pediomelum***	***Rupertia***	***Ladeania***	***Hoita***	***Orbexilum***	***SAm Otholobium***	***Otholobium***	***Psoralea***	***Bituminaria***	***Cullen***
Stem without secondary growth	A	A	A	A	A	X	B	B	A	B
Compound leaves	A	A	A	A	B	A	B	B	A	A
Leaves apex forming recurved mucro	X	X	X	X	X	X	A	X	X	X
Persistent bracts	A	X	X	B	B	B	A	B	A	A
Presence of cupulum	X	X	X	X	X	X	X	A	X	X
Flat or oblique Receptacle	A	X	A	A	A	A	A	B	A	A
Flowers borne in triplets subtended by a single bract	A	A	A	A	A	B	A	X	B	A
Continuous floral vasculature	A	A	A	A	A	A	A	A	X	X
Fruits with thick, ruggose walls	X	X	X	X	A	X	X	X	X	X
Single seeded in dehiscent fruit	X	A	A	A	A	A	A	A	A	A

Grimes (1997) presented a revision of the genus *Cullen* and reconstructed evolutionary relationships using morphological data, whereas Egan & Crandall (2008a & b) sampled five *Cullen* species in their molecular phylogeny. Although Kroiss *et al.* (2009) placed data for Australian and Asian *Cullen* on GenBank, they never published their findings. Here, I combined the taxonomic sampling of Kroiss *et al.* (2009) and Egan & Crandall (2008a & b), making the present study the first to reveal relationships in the genus *Cullen* using molecular phylogenetic data, sampling more than half the described species. The Mediterranean *C. americanum* forms a basal grade together with the African (*C. obtusifolia* and *C. biflorum*) / Eurasian (*C. corylifolium* and *C. drupacea*) species, all together sister to Australian *Cullen* (BS = 95%, PP = 1.00). Egan & Crandall (2008a) found similar evolutionary relationships. Grimes's (1997) phylogeny, based on morphological characters, found evidence for two clades (one including species the stems of which are branched at the base, and one in which the species are unbranched below), but these were not geographically aligned, and *C. americanum* was sister to *C. drupacea* but in a clade that included some Australian species.

The present study confirmed the phylogenetic position of *Bituminaria* as sister to SAm *Otholobium* (Figure 3 and 4) as shown by Dludlu *et al.* (2013). Despite the additional sample of *B. bituminosa* from Ukraine in this study, the sampling at the species level has not been improved beyond that of the Egan & Crandall (2008a) and Dludlu *et al.* (2013) since several new species of *Bituminaria* have recently been described (Minissale *et al.* 2013; del Galdo *et al.* 2015). There is therefore the need to increase the sampling of the genus so as to confirm its monophyly and its phylogenetic position within Psoraleeae.

2.4.3. Monophyly of the genus *Psoralea*

Psoralea is nested within *Otholobium*, confirming the paraphyly of the SAf *Otholobium* as shown by Dludlu *et al.* (2013). However, there are multiple distinct morphological characters that separate *Psoralea* from *Otholobium* as well as the rest of Psoraleeae. *Psoralea* can be distinguished based on the presence of a cupulum, the lack of three flowers per node subtended by a bract, the absence of recurved mucronate leaves, and the presence of proanthocyanidines (Bello *et al.* 2015b; Boardley *et al.* 1986). Given these diagnostic features and the fact that the present phylogeny strongly supports *Psoralea* as a clade (BS = 70%, PP = 1. 00), the classification by Stirton (1981), which recognises *Otholobium* as distinct from *Psoralea*, will be maintained in this study pending a better resolved phylogeny. More studies to resolve the polyphyly of *Otholobium* are in progress, and decisions on generic re-alignments will take into consideration the current paradigm to recognise monophyletic taxa while maximising clade stability and phenotypic diagnosability (Vences *et al.* 2013).

2.4.4. Ancestral state reconstructions

The ancestral states reconstruction was carried out to evaluate the utility of four morphological diagnostic characters used by Stirton (1981) and Grimes (1990) in delimiting the sections and genera of Psoraleeae. This exercise was useful especially in delimiting the two southern African genera (*Otholobium* and *Psoralea*), based on Dludlu *et al.* (2013). Our results showed that the ancestral growth habit in Psoraleeae is herbaceous. Three major shifts occurred from the herbaceous ancestor to the woody habit, with multiple secondary changes from woody to herbaceous in the southern African clade. The habit switch from herbaceousness back to the ancestral woody habit in angiosperms is supported in this study. Species of Psoraleeae are perennial herbs, subshrubs, sarmentose shrubs, shrubs or trees (Grimes 1990). In Africa and Australasia, both herb and shrub growth forms occur. The SAm species are shrubs or trees, and the NAm species are perennial herbs adapted to woodlands, grasslands, prairies, plains and deserts (Grimes 1990). Grimes (1990) showed that the radiation of many species into more xeric habitats led to a reduction of the plant body in the North American, African and Australian Psoraleeae.

The ancestral state for leaf type in Psoraleeae was shown to be compound leaves while simple leaves are a derived state (Figure 5b) as shown by Dludlu *et al.* (2013). All of the genera of Psoraleeae have compound leaves with the exception of the SAf Psoraleeae where a switch from compound to simple leaves occurred once in both genera (*Otholobium* and *Psoralea*). This shows that leaf type is of limited value for taxonomic grouping in Psoraleeae. It can only be a useful diagnostic character within each of the two SAf Psoraleeae (*Otholobium* and *Psoralea*) but not between the genera. A secondary loss of leaves at the adult stage among leaf species with leafy seedlings is seen to have evolved multiple independent times (Figure 5b) and is associated with members of the *Psoralea aphylla* complex (Dludlu *et al.* 2015). However, the taxonomic value of this character cannot be established here due to poor resolution within species of *Psoralea*. It is anticipated that members of this complex might form a clade in a well resolved phylogeny on the basis of this synapomorphy.

The ancestral character state reconstruction shows that the cupulum is a valuable diagnostic character in Psoraleeae. The lumping of *Hallia* into *Psoralea* (Salter 1939; Stirton 1989;

Crow *et al.* 1997) was based on this character, a pattern supported by the results here and that of Dludlu *et al.* (2013). The cupulum evolved only once in the Psoraleeae as a synapomorphy to *Psoralea*. This supports the recognition of *Psoralea* as a distinct genus.

Although indehiscence is the ancestral character state in Psoraleeae, dehiscent fruits evolved once in *Pediomelum.* However, this dehiscence is by transverse rupture of the pod and not along sutures (Grimes 1990). This is a valuable diagnostic character in Psoraleeae as it arises only once as synapomorphy in *Pediomelum.*

2.4.5. Divergence dates

The dates obtained from the present study confirmed the recent and rapid radiation of Psoraleeae as shown by previous studies (Egan & Crandall 2008b; Dludlu 2009; Li *et al.* 2013). In their treatment of NAm Psoraleeae, including only one SAf *Otholobium* and three *Cullen* species, Egan & Crandall (2008b) placed the origin of Psoraleeae at ca. 6.7 mya. Li *et al.* (2013) revised the estimate to 6.1 mya but included only six of Egan & Crandall (2008b) accessions. Based on my estimates, the crown node of Psoraleeae dates back to 8 mya (Figure 5). The disparity of the dates observed in this study compared with other studies might be influenced by a wide coverage of the majority of the species in Psoraleeae. The results here, showed that the diversification of several genera of Psoraleeae is estimated to have started in the early Pliocene (Figure 5), corresponding largely to the dates reported by Egan & Crandall (2008b), Dludlu (2009) and Li *et al.* (2013). These suggest that the divergence date estimates for Psoraleeae in the present study are reliable.

Of particular interest is the divergence and diversification of the SAf Psoraleeae, which began ca. 6.7 mya (Figure 5), with the split of *Otholobium*. This was followed by the split of *Psoralea* around 4 mya (Figure 5) constituting the main body of species richness in Psoraleeae as shown by the results of our biogeographic analyses in RASP. A similar pattern of diversification was observed in the NAm clade, where the main body of species richness is observed in one of the youngest lineage, *Pediomelum*, ca. 3 myo (Figure 5; Egan & Crandall 2008b). Within other phaseoloid legumes, the MRCA of *Pueraria loata* and *Pueraria phaseoloides* is ca.13.8 mya (Li *et al.* 2013; Wang *et al.* 2010). Compared with other Fabaceae co-occurring with Psoraleeae in the GCFR, the Crotalarieae are estimated to have diversified ca. 46.3 mya, while the Podalyrieae are placed ca. 44.6 mya (Edwards & Hawkins 2007). Indigofereae on the other hand, are estimated to have radiated ca. 20 mya (Schrire *et al.* 2009). Putting the age of Psoraleeae and its genera in the context of other phaseoloid legumes and the Fabaceae in general, suggests that Psoraleeae is the youngest clade in phaseoloid legumes and is an example of rapid recent radiations, findings which agree with several others (Egan & Crandall 2008b; Dludlu 2009; Li *et al.* 2013).

2.4.6. Biogeography

Biogeographic studies have suggested several ways to explain disjunct distributions among many plant lineages. Some studies have suggested vicariance events to explain the disjunct patterns in legume clades *Cercis*, *Schotia* and *Baudouinia* (Schrire *et al.* 2005a), whereas dispersal was the main mechanism involved in Fabeae (Schaefer *et al.* 2012). Dispersal is favoured to explain the current disjunct distribution pattern in Psoraleeae because the ages of the clades presented here and in previous studies on Psoraleeae are too recent to be the result of a vicariance event; its diversification occurs after the Tethyan vicariance (Schrire *et al.* 2005a & b).

The result of this study (Figure 7b) agrees with previous hypotheses (Schrire *et al.* 2005a & b), that the Psoraleeae is largely a temperate biome clade. Our broadly sampled molecular trees shows that the ancestor of Psoraleeae is either African (MP = 53%) or North American (MP = 40%), with radiations in each of these areas for their respective clades. The results here also revealed that intercontinental dispersals occurred from North America to South America, Europe, and Eurasia-Africa then to Australia (in the case of *Cullen*). This late Miocene temperate biome radiation is consistent with the southern African climate-driven, rampant speciation (Dupont *et al.* 2011; Linder & Verboom 2015) and the presence of glaciation-induced climate oscillations in North America (Egan & Crandall 2008a). A similar divergence and distribution pattern is observed in the warm-temperate *Sarcocornia* (Amaranthaceae) clade, which evolved during the Mid-Miocene from Eurasian ancestors and then dispersed into the Americas, southern Africa and Australia (Steffen *et al.* 2015).

The diversification of Psoraleeae in Africa started ca. 6.5 mya. This was followed by rapid speciations (ca. 124 speciation events). Climate changes in the late Miocene and edaphic heterogeneity have been proposed to explain the rapid diversification of plant lineages in the Cape (Linder 2003; Verboom *et al.* 2009). SAf Psoraleeae originated independently in the MS biome. The majority of the species in *Psoralea* (ca. 90%) maintained their original biome with only six dispersals into the TG biome (*P. glabra*, *P. latifolia*, *P. margaretiflora*, *P. arida*, *P. rhizotoma*, and *P. karooensis*), only two dispersals into the S biome (*P. glaucescens* and *P. tenuifolia*), and a single dispersal into MS/S biomes (*P. abbottii*). A similar pattern is observed in SAf *Otholobium*, where some species dispersed into the drier S biome, as seen in a small clade (*O. flexuosum*, *O. striatum*, *O. arborescens*, *O. argenteum* and *O. pustulatum*). Other dispersals into the S/TG biome were also observed in *Otholobium*, occurring in *O. caffrum*, *O. polystictum*, and *O. pictum*. These findings are consistent with several other studies (Schrire *et al.* 2005b, 2009; Dupont *et al.* 2011; Linder & Verboom 2015).

The *in situ* diversification of a clade consisting of the Americas (*Pediomelum*, *Rupertia*, *Ladeania*, *Orbexilum*, SAm *Otholobium* and *Hoita*), Mediterranean (*Bituminaria*) and Australian (*Cullen*) species started ca. 6.7 mya. Among the five genera of the NAm Psoraleeae, *Orbexilum* was the only clade shown to have colonised the TF biome (MP = 87.6%, Table 4) at ca. 3.1 mya. *Hoita* diversified ca. 1 mya within the MN biome. The subclade consisting of *Pediomelum*, *Rupertia*, and *Ladeania* originated in the S biome (MP = 60%, Figure 7) and then *Rupertia* dispersed into the MN biome. A S biome origin of legumes has been previously hypothesised (Schrire *et al.* 2005a & b) followed by dispersal to and diversification within other biomes. This is demonstrated here, where, a largely S *Pediomelum* clade, of ca. 10 species dispersed into the TG biome. Glaciation-induced climate changes during the Quaternary have been proposed to influence the diversification of the NAm Psoraleeae (Egan & Crandall 2008b).

2.5. Conclusions

This study supports the monophyly of Psoraleeae as a clade of phaseoloid legumes. The results here do not support the recognition of Psoraleeae as a subtribe of the Phaseoleae (Schrire 2005; Dludlu *et al.* 2013; LPWG 2013) until more sampling of the various subtribes of Phaseoleae is achieved. The phylogenetic relationship within the genus *Cullen*, revealed for the first time in this study, showed that the Mediterranean (*C. americanum*), African (*C. obtusifolia* and *C. biflorum*) and Eurasian (*C. corylifolium* and *C. drupacea*) taxa form a basal grade sister to Australian *Cullen* (BS = 95%, PP = 1.00). The addition of more molecular markers on the three previously studied by Dludlu *et al.* (2013) together with an extensive sampling of various genera helped resolve the phylogenetic position of southern African Psoraleeae (*Otholobium* and *Psoralea*) as sister to the rest of Psoraleeae. However, neither the New World nor the Old World Psoraleeae is supported as monophyletic, contrary to the results of Egan & Crandall (2008a & b) which showed a monophyletic New World Psoraleeae. Instead, the NAm Psoraleeae formed a strongly supported clade together with the European *Bituminaria*, sister to the SAf Psoraleeae (*Otholobium* and *Psoralea*). The African *Cullen* on the other hand formed a basal grade sister to the Australian *Cullen*. Furthermore, the polyphyly of *Otholobium* is reported here, supporting the findings of Egan & Crandall (2008a & b) and Dludlu *et al.* (2013). A new genus has been proposed in this study to accommodate all the species of SAm *Otholobium*, to be formally described in a later publication. The paraphyly of the SAf *Otholobium* as reported by Dludlu *et al.* (2013) has been confirmed in this study. However, due to multiple distinct morphological characters separating *Psoralea* from SAf *Otholobium* and the rest of Psoraleeae and strong support from the molecular phylogeny, the classification of Stirton (1981), which recognises *Otholobium* as distinct from *Psoralea*, is maintained in this study pending a more resolved phylogenetic relationship. More studies to resolve the polyphyly of *Otholobium* are in progress and decision on generic re-alignments will take into consideration the current paradigm to recognise monophyletic taxa while still maximising clade stability and phenotypic diagnosability (Vences *et al.* 2013).

According to the results of the ancestral character state reconstructions, the ancestors of Psoraleeae had an herbaceous growth habit, compound leaves, and indehiscent fruits. The presence of a cupulum and a secondary dehiscence by transverse rupture of the pod are synapomorphies unique to *Psoralea* and *Pediomelum* respectively. South African Psoraleeae evolved woodiness and diversified into the Mediterranean South biome, characterised by fynbos vegetation. More recently, SAf Psoraleeae dispersed into the Succulent biome, as well as occasional moving into afro-temperate highlands in Eastern Africa. In North America, earlier occupation was in the Succulent biome as herb, but multiple diversifications into temperate biomes and shifts into a woody life form were observed in South America.

This chapter suggests that Psoraleeae originated in the MS biome of southern Africa in Miocene, ca. 8.2 mya. It then dispersed into the S biome of Americas and MN biome of Mediterranean Europe ca. 6.7 mya. This was followed by a series of both local and transoceanic dispersal and speciation events. Among these were widespread dispersals of *Cullen* into the MS and TG biomes of Africa and transcontinental dispersal into TG and S biomes of Australia. Dispersal is favoured to explain the distribution of Psoraleeae due to its younger age. The current disjunct distribution pattern in Psoraleeae can be explained by ca. 27 dispersals events. The highest speciation events of ca. 124 occurred in Africa followed by 44 in North America, 18 in Australia and one each in South America and Mediterranean regions as shown by the current data. The results showed ca. 126 global dispersal events across the biomes. The highest speciation event of 111 occurred within the MS biome followed by 28 in S, 15 in TG, 11 in TF and 9 in MN biome. Climate changes in the late

Miocene, changes induced by glaciation during the Quaternary and environmental heterogeneity were proposed as the major drivers of speciation in *Psoralea* and Psoraleeae.

CHAPTER THREE: DNA BARCODES REVEAL MICROEVOLUTIONARY SIGNALS IN FIRE RESPONSE TRAIT IN TWO AFRICAN PSORALEEAE GENERA

Abstract

Large-scale DNA barcoding provides a new technique for species identification and evaluation of relationships across various levels (populations and species) and may reveal fundamental processes in recently diverged species. Here, we analysed DNA sequence variation in the recently diverged legumes from the Psoraleeae (Fabaceae) occurring in the Cape Floristic Region (CFR) of southern Africa to test the utility of DNA barcodes in species identification and discrimination. We further explored the phylogenetic signal on fire response trait (reseeding and resprouting) at species and generic levels. We showed that Psoraleoid legumes of the CFR exhibit a barcoding gap yielding the combination of *matK* and *rbcLa* (*matK* + *rbcLa*) dataset as a better barcode than single regions. We found a high score (100%) of correct identification of individuals to their respective genera but very low score (<50%) in identifying them to species. We found a less considerable match (54%) between genetic species and morphologically-delimited species. We also found that different lineages showed a weak but significant phylogenetic conservatism in their response to fire as reseeders or resprouters, with more clustering of resprouters than would be expected by chance. These novel microevolutionary patterns might be acting continuously over time to produce multi-scale regularities of biodiversity. This study provides the first insight into the DNA barcoding campaign of land plants in species identification and detection of phylogenetic signal in recently diverged lineages of the CFR.

Keywords: Fabaceae; *Otholobium*; *Psoralea*; reseeders; resprouters; South Africa.

3.1. Introduction

The primary goal of DNA barcoding is the identification of an unknown sample by correctly matching a specific genetic marker to a reference sequence library. However, DNA barcoding can also be used as a tool for addressing fundamental questions in ecology, evolution and conservation biology (Kress *et al.* 2014). For evolutionary biologists and ecologists, one of the goals of DNA barcoding is to understand the origin of species and the factors causing the difference in species richness in different biomes across the globe. Generally, the full diversity of species in most diverse habitats is still poorly known (Kress *et al.* 2014). The primary focus of this paper is to explore the application of DNA barcoding in some recently diverged lineages of an exceptionally unique fire derived biodiversity hotspot to determine its efficacy in species identification and detection of microevolutionary signals.

The Greater Cape Floristic Region is a home to Fynbos and the Succulent Karoo biomes− two major biodiversity hotspots located in the winter rainfall area of southern Africa (Myers *et al.* 2000) (Figure1). The Fynbos biome (also called the Cape Floristic Region, CFR) is famed for its high species diversity consisting of about 9 000 species of vascular plants packed into an area of 90 760 km^2 of which about 69% are endemic (Manning and Goldblatt 2012). The family Fabaceae consists of about 764 species in 43 genera. It is the second largest family in the CFR flora after Asteraceae. Three of the major clades of Fabaceae include the Crotalarieae (300 species), Podalyrieae (125 species) and African Psoraleeae (120 species). These legume lineages differ in their patterns of diversification, with Crotalarieae and Podalyrieae originating in the Eocene c. 40 Ma (Edwards and Hawkins 2007, Schnitzler *et al.* 2011) and the African Psoraleeae originating during the Pliocene c. 5 Ma (Egan and Crandall 2008b). This suggests that the African Psoraleeae is a young lineage which has undergone rapid recent radiation giving rise to c. 75 species of *Psoralea* L. and c. 53 species of *Otholobium* C.H.Stirt. (Manning and Goldblatt 2012; Stirton 2005). Majority of species in *Otholobium* and *Psoralea* have a narrow distribution and are frequently restricted to a single mountain stream or slope or a single soil type. In addition, several species are listed in the IUCN Red List under different levels of conservation categories ranging from extinct in the wild (e.g. *Psoralea gueinzii* and *P. cataracta*), to endangered (e.g. *Otholobium bowieanum*, *O. incanum, P. fascicularis* and *P. filifolia*) and vulnerable (*O. hamatum*, *O. venustum, P. abbottii* and *P. alata*) (Raimondo *et al.* 2009).

Fynbos is a fire prone vegetation that requires regular burning for its persistence. The high species richness in the Fynbos biome has been ascribed to fire (Cowling *et al.* 1996, Linder

Figure 1. Map of the GCFR showing the Fynbos and the Succulent Karoo Biomes constructed based on Mucina and Rutherford (2006).

Figure 2. A recent fire burn in the Cape Fynbos, Table Mountain on 5 March 2015. Photograph: A Bello

2003, Power *et al.* 2011). Plants adapt to fires in two major ways: as resprouters or reseeders (Bell 2001). Resprouting plants survive fire as individuals and then replace the lost structures by resprouting from surviving tissues. Conversely, reseeding individuals are often killed by fire (Figure 2) and the population is re-established by a new generation growing from seeds (Bell 2001). Fire-survival and regeneration strategies of plants have been the subject of numerous studies (e.g. Keeley 1977; Bond 1985; Le Maitre and Midgley 1992; Pausas and Keeley 2014; Schutte *et al.* 1995; Scott *et al.* 2014). Cowling (1987) postulated that the high species diversity in the Gondwanan floras (Australian kwongan and Cape fynbos) may be ascribed to recurrent fires, edaphic specialisation and short dispersal distance. There are noticeable differences in the allocation of resources to reserve storage, vegetative growth and reproductive effort linked with these fire survival strategies (Bond and van Wilgen 1996, Bond and Midgley 2001, Bell 2001, Scott *et al.* 2014). For example, while reseeders are generally characterised by a shorter life span, they tend to grow rapidly and taller with much allocation of resources predominantly above ground. Resprouters on the other hand have longer life spans, slower growth, produce fewer seeds and have a below ground resource allocation in starch-rich lignotubers (Bell and Ojeda 1999; Hansen *et al.* 1991). Reseeders produce larger numbers of viable seeds than do resprouters due to their greater reliance on seed for survival (Hansen *et al.* 1991; Bell 2001), resulting in elevated post-fire recruitment. There are also reported differences in seed yield and quality with reseeders having higher N and P concentrations in the seeds than those of congeneric resprouters (Hansen *et al.* 1991). Other differences include nutritional requirements with reseeders requiring more nutrients than the resprouters due to the high nutritional costs of seed production and growth (Hansen *et al.* 1991; Bell 2001). These strategies influence speciation rates in woody genera in the fynbos (Wells 1969, Litsios *et al.* 2013), with reseeders shown to have higher diversification rates than resprouters (Litsios *et al.* 2013). Other studies have shown that fire survival and regeneration strategy (reseeding/resprouting) is a character of taxonomic, ecological and evolutionary importance in Fynbos legumes (Schutte *et al.* 1995; Litsios *et al.* 2013; Scott *et al.* 2014).

Traditionally species identification depends primarily on morphological features (morphospecies). As molecular data became increasingly available and new techniques such as DNA barcoding emerged, species identification is becoming fast, reliable and more accurate. Here, we use *matK* and *rbcLa* and the combination of the two regions (*matK* + *rbcLa*), based on their recognition as core plant barcode markers by the Consortium for the Barcode of Life Plant Working Group (CBOL 2009) to (i) test their efficacy in identifying species of two southern African Psoraleoid genera (*Otholobium* and *Psoralea*); (ii) explore the potential of the DNA barcode markers in grouping Psoraleoid legumes sequences into molecular operational taxonomic units (MOTUs) or genetic species units and; (iii) test the power of DNA barcodes in revealing microevolutionary patterns including fire survival and regeneration strategies. The genera *Otholobium* and *Psoralea* were chosen for this study because they both have species with reseeding and resprouting mode of regeneration (Figure 3). Furthermore, although the two genera are closely related (Dludlu *et al.* 2013), they differ markedly in terms of their morphology and ecology. For example, *Otholobium* species differ from *Psoralea* by the absence of a cupulum on the flower pedicel (unique structure in *Psoralea*, Tucker and Stirton 1990); possession of entire obovate to oblanceolate recurved mucronate leaflets (1–3); and inflorescences characterised by bracteate triplets of flowers, with each triplet subtended by a single variously shaped bract (Stirton 1981). Leaves of *Psoralea* range from 1–19-foliolate compound structures or reduced to small scale-like structures with only *P. aculeata* having a recurved mucro (Stirton 1989; Manning and Goldblatt 2012) and each flower is subtended by a pair of free minute bracts. The two genera also differ in terms of habitat preferences. Eighty percent of *Psoralea* species inhabit seeps, marshes, riverbanks and/or moist, mist laden high-altitude habitats, while *Otholobium* species occur predominantly in drier habitats, with only 11% of species occupying the moist habitats favoured by *Psoralea* (Stirton 1989; Manning and Goldblatt 2012).

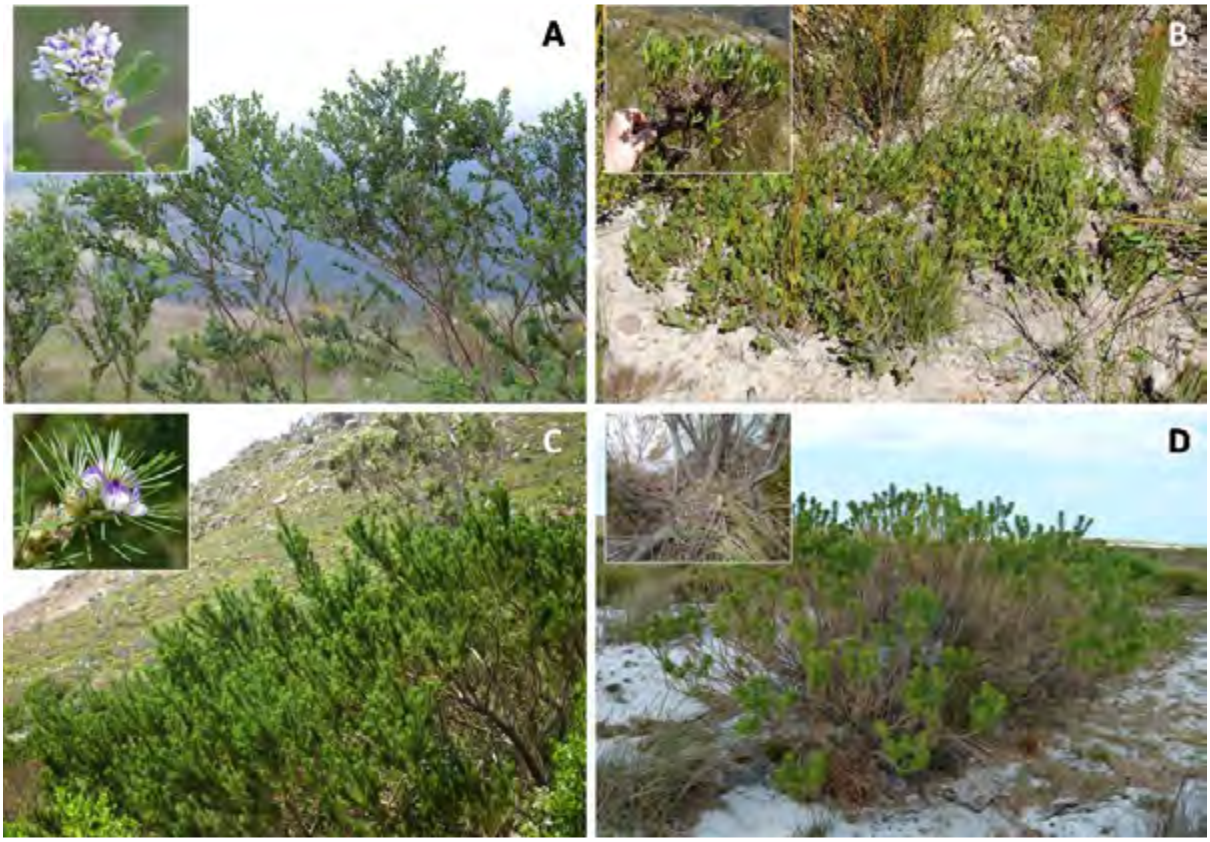


Figure 3. Habit in *Otholobium* and *Psoralea* species: (A) reseeding, *O. spicatum*; (B) resprouting, *O. rotundifolium*; (C) reseeding, *P. pinnata*; (D) resprouting, *Psoralea* sp. nov. Photographs: C.H.S. (A–C) and A.B. (D).

3.2. Methods

3.2.1. Taxon sampling

We collected 172 samples representing 26 species of *Otholobium* and 43 species of *Psoralea* across their distribution range in the CFR. Where possible, each species was represented by two or more different samples. In all, we collected 72 samples of *Otholobium* and 100 of *Psoralea* (voucher specimens are deposited at the Bolus Herbarium and listed in Table 1). Of these samples, 23 out of the 26 species of *Otholobium* and 26 out of 43 species of *Psoralea* are represented by more than one sample. Only samples for which sequences for both genes (*matK* and *rbcLa*) are available were included in the analyses. The final dataset used in the analyses included 4 reseeding (27 samples) and 22 resprouting (35 samples) species of *Otholobium*, and 35 (43 samples) reseeding and 8 (56 samples) resprouting species of *Psoralea*. Information on fire response strategy was extracted from Manning and Goldblatt (2012); Snijman (2013) and Stirton (1989). To our knowledge, no species included in our analysis show both fire response strategies in wild populations. Collection details including GPS coordinates, altitude and photographs of specimens are available online in the Barcode of Life Data Systems (BOLD; www.boldsystems.org) together with DNA sequences.

3.2.2. DNA extraction, sequencing and alignment

All the samples were sent to the Canadian Centre for DNA Barcoding (CCDB) in Canada, where total DNA was extracted and the two core DNA barcodes (*matK* and *rbcLa*) were sequenced according to standard CCDB protocols (Ivanova *et al.* 2006). Sequence alignment was performed using Multiple Sequence Comparison by Log Expectation (MUSCLE vs. 3.8.31, Edgar 2004) plugin in Geneious v.8.0.4 (Kearse *et al.* 2012) and manually adjusted using MESQUITE v.2.5 (Maddison and Maddison 2008). The two regions were aligned separately and then combined.

3.2.3. Evaluation of DNA barcodes

First, we evaluated the performance of the DNA markers (*matK, rbcLa* and *matK* + *rbcLa*) in species identification and delimitation of African Psoraleoid legumes at species and generic levels by applying two criteria commonly used to evaluate the utility of the DNA barcodes in species discrimination: the barcode gap of Meyer and Paulay (2005) and discriminatory power (Hebert *et al.* 2004). Barcode gap was assessed by comparing intraspecific variation (i.e. the amount of genetic variation within species) to interspecific variation (between species). A good barcode should exhibit a significant gap, meaning that sequence variation within species should be significantly lower than between species. Statistical significance between intra- and interspecific variation was assessed using Wilcoxon test in R (R Core Team 2013).

Table 1. List of voucher specimens and the DNA sequence BOLD ID reference number.

Taxon name	**Collector**	**Number**	**BOLD ID**	**Herbarium**	**Distribution**
Otholobium acuminatum	Muasya & Stirton	AMM3850	FAUCT199-11	BOL	Africa
Otholobium acuminatum	Muasya & Stirton	AMM3603	FAUCT144-11	BOL	Africa
Otholobium arborescens	Muasya & Stirton	AMM3279	FAUCT051-11	BOL	Africa
Otholobium beanianum sp. nov.	Muasya & Stirton	AMM3350	FAUCT067-11	BOL	Africa
Otholobium bracteolatum	Muasya & Stirton	AMM3963	FAUCT229-11	BOL	Africa
Otholobium bracteolatum	Muasya & Stirton	AMM3164	FAUCT002-11	BOL	Africa
Otholobium bracteolatum	Muasya & Stirton	AMM3879	FAUCT208-11	BOL	Africa
Otholobium bracteolatum ssp. limnophilum ssp. nov.	Muasya & Stirton	Stirton13155	FAUCT367-11	BOL	Africa
Otholobium bracteolatum ssp. limnophilum ssp. nov.	Muasya & Stirton	AMM3204	FAUCT030-11	BOL	Africa
Otholobium candicans	Muasya & Stirton	AMM3911	FAUCT223-11	BOL	Africa
Otholobium candicans	Muasya & Stirton	AMM3369	FAUCT072-11	BOL	Africa
Otholobium candicans	Muasya & Stirton	AMM3563	FAUCT130-11	BOL	Africa
Otholobium crewii sp. nov.	Muasya & Stirton	AMM3264	FAUCT041-11	BOL	Africa
Otholobium flexuosum	Muasya & Stirton	AMM3276	FAUCT049-11	BOL	Africa
Otholobium flexuosum	Muasya & Stirton	AMM3280	FAUCT052-11	BOL	Africa
Otholobium fruticans	Muasya & Stirton	AMM3480	FAUCT106-11	BOL	Africa
Otholobium fruticans	Muasya & Stirton	AMM3397	FAUCT081-11	BOL	Africa
Otholobium hamatum	Muasya & Stirton	AMM3310	FAUCT060-11	BOL	Africa

Otholobium hamatum	Muasya & Stirton	AMM3306	FAUCT059-11	BOL	Africa
Otholobium hirtum	Muasya & Stirton	AMM3326	FAUCT063-11	BOL	Africa
Otholobium hirtum	Muasya & Stirton	AMM3991	FAUCT232-11	BOL	Africa
Otholobium hirtum	Muasya & Stirton	AMM3190	FAUCT018-11	BOL	Africa
Otholobium hirtum	Muasya & Stirton	AMM3373	FAUCT074-11	BOL	Africa
Otholobium hirtum	Muasya & Stirton	AMM3372	FAUCT073-11	BOL	Africa
Otholobium hirtum	Muasya & Stirton	AMM3499	FAUCT112-11	BOL	Africa
Otholobium hirtum	Muasya & Stirton	AMM3878	FAUCT207-11	BOL	Africa
Otholobium lucens sp. nov.	Muasya & Stirton	AMM3570	FAUCT133-11	BOL	Africa
Otholobium mundianum	Muasya & Stirton	AMM3885	FAUCT211-11	BOL	Africa
Otholobium obliquum	Muasya & Stirton	AMM3198.1	FAUCT023-11	BOL	Africa
Otholobium parviflorum	Muasya & Stirton	AMM3199	FAUCT024-11	BOL	Africa
Otholobium parviflorum	Muasya & Stirton	AMM3542	FAUCT119-11	BOL	Africa
Otholobium prodiens	Muasya & Stirton	AMM3845	FAUCT196-11	BOL	Africa
Otholobium prodiens	Muasya & Stirton	AMM3854	FAUCT201-11	BOL	Africa
Otholobium pustulatum	Muasya & Stirton	AMM3286	FAUCT054-11	BOL	Africa
Otholobium rotundifolium	Muasya & Stirton	AMM3929	FAUCT227-11	BOL	Africa
Otholobium rotundifolium	Muasya & Stirton	AMM3173	FAUCT009-11	BOL	Africa
Otholobium rubicundum	Muasya & Stirton	AMM5982	FAUCT359-11	BOL	Africa
Otholobium schutteae sp. nov.	Muasya & Stirton	AMM3575	FAUCT134-11	BOL	Africa
Otholobium spicatum	Muasya & Stirton	AMM3445	FAUCT097-11	BOL	Africa
Otholobium spicatum	Muasya & Stirton	AMM3498	FAUCT111-11	BOL	Africa
Otholobium spicatum	Muasya & Stirton	AMM3906	FAUCT220-11	BOL	Africa
Otholobium spicatum	Muasya & Stirton	AMM3568	FAUCT132-11	BOL	Africa
Otholobium stachyerum	Muasya & Stirton	AMM3837	FAUCT194-11	BOL	Africa
Otholobium stachyerum	Muasya & Stirton	AMM3872	FAUCT206-11	BOL	Africa
Otholobium stachyerum	Muasya & Stirton	AMM3791	FAUCT183-11	BOL	Africa
Otholobium stachyerum	Muasya & Stirton	AMM3604	FAUCT145-11	BOL	Africa
Otholobium stachyerum	Muasya & Stirton	AMM3851	FAUCT200-11	BOL	Africa
Otholobium striatum	Muasya & Stirton	AMM3339	FAUCT064-11	BOL	Africa
Otholobium striatum	Muasya & Stirton	AMM3363	FAUCT071-11	BOL	Africa
Otholobium striatum	Muasya & Stirton	AMM3561	FAUCT129-11	BOL	Africa
Otholobium striatum	Muasya & Stirton	AMM4106	FAUCT247-11	BOL	Africa
Otholobium striatum	Muasya & Stirton	AMM3351	FAUCT068-11	BOL	Africa
Otholobium striatum	Muasya & Stirton	AMM3318	FAUCT062-11	BOL	Africa
Otholobium thomii	Muasya & Stirton	AMM3187	FAUCT016-11	BOL	Africa
Otholobium uncinatum	Muasya & Stirton	AMM3175	FAUCT010-11	BOL	Africa
Otholobium uncinatum	Muasya & Stirton	AMM3263	FAUCT040-11	BOL	Africa
Otholobium uncinatum	Muasya & Stirton	AMM3261	FAUCT038-11	BOL	Africa
Otholobium velutinum	Muasya & Stirton	Stirton13106	FAUCT362-11	BOL	Africa
Otholobium virgatum	Muasya & Stirton	AMM3908	FAUCT222-11	BOL	Africa
Otholobium virgatum	Muasya & Stirton	AMM3395	FAUCT079-11	BOL	Africa
Otholobium virgatum	Muasya & Stirton	AMM3163	FAUCT001-11	BOL	Africa
Otholobium virgatum	Muasya & Stirton	AMM3191	FAUCT019-11	BOL	Africa
Psoralea aculeata	Muasya & Stirton	AMM3183	FAUCT012-11	BOL	Africa
Psoralea aculeata	Muasya & Stirton	AMM3405	FAUCT088-11	BOL	Africa
Psoralea aculeata	Muasya & Stirton	AMM3550	FAUCT124-11	BOL	Africa
Psoralea aculeata	Muasya & Stirton	AMM3170	FAUCT006-11	BOL	Africa
Psoralea affinis	Muasya & Stirton	AMM3903.2	FAUCT215-11	BOL	Africa
Psoralea affinis	Muasya & Stirton	AMM3868	FAUCT203-11	BOL	Africa
Psoralea alata	Muasya & Stirton	AMM3262	FAUCT039-11	BOL	Africa
Psoralea alata	Muasya & Stirton	AMM3398	FAUCT082-11	BOL	Africa
Psoralea alata	Muasya & Stirton	AMM3880	FAUCT209-11	BOL	Africa
Psoralea alata	Muasya & Stirton	AMM3901	FAUCT213-11	BOL	Africa

Psoralea aphylla	Muasya & Stirton	AMM3400	FAUCT084-11	BOL	Africa
Psoralea arborea	Muasya & Stirton	AMM3212	FAUCT032-11	BOL	Africa
Psoralea arborea	Muasya & Stirton	AMM3248	FAUCT037-11	BOL	Africa
Psoralea arida sp. nov.	Muasya & Stirton	AMM3526	FAUCT113-11	BOL	Africa
Psoralea arida sp. nov.	Muasya & Stirton	AMM4098	FAUCT246-11	BOL	Africa
Psoralea asarina	Muasya & Stirton	AMM3907	FAUCT221-11	BOL	Africa
Psoralea asarina	Muasya & Stirton	AMM3476	FAUCT105-11	BOL	Africa
Psoralea asarina	Muasya & Stirton	AMM3552	FAUCT126-11	BOL	Africa
Psoralea axillaris	Muasya & Stirton	AMM3834	FAUCT192-11	BOL	Africa
Psoralea axillaris	Muasya & Stirton	AMM3848	FAUCT198-11	BOL	Africa
Psoralea axillaris	Muasya & Stirton	AMM3827	FAUCT191-11	BOL	Africa
Psoralea axillaris	Muasya & Stirton	AMM5874	FAUCT356-12	BOL	Africa
Psoralea brilliantissima sp. nov.	Muasya & Stirton	AMM3621	FAUCT152-11	BOL	Africa
Psoralea cf. latifolia	Muasya & Stirton	AMM4028	FAUCT234-11	BOL	Africa
Psoralea congesta	Muasya & Stirton	AMM5462	FAUCT328-11	BOL	Africa
Psoralea elegans sp. nov.	Muasya & Stirton	AMM3591	FAUCT139-11	BOL	Africa
Psoralea filifolia	Muasya & Stirton	AMM4321	FAUCT278-11	BOL	Africa
Psoralea fleta	Muasya & Stirton	AMM3273	FAUCT047-11	BOL	Africa
Psoralea fleta	Muasya & Stirton	AMM3341	FAUCT065-11	BOL	Africa
Psoralea fleta	Muasya & Stirton	AMM3342	FAUCT066-11	BOL	Africa
Psoralea forbesii sp. nov.	Muasya & Stirton	AMM3578	FAUCT135-11	BOL	Africa
Psoralea forbesii sp. nov.	Muasya & Stirton	AMM3592	FAUCT140-11	BOL	Africa
Psoralea gigantean	Muasya & Stirton	AMM3203	FAUCT029-11	BOL	Africa
Psoralea glaucescens	Muasya & Stirton	AMM3289	FAUCT056-11	BOL	Africa
Psoralea glaucescens	Muasya & Stirton	AMM3312	FAUCT061-11	BOL	Africa
Psoralea imbricate	Muasya & Stirton	AMM4030	FAUCT235-11	BOL	Africa
Psoralea imbricate	Muasya & Stirton	AMM3439	FAUCT094-11	BOL	Africa
Psoralea imbricate	Muasya & Stirton	AMM3544	FAUCT120-11	BOL	Africa
Psoralea imbricate	Muasya & Stirton	AMM3904	FAUCT218-11	BOL	Africa
Psoralea imbricate	Muasya & Stirton	AMM3396	FAUCT080-11	BOL	Africa
Psoralea imbricate	Muasya & Stirton	AMM3399	FAUCT083-11	BOL	Africa
Psoralea imminens	Muasya & Stirton	AMM3596	FAUCT141-11	BOL	Africa
Psoralea ivumba sp. nov.	Muasya & Stirton	AMM3374	FAUCT075-11	BOL	Africa
Psoralea ivumba sp. nov.	Muasya & Stirton	AMM3165	FAUCT003-11	BOL	Africa
Psoralea keetii	Muasya & Stirton	AMM3599	FAUCT143-11	BOL	Africa
Psoralea laevigata	Muasya & Stirton	AMM3457	FAUCT099-11	BOL	Africa
Psoralea laxa	Muasya & Stirton	AMM3646	FAUCT156-11	BOL	Africa
Psoralea laxa	Muasya & Stirton	AMM4325	FAUCT279-11	BOL	Africa
Psoralea laxa	Muasya & Stirton	AMM3548	FAUCT122-11	BOL	Africa
Psoralea laxa	Muasya & Stirton	AMM3870	FAUCT205-11	BOL	Africa
Psoralea muirii sp. nov.	Muasya & Stirton	AMM4181	FAUCT257-11	BOL	Africa
Psoralea odoratissima	Muasya & Stirton	AMM3532	FAUCT116-11	BOL	Africa
Psoralea odoratissima	Muasya & Stirton	AMM3557	FAUCT127-11	BOL	Africa
Psoralea oligophylla	Muasya & Stirton	AMM3798	FAUCT185-11	BOL	Africa
Psoralea oreophila	Muasya & Stirton	AMM3463	FAUCT102-11	BOL	Africa
Psoralea oreophila	Muasya & Stirton	AMM3464	FAUCT103-11	BOL	Africa
Psoralea oreopola sp. nov.	Muasya & Stirton	AMM4370	FAUCT283-11	BOL	Africa
Psoralea oreopola sp. nov.	Muasya & Stirton	AMM4376	FAUCT285-11	BOL	Africa
Psoralea oreopola sp. nov.	Muasya & Stirton	AMM3271	FAUCT044-11	BOL	Africa
Psoralea pinnata	Muasya & Stirton	AMM3169	FAUCT005-11	BOL	Africa
Psoralea pinnata	Muasya & Stirton	AMM3403	FAUCT086-11	BOL	Africa
Psoralea pinnata	Muasya & Stirton	AMM3186	FAUCT015-11	BOL	Africa
Psoralea pinnata	Muasya & Stirton	AMM3547	FAUCT121-11	BOL	Africa
Psoralea pinnata	Muasya & Stirton	AMM3172	FAUCT008-11	BOL	Africa

Psoralea pinnata	Muasya & Stirton	AMM3171	FAUCT007-11	BOL	Africa
Psoralea pinnata	Muasya & Stirton	AMM3189	FAUCT017-11	BOL	Africa
Psoralea plauta	Muasya & Stirton	AMM3611	FAUCT149-11	BOL	Africa
Psoralea pullata	Muasya & Stirton	AMM3178	FAUCT011-11	BOL	Africa
Psoralea pullata	Muasya & Stirton	AMM3903.1	FAUCT214-11	BOL	Africa
Psoralea repens	Muasya & Stirton	AMM3809	FAUCT186-11	BOL	Africa
Psoralea repens	Muasya & Stirton	AMM3168	FAUCT004-11	BOL	Africa
Psoralea restioides	Muasya & Stirton	AMM3216	FAUCT033-11	BOL	Africa
Psoralea rhizotoma	Muasya & Stirton	AMM3659	FAUCT158-11	BOL	Africa
Psoralea rigidula	Muasya & Stirton	AMM3390	FAUCT077-11	BOL	Africa
Psoralea sordid	Muasya & Stirton	AMM3579	FAUCT136-11	BOL	Africa
Psoralea sordid	Muasya & Stirton	AMM3580	FAUCT137-11	BOL	Africa
Psoralea sparsa sp. nov.	Muasya & Stirton	AMM3567	FAUCT131-11	BOL	Africa
Psoralea speciosa	Muasya & Stirton	AMM3458	FAUCT100-11	BOL	Africa
Psoralea speciosa	Muasya & Stirton	AMM3610	FAUCT148-11	BOL	Africa
Psoralea speciosa	Muasya & Stirton	AMM3456	FAUCT098-11	BOL	Africa
Psoralea speciosa	Muasya & Stirton	AMM3607	FAUCT146-11	BOL	Africa
Psoralea suaveolens	Muasya & Stirton	AMM4396	FAUCT286-11	BOL	Africa
Psoralea suaveolens	Muasya & Stirton	AMM4975	FAUCT303-11	BOL	Africa
Psoralea triflora	Muasya & Stirton	AMM3862	FAUCT202-11	BOL	Africa
Psoralea usitata	Muasya & Stirton	AMM4344	FAUCT281-11	BOL	Africa
Psoralea usitata	Muasya & Stirton	AMM4071	FAUCT244-11	BOL	Africa
Psoralea usitata	Muasya & Stirton	AMM3440	FAUCT095-11	BOL	Africa
Psoralea usitata	Muasya & Stirton	AMM3528	FAUCT114-11	BOL	Africa
Psoralea usitata	Muasya & Stirton	AMM3541	FAUCT118-11	BOL	Africa
Psoralea usitata	Muasya & Stirton	AMM3194	FAUCT020-11	BOL	Africa
Psoralea usitata	Muasya & Stirton	AMM3414	FAUCT092-11	BOL	Africa
Psoralea usitata vigilans sp. nov.	Muasya & Stirton	AMM3415	FAUCT093-11	BOL	Africa
Psoralea usitata vigilans sp. nov.	Muasya & Stirton	AMM4340	FAUCT280-11	BOL	Africa
Psoralea verrucosa	Muasya & Stirton	AMM3357	FAUCT070-11	BOL	Africa
Psoralea verrucosa	Muasya & Stirton	AMM3905	FAUCT219-11	BOL	Africa
Psoralea verrucosa	Muasya & Stirton	AMM3353	FAUCT069-11	BOL	Africa
Psoralea verrucosa	Muasya & Stirton	AMM3269	FAUCT042-11	BOL	Africa
Psoralea verrucosa	Muasya & Stirton	AMM4371	FAUCT284-11	BOL	Africa
Psoralea verrucosa	Muasya & Stirton	AMM3270	FAUCT043-11	BOL	Africa

The discriminatory power of DNA barcoding was tested by evaluating the proportion of correct species identification at different taxonomic level (species and generic) using *matK*, *rbcLa* and *matK* + *rbcLa* regions. All sequences were labelled according to the names of the species from which the sequences were generated. The test of discriminatory power was carried out using two methods: the “best close match” (Meier *et al.* 2006) and the “near neighbour” using the functions *bestCloseMatch* and *nearNeighbour* implemented in the R package Spider (Brown *et al.* 2012). Before the test, we determined the optimised genetic distance suitable as threshold for taxon identification using the function *localMinima* also implemented in Spider (Brown *et al.* 2012).

The function *bestCloseMatch* conducts the “best close match” analysis (Meier *et al.* 2006) by searching for the closest individual in the dataset. If the closest individual is within a given threshold, the outcome is scored as “correct”; and if it is further, then the result is “no ID” (no identification). If more than one species are tied for closest match, the outcome of the test is an “ambiguous” identification. When all matches within the threshold are different species to the query, the result is scored as “incorrect”. The *nearNeighbour* function finds the closest individual and returns the score “true” (similar to “correct” in the *bestCloseMatch* method) if their names are the same, but if the names are different, the outcome is scored as “false” (similar to “incorrect” in the *bestCloseMatch* method).

3.2.4. Barcode test of species delimitation

Apart from investigating the potential of DNA markers in identifying species, we explored their ability in assigning morphologically-delimited species into genetic units i.e. molecular operational taxonomic units “MOTUs” or “genetic species” (sensu Saunders and McDevit 2013). We considered MOTUs as groupings or clusters of specimens that fall around a medoid. The goal is to verify the optimal number of clusters (species) that may be inferred from the pair wise genetic distance matrices of Psoraleoid legumes. A match between our genetic species and morphologically-delimited species would indicate that one could serve as a surrogate for the other (see Stahlhut *et al.* 2013), and thus lend support to the discriminatory power of DNA barcoding. We used partition around medoids (PAM) approach using the R package Cluster (Maechler *et al.* 2015) (R Core Team 2015). Our decision in choosing PAM was made after testing the performance of several clustering algorithms including: "Agglomerative Nesting (agnes)", "Divisive Analysis Clustering (diana)" and "Fuzzy Analysis Clustering (fanny)". Results from these other approaches were not reported for at least one of two main reasons. Firstly, they yielded identical results to PAM and are less straight forward to explain. For example, fanny does not produce unique clusters. Instead, it groups each species (probabilistically) to multiple clusters. The second reason was that the methodologies employed by some of the algorithms do not easily accommodate the restriction of cluster sizes.

The PAM algorithm works as follows: given a specific number of clusters (k), desired from a distance matrix, PAM searches for species (here referred to as medoids) that are representative of the data. The number of medoids sought is usually the same as the number of desired clusters k. Each cluster is then constructed such that the distance of any other sample, in the cluster, from its medoid is minimal. Cluster sizes between 2 and 69 were first investigated for each distance matrix. An optimal cluster size was then chosen as the one that yielded the maximum *silhouette coefficient* (Kaufman and Rousseeuw 1990). A silhouette coefficient measures the quality of clustering, derived as an average of the silhouette widths over all species. We used the silhouette width as an aggregate of a measure of the suitability of a cluster for each observation it contains relative to the next best cluster for the observations. Silhouette coefficients range between 0 and 1.

3.2.5. Barcode test for phylogenetic signal

We explored the potential of the DNA barcode data to reveal microevolutionary patterns by testing for phylogenetic signal in the affinity of lineages to fire survival and regeneration strategies. We used a phylogeny of the southern African Psoraleoid species and a binary matrix of reseeders versus resprouters. The phylogeny was reconstructed using a combination of *matK* and *rbcLa* datasets, based on a maximum likelihood approach (Stamatakis *et al.* 2008), enforcing topological constraints from a consensus tree of the Bayesian analysis of the dataset. We used the GTR+G+I substitution model based on the result of AIC from Modeltest v.2.3 (Nylander 2004), and ran 1000 maximum likelihood (ML) searches. Phylogenetic signal was tested on the ML best tree and binary matrix of reseeders versus resprouters using the D statistics of Fritz and Purvis (2010) in the R package Caper (Orme *et al.* 2012). The D statistics calculates the sum of changes of a binary trait along the branches of a phylogeny, and compares it with a random model and clumping expected under a Brownian evolution. Significance was assessed by shuffling the trait values 999 times at the tips of the phylogeny. $D = 1$ corresponds to a random distribution of traits at the tip of the phylogeny; $D = 0$ corresponds to a Brownian motion model (Fritz and Purvis 2010).

3.3. Results

For the core barcode loci, we obtained 332 sequences (165 and 167 for *matK* and *rbcLa* respectively) from 172 specimens representing 72 *Otholobium* and 100 *Psoralea*. Sequence recoverability was higher for *rbcLa* than *matK* (98.1% and 97.1% of specimens, respectively, Figure 4). The combined *matK* + *rbcLa* sequence data were obtained from 98.1% of the specimens sampled (Figure 4). For *rbcLa,* we recovered 95.7% of the 69 species sequenced, and 93.6% for *matK* and when combined with *rbcLa* i.e. *matK* + *rbcLa*. Both barcodes combined yielded a total of 1326 base pairs (770 bp for *matK* and 549 bp for *rbcLa*).

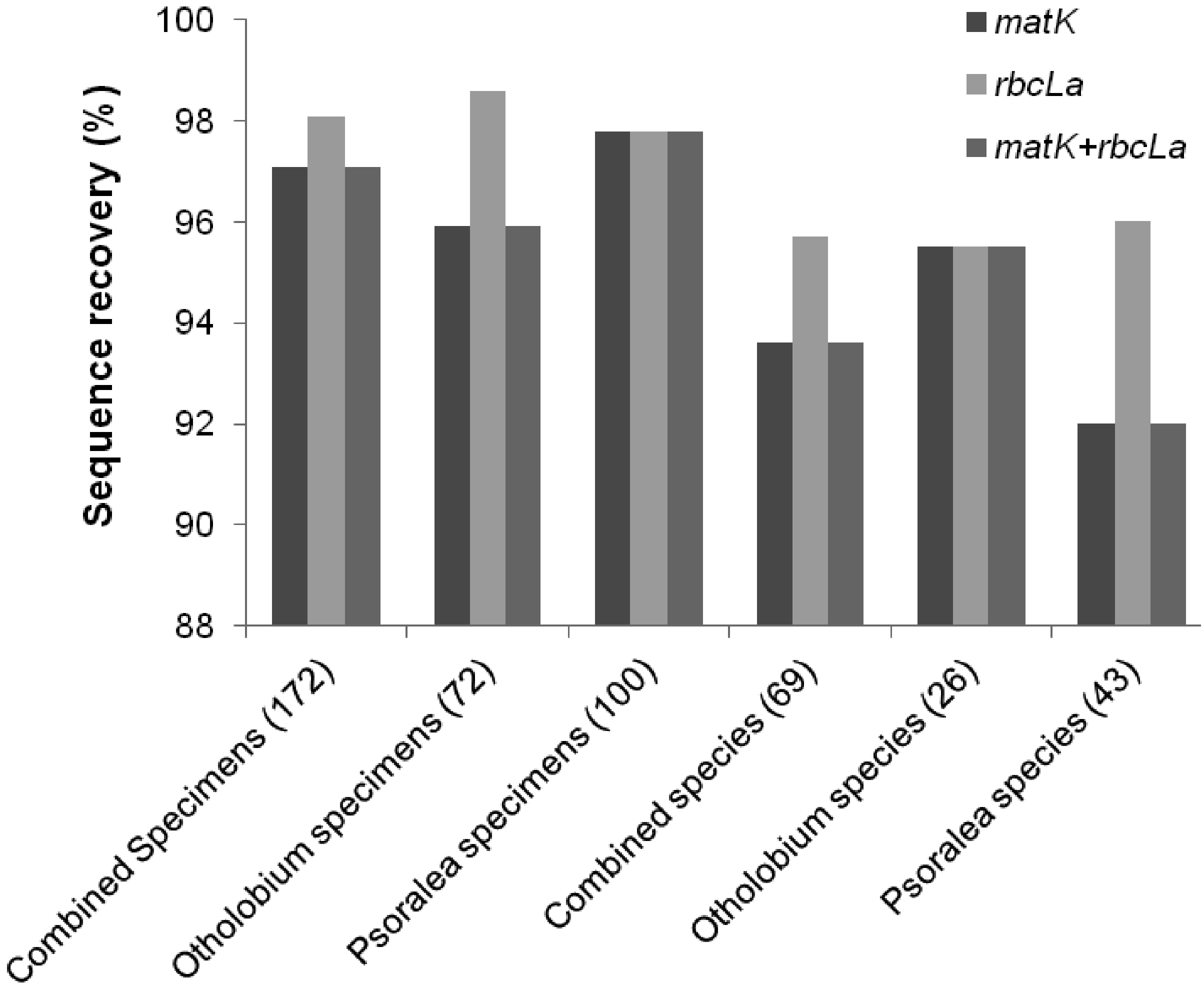


Figure 4. Percentage of specimens and species of *Otholobium* and *Psoralea* from which *rbcL* and *matK* barcodes were recovered. Numbers in parentheses are the total number of individuals (specimens, species).

The mean interspecific distances for the single and combined regions are lower than 1%, ranging from 0.002013 in *rbcLa* to 0.008612 in *matK*. The mean intraspecific variation for each and combined DNA regions was also low ranging from 0.000108 in *rbcLa* to 0.001251 in the combined dataset, *matK* + *rbcLa*. The mean intraspecific distances in all cases are significantly lower than interspecific distances (Wilcoxon test, $p < 0.0001$). The minimum interspecific genetic distance is greater than the maximum intraspecific genetic distance in *matK* + *rbcLa* dataset (Figure 5A) indicating the existence of a barcode gap in the dataset. The comparison between the lowest interspecific distances (red lines) versus the maximum intraspecific distances (black lines) is shown in Figure 5B. Further, we found 72% (116) of the individuals with barcode gap and 28% (45) without a barcode gap in *matK* + *rbcLa* dataset. We also found 12% (19) of the individuals with barcode gap and 88% (152) without a barcode gap in *matK* dataset. Lastly, we found only 3% (2) of the individuals with barcode gap in *rbcLa* dataset and 97% (168) without a barcode gap.

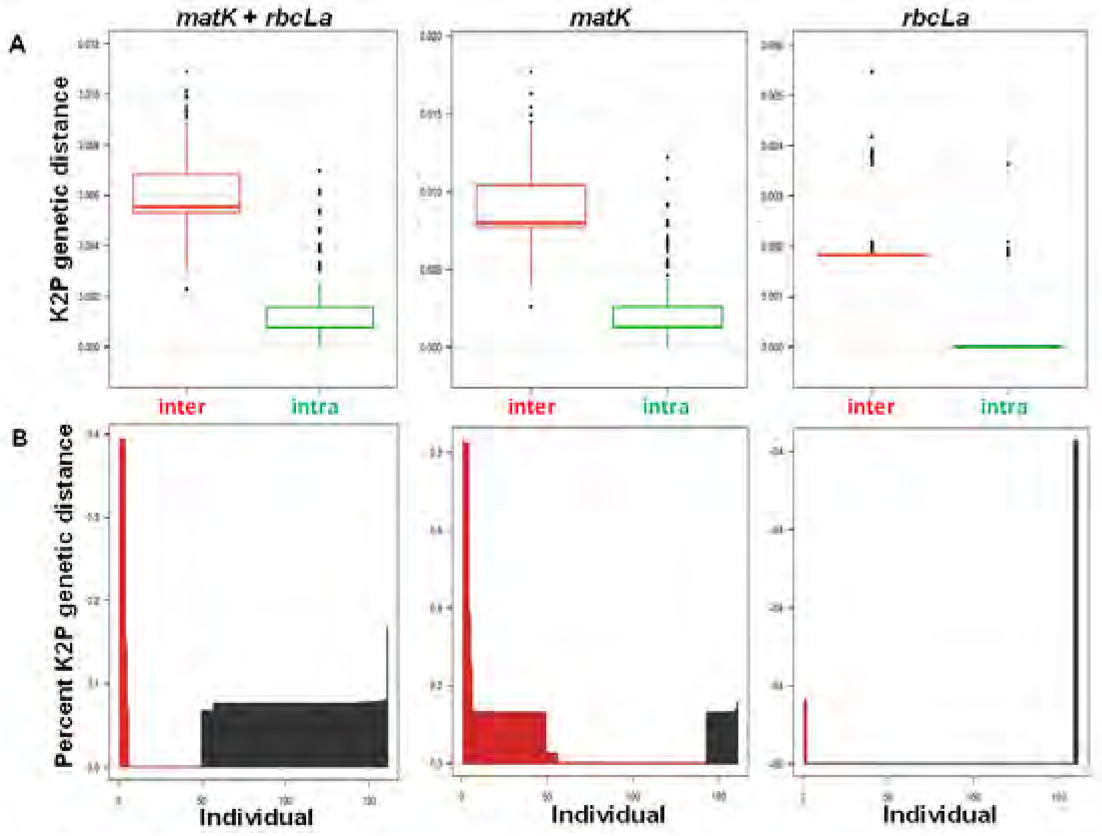


Figure 5. **A** Evaluation of barcode gap in the dataset. Boxplot of the interspecific (inter) and intraspecific (intra) genetic distances for *matK*+*rbcLa*, *matK* and *rbcLa* datasets indicating the existence of a barcode gap i.e. minimum interspecific distance is greater than the maximum intraspecific distance. The bottom and top of the boxes show the first and third quartiles respectively, the median is indicated by the horizontal line, the range of the data by the vertical line and outliers by dots. **B** Line plot of the barcode gap for the 171 Psoraleiod individuals. The black lines indicate where the minimum interspecific distance is greater than the maximum intraspecific distance (an indication of a barcode gap); the red lines show where this pattern is reversed, i.e., the situation where there is no barcoding gap.

Testing the efficacy of DNA barcoding based on discriminatory potential, shows that the calculated thresholds ranged from 0.045 in *matK* to an optimized value of 0.36 for the full dataset (*matK* + *rbcLa*). Using these cut-offs, we found 100% true and correct identification in all the datasets for the near neighbour and best close match analyses respectively in identifying the individuals to their respective genera (*Psoralea* or *Otholobium*). In terms of identifying the individuals at the species level, we found 25% success rate for *matK* compared to 4% in *rbcLa* for the near neighbour method, which did not improve when the two barcodes were combined (*matK* + *rbcLa*) (Table 2). Similarly, for the best close match analysis, *matK* + *rbcLa* and *matK* exhibited 11% correct identification rate as opposed to failure in *rbcLa* (0%) dataset (Table 2).

Table 2. Performance of the DNA barcodes in identification of individuals to species or genera of Psoraleoid legumes evaluated based on discriminatory potential. Values in parenthesis represent identification of individuals to genera. 'True' indicates instances when the near neighbour method finds the closest individual in the dataset and their names are the same or 'False' if different. 'Correct', 'Incorrect', 'Ambiguous' and 'No id' are used in the best close match method, when the name of the closest match is the same, different, more than one species is the closest match, or no species are within the threshold distance, respectively".

DNA barcoding regions	**Number of genetic species (MOTUs)**	**Near neighbour**		**Best Close Match**			
		True (%)	**False (%)**	**Ambiguous (%)**	**Correct (%)**	**Incorrect (%)**	**No ID (%)**
matK + *rbcLa*	37	25 (100)	75 (0)	51 (0)	11 (100)	38 (0)	0
matK	33	25 (100)	75 (0)	53 (0)	11 (100)	36 (0)	0
rbcLa	7	4 (100)	96 (0)	79 (0)	0 (100)	21 (0)	0

Of the 69 morphologically-delimited species included in the analyses, varying discriminatory power in the performance of the DNA markers in grouping specimens into genetic species (MOTUs) was found. *rbcLa* grouped all the specimens into 7 genetic species only (*silhouette coefficient* = 0.98), followed by *matK* (33 genetic species (*silhouette coefficient* = 0.84; Table 3). The combination of *matK* + *rbcLa* grouped specimens into 37 genetic species unit (*silhouette coefficient* = 0.84) which is equivalent to 54% of the morphologically delimited species). We therefore discussed our results based on the combined data of core barcodes (i.e. *matK* + *rbcLa* dataset).

Lastly, we found a weak but significant phylogenetic signal in the affinity of lineages to fire survival and regeneration strategies. This was significant under the Brownian motion model ($D_{resprouters} = 0.797$, $p = 0.003$ and $D_{reseeders} = 0.798$, $p = 0.002$, where $D = 0$ corresponds to a Brownian motion model, and $D = 1$ indicates no phylogenetic signal) (Figure 6). Multiple origin of reseeder habit is observed in both genera, but is predominant in *Psoralea* (Figure 6).

Table 3. Genetic species delimited using the best DNA barcode region (*matK* + *rbcLa*) identified in this study.

No	**Composition of genetic species or molecular operational taxonomic units (MOTUs)**
1	[1] *O acuminatum* Muasya&Stirton3603 [2] *O acuminatum* Muasya&Stirton3850 [3] *O arborescens* Muasya&Stirton3279 [4] *O candicans* Muasya&Stirton3369 [5] *O flexuosum* Muasya&Stirton3276 [6] *O flexuosum* Muasya&Stirton3280.1 [7] *O hirtum* Muasya&Stirton3499 [8] *O obliquum* Muasya&Stirton3198.1 [9] *O parviflorum* Muasya&Stirton3199 [10] *O pustulatum* Muasya&Stirton3286 [11] *O rotundifolium* Muasya&Stirton3173 [12] *O rotundifolium* Muasya&Stirton3929 [13] *O spicatum* Muasya&Stirton3498 [14] *O spicatum* Muasya&Stirton3568 [15] *O spicatum* Muasya&Stirton3906 [16] *O stachyerum* Muasya&Stirton3604 [17] *O stachyerum* Muasya&Stirton3851 [18] *O stachyerum* Muasya&Stirton3872 [19] *O striatum* Muasya&Stirton3318 [20] *O striatum* Muasya&Stirton3339 [21] *O striatum* Muasya&Stirton3351 [22] *O striatum* Muasya&Stirton3363 [23] *O striatum* Muasya&Stirton3561 [24] *O striatum* Muasya&Stirton4106 [25] *O thomii* Muasya&Stirton3187 [26] *O uncinatum* Muasya&Stirton3261 [27] *O uncinatum* Muasya&Stirton3263
2	[1] *O beanianum sp. nov.* Muasya&Stirton3350
3	[1] *O bracteolatum limnophilum sp. nov.* Muasya&Stirton3204
4	[1] *O bracteolatum limnophilum sp. nov.* Stirton13155 [2] *O fruticans* Muasya&Stirton3397 [3] *O fruticans* Muasya&Stirton3480 [4] *O hirtum* Muasya&Stirton3373 [5] *O mundianum* Muasya&Stirton3885 [6] *O parviflorum* Muasya&Stirton3542
5	[1] *O bracteolatum* Muasya&Stirton3164 [2] *O bracteolatum* Muasya&Stirton3879 [3] *O bracteolatum* Muasya&Stirton3963
6	[1] *O candicans* Muasya&Stirton3563 [2] *O schutteae* Muasya&Stirton3575
7	[1] *O candicans* Muasya&Stirton3911
8	[1] *O crewii* Muasya&Stirton3264 [2] *O virgatum* Muasya&Stirton3163 [3] *O virgatum* Muasya&Stirton3191 [4] *O virgatum* Muasya&Stirton3395 [5] *O virgatum* Muasya&Stirton3908
9	[1] *O hamatum* Muasya&Stirton3306 [2] *O hamatum* Muasya&Stirton3310
10	[1] *O hirtum* Muasya&Stirton3190 [2] *O hirtum* Muasya&Stirton3326 [3] *O hirtum* Muasya&Stirton3372 [4] *O hirtum* Muasya&Stirton3878 [5] *O hirtum* Muasya&Stirton3991
11	[1] *O lucens* Muasya&Stirton3570
12	[1] *O prodiens* Muasya&Stirton3845 [2] *O prodiens* Muasya&Stirton3854
13	[1] *O rubicundum* Muasya&Stirton5982
14	[1] *O spicatum* Muasya&Stirton3445
15	[1] *O stachyerum* Muasya&Stirton3791
16	[1] *O stachyerum* Muasya&Stirton3837
17	[1] *O uncinatum* Muasya&Stirton3175
18	[1] *O velutinum* Stirton13106
19	[1] *P aculeata* Muasya&Stirton3170 [2] *P oreopola* Muasya&Stirton4370 [3] *P plauta* Muasya&Stirton3611 [4] *P verrucosa* Muasya&Stirton3269 [5] *P verrucosa* Muasya&Stirton3905
20	[1] *P aculeata* Muasya&Stirton3183 [2] *P aculeata* Muasya&Stirton3405 [23] *P oreophila* Muasya&Stirton3464 [24] *P oreopola* Muasya&Stirton3271

	[3] *P aculeata* Muasya&Stirton3550 [4] *P affinis* Muasya&Stirton3868 [5] *P affinis* Muasya&Stirton3903 2 [6] *P aphylla* Muasya&Stirton3400 [7] *P arida* Muasya&Stirton4098 [8] *P asarina* Muasya&Stirton3907 [9] *P axillaris* Muasya&Stirton3848 [10] *P axillaris* Muasya&Stirton5874 [11] *P cf latifolia* Muasya&Stirton4028 [12] *P elegans* Muasya&Stirton3591 [13] *P fleta* Muasya&Stirton3341 [14] *P forbesii* Muasya&Stirton3578 [15] *P forbesii* Muasya&Stirton3592 [16] *P gigantea* Muasya&Stirton3203 [17] *P imminens* Muasya&Stirton3596 [18] *P ivumba* Muasya&Stirton3374 [19] *P keetii* Muasya&Stirton3599 [20] *P laevigata* Muasya&Stirton3457 [21] *P latifolia* Muasya&Stirton3646 [22] *P odoratissima* Muasya&Stirton3557	[25] *P oreopola* Muasya&Stirton4376 [26] *P pinnata* Muasya&Stirton3403 [27] *P pinnata* Muasya&Stirton3407 [28] *P pinnata* Muasya&Stirton3547 [29] *P pullata* Muasya&Stirton3903 1 [30] *P rhizotoma* Muasya&Stirton3659 [31] *P rigidula* Muasya&Stirton3390 [32] *P sordida* Muasya&Stirton3579 [33] *P sordida* Muasya&Stirton3580 [34] *P speciosa* Muasya&Stirton3458 [35] *P speciosa* Muasya&Stirton3607 [36] *P speciosa* Muasya&Stirton3610 [37] *P suaveolens* Muasya&Stirton4975 [38] *P triflora* Muasya&Stirton3862 [39] *P usitata* Muasya&Stirton3194 [40] *P usitata* Muasya&Stirton3440 [41] *P usitata* Muasya&Stirton3528 [42] *P usitata* Muasya&Stirton3541 [43] *P usitata* Muasya&Stirton4071 [44] *P verrucosa* Muasya&Stirton4371
21	[1] *P alata* Muasya&Stirton3262 [2] *P alata* Muasya&Stirton3398 [3] *P alata* Muasya&Stirton3901	
22	[1] *P alata* Muasya&Stirton3880 [2] *P laxa* Muasya&Stirton3548 [3] *P laxa* Muasya&Stirton3870	
23	[1] *P arborea* Muasya&Stirton3212 [2] *P axillaris* Muasya&Stirton3827 [3] *P axillaris* Muasya&Stirton3834 [4] *P brilliantissima* Muasya&Stirton3621 [5] *P congesta* Muasya&Stirton5462 [6] *P filifolia* Muasya&Stirton4321	[7] *P glaucescens* Muasya&Stirton3289 [8] *P ivumba* Muasya&Stirton3165 [9] *P pinnata* Muasya&Stirton3169 [10] *P pinnata* Muasya&Stirton3172 [11] *P repens* Muasya&Stirton3168 [12] *P repens* Muasya&Stirton3809
24	[1] *P arborea* Muasya&Stirton3248 [2] *P arida* Muasya&Stirton3526 [3] *P asarina* Muasya&Stirton3476 [4] *P asarina* Muasya&Stirton3552	[5] *P odoratissima* Muasya&Stirton3532 [6] *P pinnata* Muasya&Stirton3171 [7] *P usitata* Muasya&Stirton4344 [8] *P usitata* vigilans *sp. nov.* Muasya&Stirton4340
25	[1] *P fleta* Muasya&Stirton3273	
26	[1] *P fleta* Muasya&Stirton3342 [2] *P imbricata* Muasya&Stirton3396 [3] *P imbricata* Muasya&Stirton3399 [4] *P imbricata* Muasya&Stirton3439 [5] *P imbricata* Muasya&Stirton3544	[6] *P imbricata* Muasya&Stirton3904 [7] *P imbricata* Muasya&Stirton4030 [8] *P verrucosa* Muasya&Stirton3353 [9] *P verrucosa* Muasya&Stirton3357
27	[1] *P glaucescens* Muasya&Stirton3312	
28	[1] *P laxa* Muasya&Stirton4325	
29	[1] *P muirii* Muasya&Stirton4181	
30	[1] *P oligophylla* Muasya&Stirton3798	
31	[1] *P oreophila* Muasya&Stirton3463	
32	[1] *P pinnata* Muasya&Stirton3186 [2] *P pinnata* Muasya&Stirton3189	

33	[1] *P pullata* Muasya&Stirton3178
34	[1] *P restioides* Muasya&Stirton3216 [2] *P sparsa* Muasya&Stirton3567 [3] *P speciosa* Muasya&Stirton3456
35	[1] *P usitata ssp. nov.* usitata Muasya&Stirton3414
36	[1] *P usitata ssp.* vigilans sp nov Muasya&Stirton3415
37	[1] *P verrucosa* Muasya&Stirton3270

3.4. Discussion

A key criterion for a standard plant barcode is universality, meaning that the DNA barcode should be easily recovered from all plants, ideally with a single primer pair (CBOL Plant Working Group 2009). Our amplification and sequencing success was higher for *rbcLa* than *matK*, consistent with the results of several other studies that sampled broadly across land plants (e.g., Lahaye *et al.* 2008; CBOL Plant Working Group 2009; Xiang *et al.* 2011a; Saarela *et al.* 2013). Recovery of *rbcLa* was higher (98.1%) than *matK* in this study. This corresponds to the results of other studies on plants in which *rbcLa* recovery ranged from 90–100% (Fazekas *et al.* 2008; Lahaye *et al.* 2008; CBOL Plant Working Group 2009; Jeanson *et al.* 2011; Pang *et al.* 2011; Kuzmina *et al.* 2012; Xiang *et al.* 2011a; Saarela *et al.* 2013).

Several other criteria have also been defined for the identification of the best DNA barcode marker (Hebert *et al.* 2004; Kress and Erickson 2007; Lahaye *et al.* 2008; CBOL Plant Working Group 2009). Firstly, it should exhibit a barcode gap i.e. higher genetic variation between than within species (Meyer and Paulay 2005). Secondly, it must provide a maximal discrimination among species. We measured the efficacy of the core plant DNA barcodes regions (*matK* and *rbcLa*) (CBOL Plant Working Group 2009) to identify African Psoraleoid legumes using the two approaches: 'barcode-gap' and discriminatory potential (Meyer and Paulay 2005). We found that interspecific distance is significantly greater than intraspecific distance. Our mean distances correspond to the results obtained in other plant groups such as Myristicaceae (Newmaster *et al.* 2008), Rosaceae (Pang *et al.* 2011), *Taxus* L. (Taxaceae) (Liu *et al.* 2011) and in regional Canadian Arctic Flora (Saarela *et al.* 2013). The second approach was that of Meier *et al.* (2008); i.e. comparing the smallest interspecific versus the greatest intraspecific distances, instead of comparing the mean distances alone. This approach also reveals the existence of a barcode gap, thus confirming the barcode potential of all the candidates. However, the combination of *matK* and *rbcla* datasets (*matK* + *rbcla*) in all the cases showed greater intraspecific variation than the individual regions alone. This supports the recommendation of the CBOL Plant Working Group (2009) that a combination of the two regions (*matK* and *rbcLa*) is the preferred standard barcode region for plants.

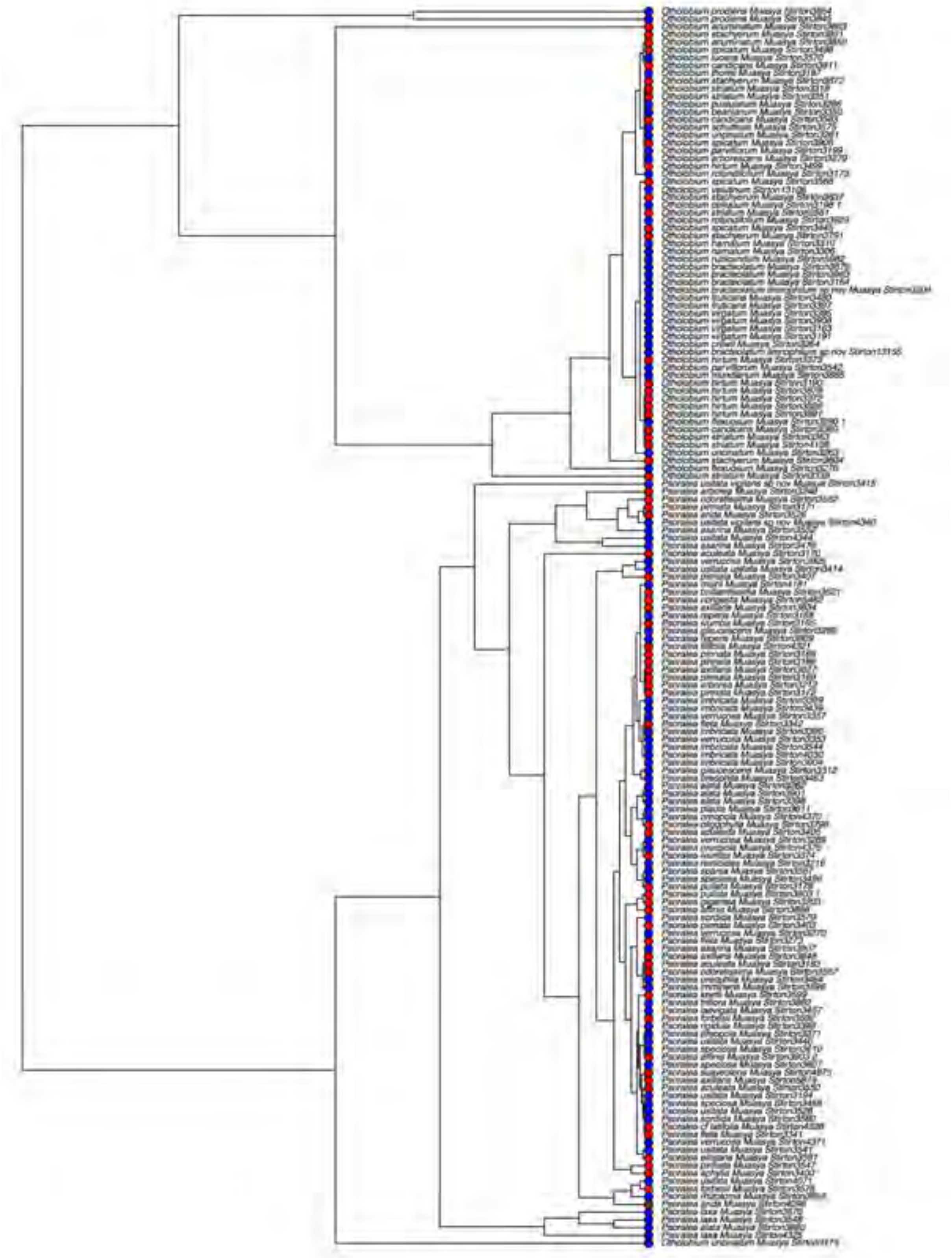


Figure 6. Maximum likelihood tree of Psoraleoid legumes derived from a combination of the core DNA barcodes *matK* and *rbcLa* showing the distribution of fire survival and regeneration strategies as reseeders (red) versus resprouters (blue).

In addition, we found that all the three datasets have a strong discriminatory power (100%) in identifying individuals to their respective genera within the Psoraleoid legumes using the near neighbour and the best close match methods. This supports the utility of DNA barcoding as a means to identify and allocate species between the two genera. Multiple other studies have demonstrated that the core barcode loci routinely provide high discrimination at the genus level, usually greater than 90% (e.g., Kress *et al* 2009, Saarela *et al.* 2013). Accordingly, we found that *rbcLa* and *matK* loci singly distinguish 100% of genera in our dataset. However, their application within species yielded a poor discrimination success i.e. < 50% with more proportion of ambiguity (51% *matK* + *rbcLa* dataset to 79% in *rbcLa* dataset; Table 2). This result is not surprising given that several other plant studies have reported poor utility of the core DNA barcodes at lower taxonomic level especially among closely related species and in taxa characterised by recent rapid radiation and hybridisation. For example, Clement and Donoghue (2012) reported low levels of discrimination and genetic variation among closely related species of *Viburnum*. Similarly, Xiang *et al.* (2011b) reported that *rbcLa* alone was unable to distinguish genera within Juglandaceae, and neither *rbcLa* nor *matK* could discriminate species of *Berberis*, *Ficus*, or *Gossypium* (Piredda *et al.* 2011). In taxa with hybridisation issues e.g. *Quercus*, *matK* and *rbcLa* were unable to distinguish any of the 12 sympatric species examined (Roy *et al.* 2010). The possible causes of the poor discrimination of the species in Psoraleoid legumes observed in this study can be attributed to their recent rapid radiation (Egan and Crandall 2008b) and multiple instances of strong hybridization (Bello *et al.* in prep; see examples in paragraph below) among the species. Given these caveats, it is clear that additional variable loci are necessary to improve the within species discrimination success as recommended by the CBOL Plant Working Group (CBOL 2009).

Another feature of less concern is the low congruence in assigning morphologically-delimited species to genetic species. Several reasons could account for this. Firstly, it could suggest that species are generally not monophyletic (Rieseberg and Brouillet 1994). Secondly, the mismatch could be due to poor performance of the DNA barcodes resulting in over-splitting of taxa. Thirdly, it could be that speciation events do not always match morphological differences; indicating that rapid changes in morphology can occur with minimal evolutionary change (Adams *et al.* 2009). Fourthly, it could indicate that taxa whose multiple accessions are appearing in diverse clades represent cryptic species, where broad morphological concepts on species are masking genetic patterns. This may be true in *Otholobium* where widespread species (*O. candicans, O. striatum* and *O. hirtum*) may be treated too broadly. Hybridization may account for some of the patterns in *Psoralea* as some of the taxa have been observed by me forming hybrids in the field e.g. *P. pinnata* × *P. aculeata, P. sordida* × *P. forbesii*, and *P. intonsa* × *P. oreopola*.

In general, there was a weak but significant phylogenetic signal in fire survival and regeneration strategies of lineages as reseeders or resprouters in Psoraleoid legumes than would be expected by chance. Lineages show significant phylogenetic conservatism in their affinity to fire survival and regeneration strategies with more clustering of resprouters at the tip of the phylogeny than might be expected by chance. Our phylogeny suggests a multiple origin of these traits implying that the species inherited the resprouting trait from their most recent common ancestor. We hypothesise that the scattering of the reseeding trait across the phylogenetic tree was the result of independent evolutionary events (convergent evolution), probably as a response to fire. It could also mean that the character was inherited from a more "basal" ancestor of the group and then "switched off" in some species but not in others again, in response to fire. However, this remains hypothetical at this stage, pending the availability of more data.

Legumes are regarded as one of the most successful families of flowering plants on Earth both from evolutionary and ecological perspectives, owing to their flexible adaptation to different environments (Rundel 1989). This is evident in the way resprouters and reseeders have evolved to survive in their respective micro-habitats in the CFR (Schutte *et al.* 1995), and frequently dominant in after-fire landscapes. Previous comparative studies on these functional groups have focused on aspects of taxonomy and physiology (Schutte *et al.* 1995, Power *et al.* 2011). Here, we provide evidence of a weak but significant phylogenetic signal in fire response trait of lineages as reseeders or resprouters in Psoraleoid legumes than expected by chance. Schutte *et al.* (1995) suggested that there is a substantial difference between resprouters and reseeders, adding that gene flow between resprouting parents and their offspring may occur over time, since the parents are not killed by fire. Seed set does occur in resprouters but is generally very poor and may not occur over a number of fire episodes. The seeds of resprouters are generally larger than those produced copiously by all reseeders (Stirton, pers. obs.).

In contrast, temporal isolation in gene flow might occur in reseeding taxa, as there is less chance of interbreeding between parents and offspring and thus, each new generation may be a cohort of its own. It is not known how much seed remains in the seed bank and it is possible that some seeds may germinate in a later fired episode. It should be borne in mind however that parents and offspring could co-exist if fires are patchy, if fire temperature affects the proportion of the seed bank that can be stimulated to germinate, if fires are too hot, and if the seed bank comprises different genetic cohorts. The consequence of these is that speciation would more readily occur in reseeders, as interbreeding between parents and their progeny is unlikely. Given these caveats our results provide some extrinsic support for the idea that reseeders speciate faster than resprouters as the number of reseeding species in our study outnumbered that of the resprouters. Schutte *et al.* (1995) reported that there is a faster rate of speciation and differentiation within reseeders, than in resprouters, but did not provide any genetic evidence for this. Most reseeding species of legumes in the CFR are short-lived (ca. 8−15 years), with few exceptions; e.g., in *Podalyria calyptrata* and in some forest margin species of *Virgilia* with relatively long life spans (more than 40 years). In the younger genus *Psoralea* there are more reseeders than resprouters whereas in the older genus *Otholobium* there are more resprouters than reseeders and fewer species overall. Among the Psoraleoid legumes, reseeders are frequently observed on wet valleys near mountain streams while resprouters are common in drier habitats, a phenomenon also observed in African Restionaceae which share increased diversification in reseeders (Litsios *et al.* 2013).

3.5. Conclusions

This study showed that DNA barcoding may be useful in identification of SAf Psoraleoid genera (*Otholobium* and *Psoralea*) and in inferring the impacts of recurrent fires on gene flow in resprouting and reseeding taxa in the CFR. In general, we showed that Psoraleoid legumes of the CFR exhibit a barcoding gap with high scores for correct identification of individuals to their respective genera. We found a less considerable match between genetic and morphologically-delimited species which is partially supporting the discriminatory power of DNA barcoding. We also found that lineages in Psoraleeae showed a weak but significant phylogenetic conservatism in their affinity for different fire response trait with more clustering of resprouters in *Psoralea* at the tip of the phylogeny than expected by chance. Our phylogeny suggests a convergent origin of the reseeding trait in African Psoraleoid genera. We conclude that these novel microevolutionary patterns might be acting continuously over time to produce multi-scale regularities of biodiversity especially in a biodiversity hotspot as the CFR.

CHAPTER FOUR: TAXONOMIC REVISION OF THE *PSORALEA PINNATA* SPECIES COMPLEX (PSORALEEAE, FABACEAE) IN SOUTHERN AFRICA

Abstract

Ambiguous species delimitations and nomenclature are a major impediment for users of biodiversity information. This is more pronounced in plant lineages that have experienced recent and rapid diversification and in plant lineages where there is a tendency to lump many species under a general name. The papilionoid legume genus *Psoralea* L. is a young lineage (ca. 4 myo, ± 75 species) which has diversified rapidly within the South African fynbos biome and adjacent habitats. All species bearing 5–19 foliolate leaves tend to be lumped within the polymorphic species *P. pinnata*, resulting in an unwieldy complex of at least 28 taxa of which only eleven species are formally named. In this chapter I sampled representative specimens of the complex, capturing their morphological and geographic diversity, and analysed the data using multivariate methods. The results support the recognition of 25 distinct species, of which 14 are described here as new: *P. azuroides* C.H. Stirt.; *P. brilliantissima* C.H. Stirt., Muasya & A. Bello; *P. elegans* C.H. Stirt.; *P. floccosa* C.H. Stirt., Muasya & A. Bello; *P. imminens*, C.H. Stirt.; *P. intonsa* C.H. Stirt., Muasya & A. Bello; *P. ivumba* C.H. Stirt., Muasya & A. Bello; *P. kougaensis* C.H. Stirt. Muasya & A. Bello; *P. montana* C.H. Stirt., Muasya & A. Bello; *P. muirii* C.H. Stirt. & Muasya; *P. rhizotoma* C.H. Stirt. & Muasya; *P. semota* C.H. Stirt.; *P. sordida* C.H. Stirt. & Muasya; and *P. suaveolens* C.H. Stirt., Muasya & A. Bello. The taxon *P. pinnata* var. *latifolia* Harv. is raised to species rank, as *P. latifolia* (Harv.) C.H. Stirt. comb. nov. The name *P. pinnata sensu stricto* is restricted here to a taxon with 5–9-leaflets and a range extending from the Cape Peninsula to the Kogelberg mountains. Most of the species in the complex have non-overlapping geographical ranges and occur in the fynbos biome in the Western Cape whereas five species occur in the grassland biome of the Eastern Cape, KwaZulu-Natal, and Mpumalanga Provinces of South Africa, Mozambique and Swaziland. A key for the identification of all twenty-five species along with full descriptions illustrations, distribution maps and notes on their conservation status is provided.

Keywords: Morphometrics; new species; nomenclature; taxonomy

4.1. Introduction

The predominantly southern African papilionoid legume genus *Psoralea* L. is a speciose (ca. 75 species), young (ca. 4 myo; see Chapter 2) lineage which has diversified rapidly within the fynbos biome and related habitats. *Psoralea* species are found throughout the extent of the GCFR where they grow almost exclusively in the fynbos vegetation, although some occur in other biomes (Chapter 2). Some species exist in the rock crevices of mountains on the arid edge of the Karoo, but the majority are found in the acidic, nutrient-poor, sandstone-derived soils typical of the fynbos or the richer Table Mountain shale soils. Outside the GCFR, *Psoralea* species are almost entirely restricted to the afromontane heathlands, such as the Drakensberg, and as far north as St Helena off the coast of Angola (Stirton 1981).

Psoralea is generally characterised by the presence of resinous, black, glandular dots on the entire plant, though mostly on the foliage (Bello *et al.* 2015a). The inflorescence can be a head, spike or raceme, axillary or terminal. The flowers can be fascicled or rarely solitary, blue, purple, or rarely yellow and white, and are faintly to strongly sweetly-scented in the majority of the species. Wing petals are longer than keel petals and have sculpturing whereas keel petals white but apically suffused with dark violet-purple on inner apex. The pistil has a small ovary, an attenuated style with a swelling at the poin of flexure. All the species are uni-ovulate with an ovate, indehiscent pod concealed within the calyx. The leaves in *Psoralea* can be digitately or pinnately 3–19-foliolate, 1-foliolate or sometimes reduced to scales as in the *Psoralea aphylla* complex (Dludlu *et al.* 2015). The stipules embrace the branches and are scarcely adnate to the petiole. *Psoralea* is also characterised by the presence of a unique, cup-shaped structure at the base of each flower pedicel, formed by the fusion and intercalary growth of three to four successive bracts, termed as the cupulum (Tucker & Stirton 1997).

Psoralea pinnata L. is the type species of *Psoralea*. It is an erect, pinnately-leaved shrub or tree up to 5 m tall bearing either blue, mauve, purple, lilac or white flowers that bloom from October to April (Harvey 1862; Forbes 1930). New plant explorations over the last century have added many new variants to the known species, and it has become unwieldy and a dumping ground for most pinnate-leaved *Psoralea* species. The *P. pinnata* complex (Fountain bush; fonteinbos, bloukeur, and penwortel (Afr.); umHlonishwa (Zulu)) occurs mostly in seepage areas in fynbos and extends into similar habitats in the Eastern Cape through KwaZulu-Natal and Mpumalanga to Swaziland and Mozambique (Figure 1). The species, as currently circumscribed, encompasses great morphological and molecular variation and forms a complex of 11 described species and ca. 17 undescribed species known only by their informal names in herbaria and in publications describing the plants of the region (e.g., Manning and Goldblatt 2012). Species delimitations and nomenclature in this complex are therefore unclear. As a number of these taxa are included in the Red Data list of South Africa (Raimondo *et al.* 2009), the absence of clear species delimitation and valid names is an impediment towards their conservation.

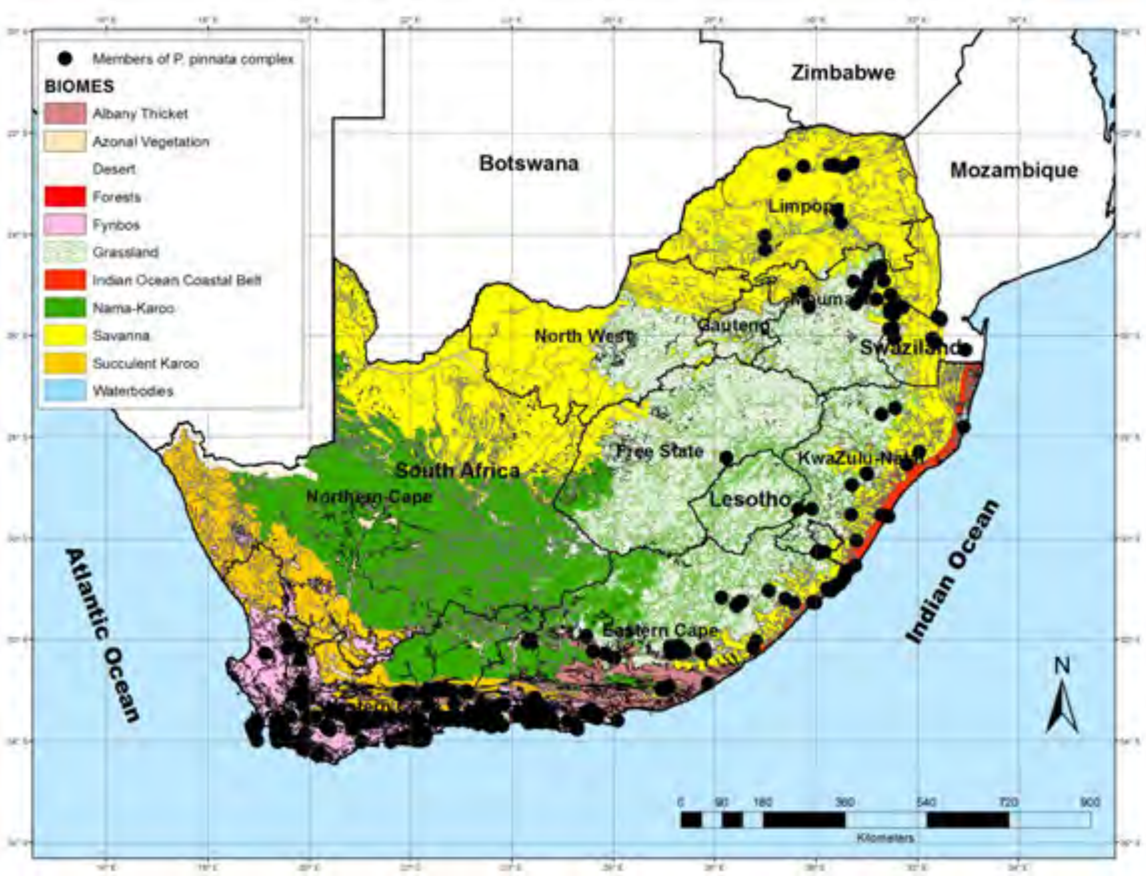


Figure 1. Distribution range of members of *P. pinnata* species complex.

Here, the concept of *P. pinnata* as described by Linnaeus (1753) is adopted. The important features of Linnaeus's concept of *P. pinnata* include: arborescent or shrubby, pubescent or glabrous, compactly branched; leaves 3−5 pairs, imparipinnate, linear or linear lanceolate, acute, very narrow; peduncle axillary, long or short, bracteolate beyond the middle; calyx very variable in incision and pubescence. A number of species consistent with Linnaeus's description of *P. pinnata* were described a century later: *P. affinis* Eckl. & Zeyh., *P. arborea* Sims, *P. glabra* E.Mey., *P. laevigata* Eckl. & Zeyh., *P. latifolia* Torr., *P. odoratissima* Jacq., and *P. speciosa* Eckl. & Zeyh. However, Harvey's (1862) revision of the genus synonymised *P. affinis*, *P. arborea*, *P. glabra*, and *P. latifolia* as varieties of a polymorphic *P. pinnata*, stating he "cannot but regard them as local forms of one variable species". He upheld *P. odoratissima*, believing it to be a garden variety, but did not mention *P. laevigata*.

In her subsequent revision of the genus, Forbes (1930) recognised *P. pinnata* and *P. affinis* as separate species, but could not convincingly separate them. She described *P. pinnata* as a tall, much-branched woody shrub, up to 3 m high with striate, virgate stems, and *P. affinis* as a tall, virgate shrub, up to 1.8 m tall. None of the other earlier published species within the complex were outlined by Forbes (1930). However, she cited some of the specimens belonging to the varieties of *P. pinnata* as depicted by Harvey (1862) under her *P. pinnata*. This shows that despite the recognition of *P. affinis* as a unique species, her concept of *P. pinnata* is similar to Harvey's (1862). Forbes also noted that *P. pinnata* had a wider distribution than any other endemic *Psoralea* and that it flowered throughout the year. She also recognised *P. pinnata* and *P. affinis* to be broadly sympatric. As the majority of the morphological characters of the two species she used greatly overlapped, it is difficult to distinguish between them. Since her revision, no other revision has been done besides the generic changes made by Stirton (1981), where *Psoralea sensu lato* was broken into six genera (*Psoralea*, *Cullen*, *Otholobium*, *Bituminaria*, *Hallia* and *Orbexilum*). Stirton and Schutte (2000, 2012) recognised a number of the validly published taxa (*P. arborea*, *P. glabra*, *P. laevigata*, *P. latifolia*, and *P. speciosa*) which had not been recognised by Forbes (1930). It is important to note that neither Harvey nor Forbes saw the majority of the species of *Psoralea* in the wild and that the herbarium material available to them excluded many of the new taxa reported here. Subsequent studies of the *P. pinnata* complex included the publication of several new species: *P. abbottii* C.H.Stirt. (Stirton 1995); *P. margaretiflora* C.H.Stirt. & V.R.Clark (Stirton *et al.* 2011); and *P. vanberkelae* C.H.Stirt., A.Bello & Muasya (Bello *et al.* 2015a).

This study adds to recent studies focusing on the taxonomy (e.g. Dludlu *et al.* 2015) and molecular identification (e.g. Bello *et al* 2015b) in *Psoralea*. In this study we evaluate whether the diversity that exists within the *P. pinnata* complex is a depiction of several local forms of a single variable species, as proposed by Harvey (1862), or whether it comprises several distinct species. Using multivariate analyses of quantitative and qualitative morphological characters, we test whether the putative species can be separated based on overall similarity. For taxa that are recognised as distinct species in this study, we provide an identification key, and revise their taxonomy and conservation status.

4.2. Materials and methods

All the data and observations were recorded from specimens collected from their natural populations as well as specimens loaned from various herbaria (for a full list, see further on).

4.2.1. Sampling and morphometric measurements

In addition to over 35 years' field experience and collections made by the second author of this paper (here referred to as chapter) C.H. Stirton, extensive field observations and collections were carried out by all authors from 2007 to date across the entire range of the distribution of putative members of the *P. pinnata* complex noting their vegetative and floral morphology and phenology. The field collections were then integrated with an examination of specimens in herbaria known to have the largest and most important collections of southern African flora: B, BLFU, BM, BOL, E, GRA, J, JRAU, K, KEI, L, LE, LINN, M, MO, NBG, NH, NU, NY, OXF, P, PRA, PRC, PRE, PRU, S, SAAS, SAM, SRGH, STE, TCD, Z, ZT and US (acronyms following Thiers 2015). Voucher materials from this study, were prepared and deposited in the Bolus herbarium (BOL), University of Cape Town and duplicates will be distributed to other herbaria where available.

After an extensive *a priori* analysis of 37 quantitative morphological characters, 20 characters that showed some degree of significance in delimiting all the specimens were selected. For qualitative characters, we considered only those characters that showed distinction between the species considered in this study. Finally, a total of 28 (20 quantitative and 8 qualitative) characters were evaluated. Flowers were soaked in water for 5 minutes and then carefully dissected prior to measurements. Both reproductive and vegetative characters were measured using callipers. Where necessary, a dissecting microscope was used for the measurements. Some of the leaf and floral characters measured are illustrated in Figure 2 and a list of all the 28 characters is presented in Table 1. To eliminate the effects of missing data on the statistical analyses, only specimens that contained all the information needed were selected (i.e. specimens that had both vegetative and mature floral parts). For each taxon, measurements were taken for up to 15 specimens where possible. Five measurements were made on each specimen and the mean of the five was recorded for each character. With rare taxa, a minimum of 5 specimens were measured. For the analyses ca. 259 specimens were suitable. *A priori* groups (i.e. groups of specimens formed on the basis of similarities and/or dissimilarities of leaf shapes, numbers, inflorescence structure, and floral features without regard to labels on herbarium sheets) were formed, and the existing published species names and names outlined in Manning and Goldblatt (2012) were used to putatively identify the groups of specimens.

4.2.2. Data analyses

Multivariate analyses were carried out sequentially by cluster analysis (CA) and principal coordinates analysis (PCoA), using the PAST 3 program (version 3.05, folk.uio.no/ohammer/past). All 28 morphological characters, for all 259 specimens, were used for the analyses and each individual specimen was considered as an operational taxonomic unit (OTU).

In all the analyses, the data were first log10-transformed for standardisation of the data matrix. CA was used to cluster the specimens, as it is better in representing distances among similar specimens (Sneath and Sokal, 1973; Sebola and Balkwill, 2013). To visualise the level of morphological similarity/dissimilarity, simple Euclidean distance was used between and within the *a priori* groups because it does not change under orthogonal transformations of the variables. This was performed by calculating the similarity matrix between OTUs using the average taxonomic distance coefficient from the standardised matrix, which clustered the OTUs by using the unpaired group method of arithmetic averages (UPGMA).

Principal coordinate analysis (PCoA) was preferred over principal component analysis (PCA) since the data set contained a mixture of quantitative and qualitative characters (Small & Brookes 1990; Small *et al.* 1999; Sebola & Balkwill 2013). PCoA is better at representing distances among groups of specimens (interspecific) (Sneath & Sokal 1973; Sebola & Balkwill 2013). PCA is only applicable to quantitative continuous characters and is not suitable for discrete qualitative characters (Sneath and Sokal 1973; Schilling & Heiser 1976; Kent & Coker 1992). PCoA is also used to test whether groups obtained by cluster analysis could be supported, as PCA is known to impose a hierarchical structure on any data (Thorpe 1983) and it is not uncommon that clusters may not be recoverable in ordination analysis (Chandler and Crisp 1998). In this study, PCoA was applied to the full dataset, since the method does not have the same constraints on the dataset nor the same assumptions as PCA (Austin 1985) and also as a means to evaluate the discriminating ability of the characters at an interspecific level (Sokal and Rohlf 1997).

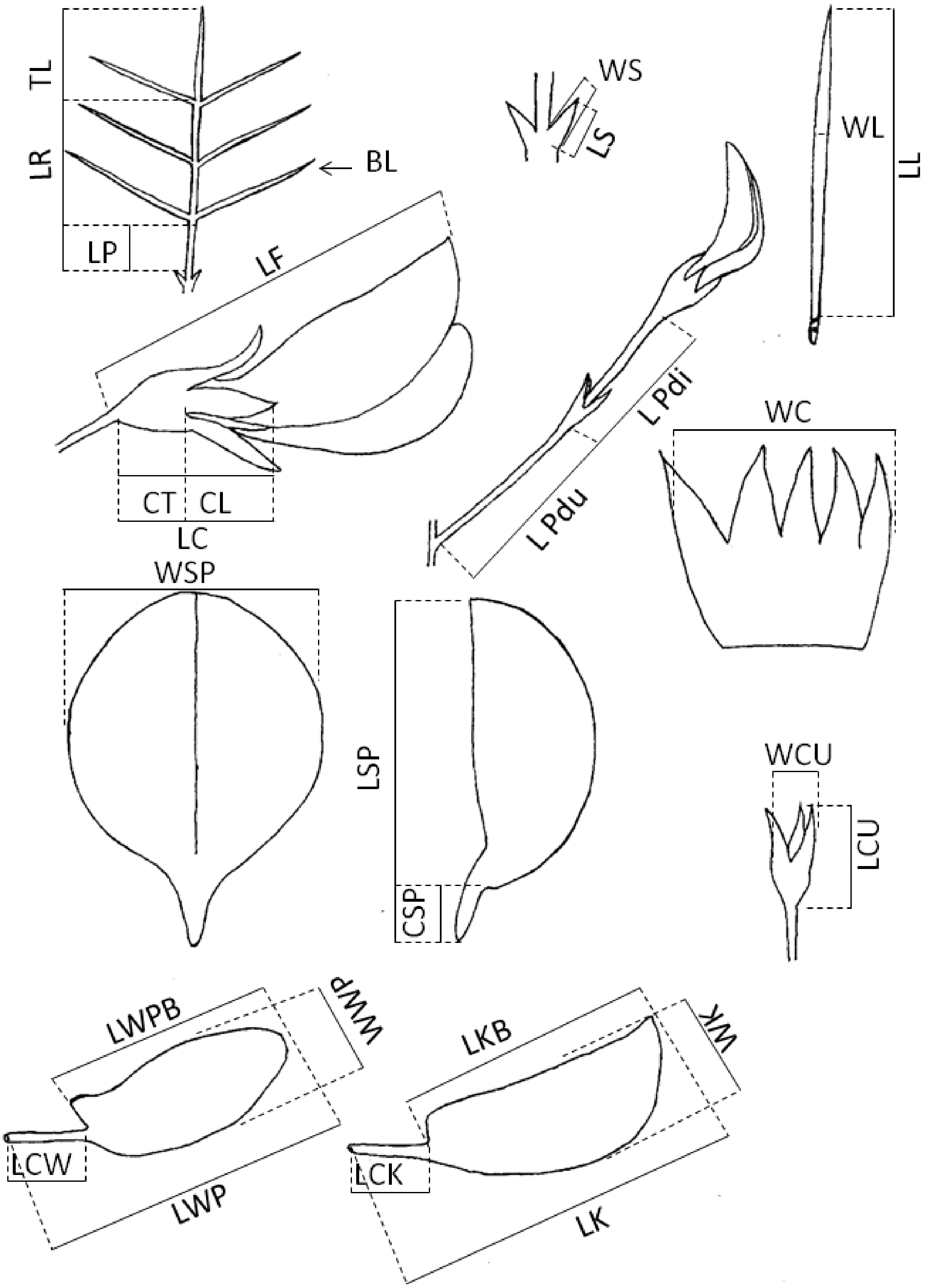


Figure 2. Illustrations of some of the vegetative and floral characters measured (TL=Terminal leaflet, LR=Length of rachis, LP= Length of petiole, BL= Basal leaflet, LS= Length of stipule, WS= Width of stipule, LL= Length of leaflet, WL= Width of leaflet, LF= Length of flower, CT=Calyx tube, CL= Calyx lobe, LC= Length of calyx, LPdu= Length of peduncle, LPdi= Length of pedicel, WC= Width of calyx, WSP= Width of standard petal, LSP= Length of standard petal, CSP= Claw of standard petal, LWP= Length of wing petal, WWP= Width of wing petal, LCW= Length of claw of wing petal, LWPB= Length of wing petal blade, LK= Length of keel, WK= Width of keel, LKB= Length of keel blade, LCK= Length of claw of keel, LCU= Length of cupulum, WCU= Width of cupulum).

In addition, we used univariate one-way ANOVA statistics on the quantitative data set using Statistica version 12 (Statsoft, Tulsa, Oklahoma, USA) to test the equality of means of individual parameters among the species. The significantly different means at $p<0.05$ were separated by Tukey's test (Sokal and Rohlf 1997; Zar 1999). The arithmetic mean ($\bar{x}$), standard deviation (SD) and coefficient of variation (CV) were calculated and analysed for each taxon. The range of variation was observed by plotting whisker-box plots of the data scored for each quantitative character.

Table 1. Morphometric characters used in the phenetic analyses of *P. pinnata* species complex (mm= millimetres)

S/No	Character	States
1	Number of leaflets (Nl)	Qualitative (5, 7, 9, 11)
2	Length of petiole (Lp)	Quantitative (mm)
3	Length of rachis (Lr)	mm
4	Length of rachis basal internode (Lrbi)	mm
5	Length of basal leaflets (Lbl)	mm
6	Width of basal leaflets (Wbl)	mm
7	Length of terminal lateral leaflet (Ltll)	mm
8	Length of terminal leaflet (Ltl)	mm
9	Width of terminal leaflet (Wtl)	mm
10	Number of flower per axil (Nfpa)	Quantitative
11	Length of peduncle (Lpe)	mm
12	Length of calyx (Lc)	mm
13	Width of calyx tube (Wct)	mm
14	Length of standard petal (Lsp)	mm
15	Width of standard petal (Wsp)	mm
16	Length of wing petal (Lwp)	mm
17	Width of wing petal (Wwp)	mm
18	Length of keel petal (Lkp)	mm
19	Length of cupulum (Lcl)	mm
20	Width of cupulum (Wcl)	mm
21	Habit (H)	Qualitative (0= Small shrub (≤ 2 m), 1=Medium shrub (> 2 –4 m), 2= Large shrubs to small trees (> 4 m))
22	Life form (Lf)	Qualitative (0= reseeder, 1= resprouter)
23	Shoot texture (St)	0=smooth, 1= intermediate, 2= rough
24	Flower colour (Fc)	0= white, yellow or cream, 1= blue, pale blue, azure blue, 2= pink, 3= mauve, 4= purple, deep purple
24	Flower congestion (Fco)	0= lax (one flower per axil), 1= congested (more than one flower per axil)
26	Inflorescence type (It)	0= indeterminate, 1= determinate
27	Inflorescence position (Ip)	0= pseudo-spicate, 1= pseudo-capitate
28	Calyx indumenta (Ci)	0= glabrous, 1= spersly pubescent, 2= pubescent

4.3. Results

4.3.1. Clustering

The CA of the entire data set separated 25 clusters (at Euclidian distance of 1.5; Figure 3: see details on names in Appendix C). The cophenetic correlation coefficient value of r = 0.85 obtained in the analysis indicates a very good fit between the triangular distance matrix and the phenogram (Sneath & Sokal 1973; Rohlf 1998). The groups were recognised as distinct taxa at different taxonomic hierarchies if all their OTUs did not mix between clusters, also considering the level of phenetic distance at which OTUs cluster together. All the *a priori* groups formed distinct clusters with the exception of *P. azuroides* and *P. bella* whose specimens intermixed and formed one large cluster (cluster number 7). A discussion on the characters and character states that played a role in the formation of distinct clusters in the clustering and the ordination is provided in the taxonomic section.

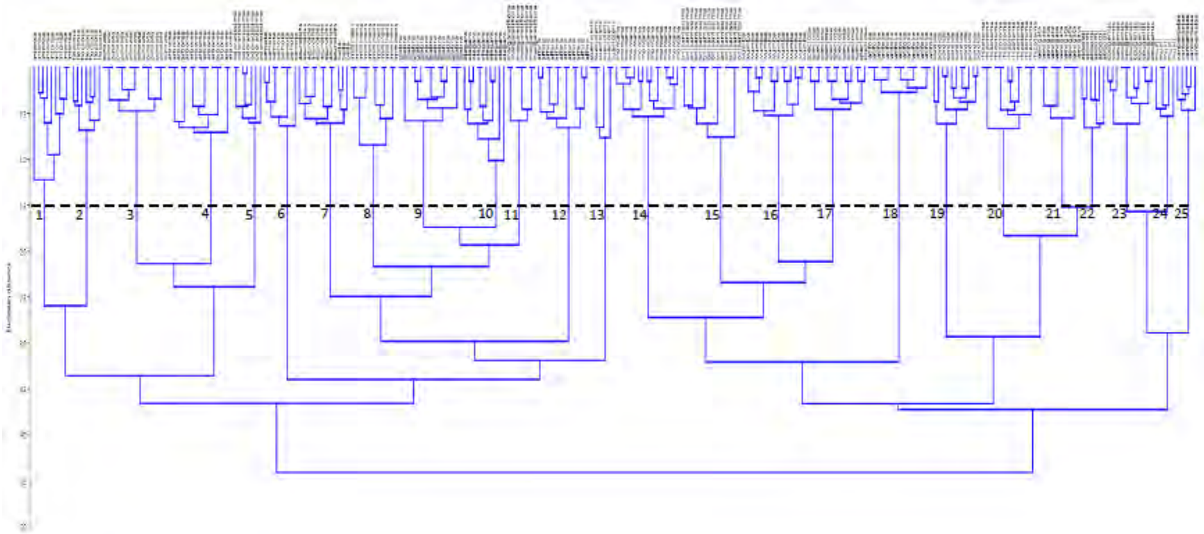
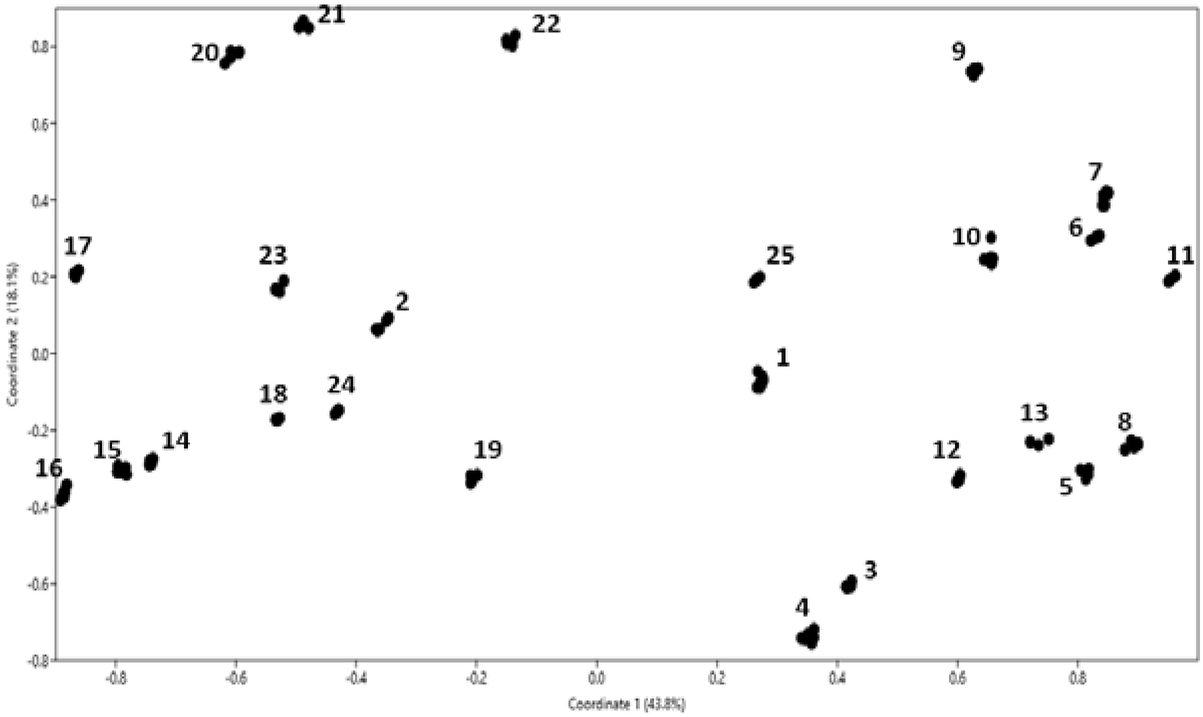


Figure 3. UPGMA phenogram resulting from cluster analysis of the full data matrix of *P. pinnata* complex. Co-phenetic correlation (r) = 0.854.Vertical dotted lines indicate phenon line. The numbers (1−25) indicate distinct clusters recognisable at species level.

4.3.2. Ordination

The ordination analysis based on the PCoA of the full data set separated the 259 specimens of *P. pinnata* complex into 25 groups corresponding largely to those obtained in the cluster analysis (Figure 4). Principal coordinate 1 (PCo1) accounted for 43.83% of the variation, principal coordinate 2 (PCo2) accounted for 18.12% of the variation and principal coordinate 3 (PCo3, not shown) accounted for 14.01% of the variation. The loadings of the first and second PCos are presented in Table 2.The characters most strongly correlated with the first PCo axis with high values (r > 0.50) were: flower colour (r = 0.96), shoot texture (r = 0.59) and inflorescence type (r = 0.53) while those correlated with the second PCo axis were habit (r = 0.82), life form (r = 0.78), flower congestion (r = 0.61), length of apical lateral leaflets (r = 0.59), inflorescence position (r = 0.56), length of basal lateral leaflets (r = 0.52) and calyx indumentum (r = 0.52).



Legend: 1= *P. intonsa*, 2= *P. floccosa*, 3= *P. pinnata*, 4= *P. arborea*, 5= *P. odoratissima*, 6= *P. semota*, 7= *P. azuroides*, 8= *P. imminens*, 9= *P. glabra*, 10= *P. elegans*, 11= *P. margaretiflora*, 12= *P. affinis*, 13= *P suaveolens*, 14= *P. montana*, 15= *P. brilliantissima*, 16= *P. ivumba*, 17= *P. speciosa*, 18= *P. latifolia*, 19= *P. sordida*, 20= *P. rhizotoma*, 21= *P. laevigata*, 22= *P. abbottii*, 23= *P. kougaensis*, 24= *P. muirii*, 25= *P. vanberkelae*.

Figure 4. Plot of the first two principal coordinates (PCo) obtained from analysis of the full morphological data set of specimens of the *P. pinnata* complex. The first and second PCo axes explain 43.8% and 18.1% of the total variation between all the taxa, respectively. The numbers (1−25) indicate distinct clusters recognisable at species level.

Two major clusters were recovered in the ordination plot, corresponding largely to those obtained by CA. The first major cluster (Figure 4) from the left hand side of the ordination plot consists of 12 individual groups while the second on the right consists of 13 individual groups. All the clusters (Figure 3, numbered as in Figure 4) were clearly separated either along the first coordinate axis or the second or both. The results of this study support a narrower circumscription of *P. pinnata sensu stricto* as a distinct taxon that is not intermixed with any other form of pinnately leaved *Psoralea*, as well as the recognition of 25 distinct species among the pinnate *Psoralea*.

The majority of the *a priori* groups identified in CA (Euclidian distance 1.5, Figure 3) were supported by the ordination analysis. Although an *a priori* analysis of the specimens used in this study showed some level of distinction in morphology between the specimens of *P. azuroides* and *P. bella*, the current morphometric analysis failed to separate the two in both the cluster and the ordination results (Figure 3 and 4). The results show that the two taxa have overlapping morphological characters. These include growth habit, life form, length and width of leaflets, rachis, and petiole (Figure 5). Other characters such as number of leaflets, length and width of standard and wing petals, and size of the cupulum also overlapped. Adopting Stace's (1989) second criteria for identifying species, that "there should be gaps between the spectra of variation exhibited by related species, and if there are no such gaps then there is a case for amalgamating the taxa as a single species", the results of this study therefore support the inclusion of all the specimens of Cluster 7 under a single species and the informal name *P. azuroides*, which has been used for a longer time, is formally recognised here as a new species. Recent fieldwork led to the discovery that *P. azuroides* and *P. montana* can hybridize and that the progeny approach of *P. bella*. On the other hand, *P. latifolia* (cluster 18) is clearly separated from *P. pinnata sensu stricto* (cluster 3) in both analyses. This level of dissimilarity indicates that the specimens of *P. latifolia* should not be mixed with any other specimens in the complex and should be considered as a distinct taxon that needs to be recognised as a distinct species.

4.3.3 Univariate

Results obtained from the one-way ANOVA showed that the taxa differed significantly ($p<0.05$) from each other in the mean values of some of the characters, while some did not differ. Characters that showed significant differences included, width of basal lateral leaflet, width of terminal leaflets, length of apical lateral leaflet, length of terminal leaflet, length of petiole, width of the cupulum lobe, length of cupulum lobe, length of pedicel, and width of calyx tube. The values of the mean ± SD of these characters were plotted in the whisker and box plots in Figure 5 (A−I). Characters showing an overlapping pattern among the taxa included, the number of leaflets, length of rachis, length of rachis basal internode, length of basal lateral leaflet, number of flowers per axil, length of calyx (carinal lobe), length and width of standard petals, length and width of wing petal, and length of keel petal.

Table 2. Loadings of the first and second coordinates in the principal coordinate analysis

Axis	**Eigenvalue**	**Percent**
1	773.89	43.829
2	319.96	18.121
3	246.8	13.978
4	114.85	6.5047
5	79.111	4.4804
6	57.471	3.2549
7	46.096	2.6106
8	27.335	1.5481
9	24.692	1.3984
10	20.651	1.1696
11	11.645	0.65952
12	10.426	0.59046
13	6.6463	0.37641
14	5.1933	0.29412
15	4.0079	0.22698
16	3.2825	0.1859
17	2.7632	0.15649
18	2.4205	0.13708
19	1.9274	0.10916
20	1.3469	0.076283
21	1.0026	0.056784
22	0.88355	0.05004
23	0.78475	0.044444
24	0.75113	0.04254
25	0.63744	0.036101
26	0.44386	0.025138
27	0.40086	0.022703
28	0.27507	0.015579

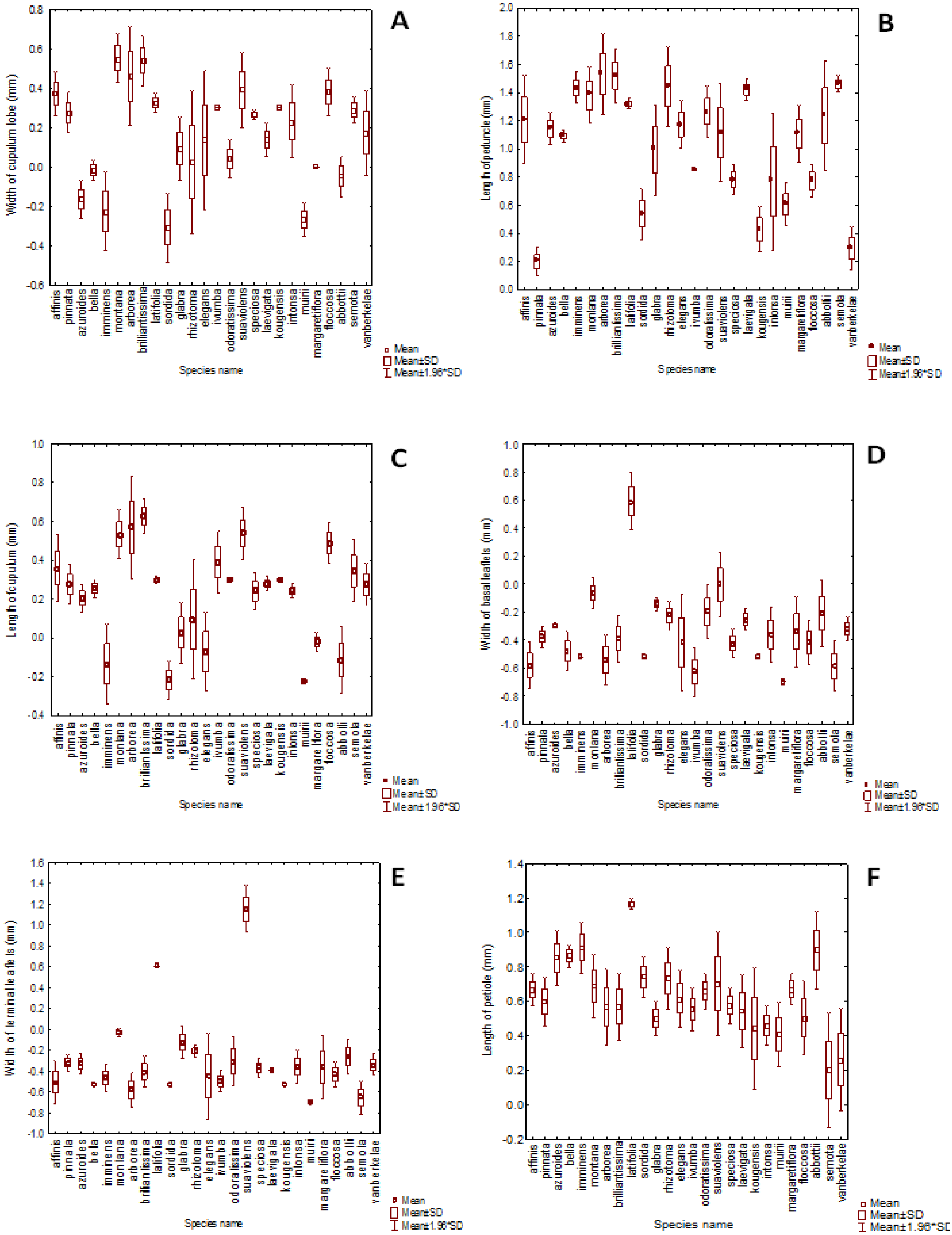


Figure 5. (A−I): Box and whisker plots of nine quantitative characters that showed significant variations (p<0.05) amongst the 25 species of *P. pinnata* complex.

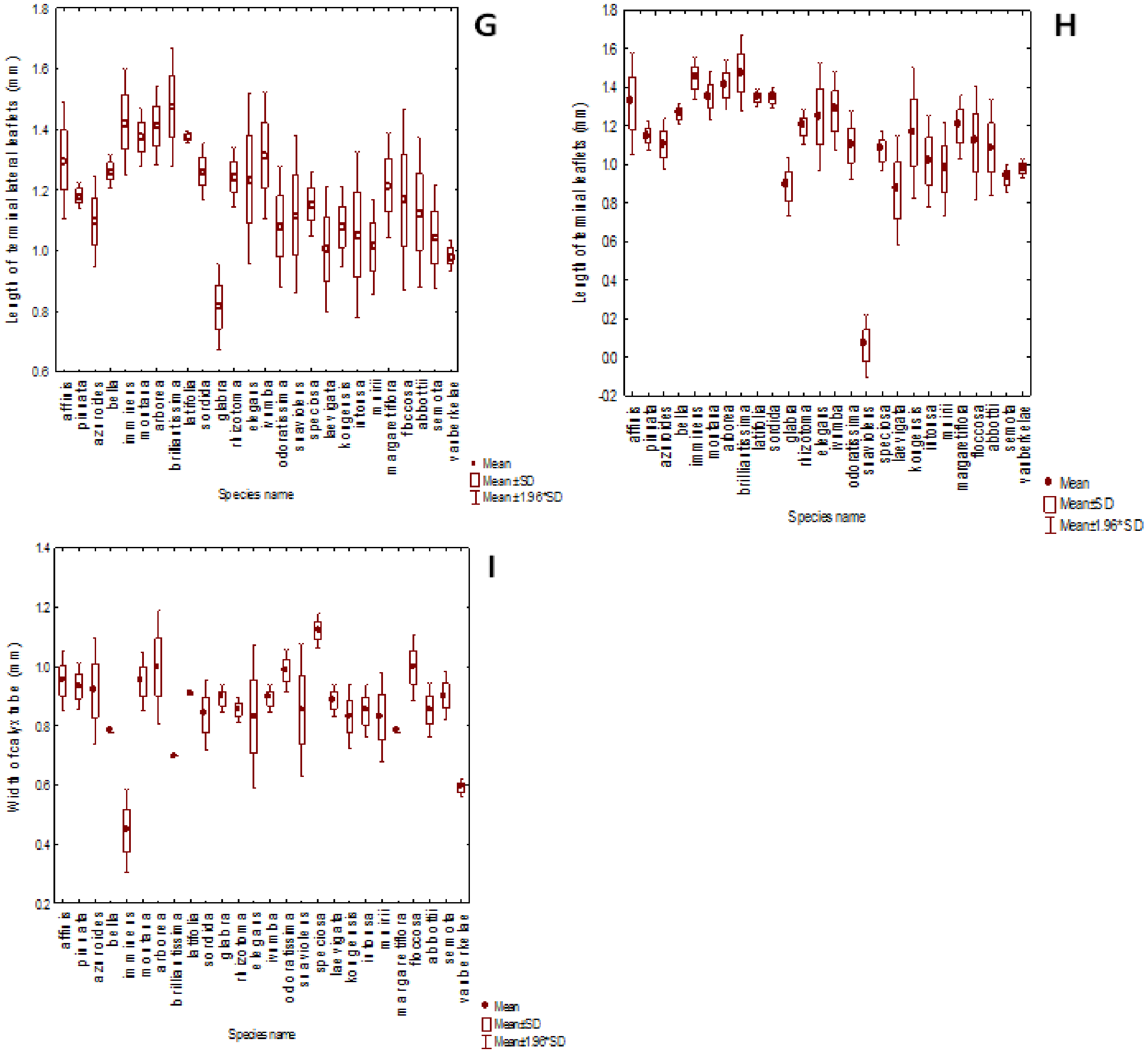


Figure 5. Continued.

4.4. Discussion

The objective of this study was to determine whether the diversity that exists within the *P. pinnata* complex is a depiction of several local forms of a single variable species as proposed by Harvey (1862) or whether it comprises distinct taxa that need to be recognised as separate species. We defined these species according to Sneath's (1976) concept of "distinct phenetic clusters" and these were evaluated according to the pattern of morphological variation. We further employed Stace's (1989) four main criteria for recognizing a species, namely: (1) individuals should bear a close resemblance to one another such that they are always readily recognisable as members of that group; (2) that there should be gaps between the spectra of variation exhibited by related species; (3) if there are no such gaps then there is a case for amalgamating the taxa as a single species; and (4) that each species occupies a definable geographical area and is demonstrably suited to the environmental conditions which it encounters. In this complex, some taxa, e.g. *P. glabra* and *P. latifolia*, are widespread in their distribution yet show morphological variation with a consistent suite of characters. Such taxa meet Sneath's (1976) concept of a distinct phenetic cluster that is not limited to geographical or ecological conditions and so have been considered as distinct species. It has been reported that the morphological variation between organisms is often considered to infer underlying genetic differences (Stace 1989). The uses of multivariate techniques for species identifications and delimitations have been used frequently (e.g. Small & Brookes 1990; Small *et al.* 1999; Sharma & Shiva 2011; Sebola & Balkwill 2013) to provide faster, more reliable information on taxonomic delimitations.

The results of this study do not support Harvey's (1862) hypothesis of a single variable species, but favours the alternative hypothesis that *P. pinnata* comprises several distinct taxa that need to be recognised as separate species. The results show that the *P. pinnata* complex represents an assemblage of 25 distinct species (Figure 3 and 4). Of these, only 11 species (*P. abbottii*, *P. affinis*, *P. arborea*, *P. glabra*, *P. laevigata*, *P. latifolia*, *P. margaretiflora*, *P. odoratissima*, *P. pinnata*, *P. speciosa* and *P. vanberkelae*) have been formally described. This study provides descriptions of 14 species new to science, including taxa that are already known by manuscript names in literature and herbaria.

4.5. Conclusion

Based on the data presented here, 25 species are recognised within the *P. pinnata* complex. Harvey's (1862) taxonomic interpretation that *P. pinnata* is a single variable species is not supported by the results of this study. A narrower circumscription of *P. pinnata* is adopted with a formal recognition of 24 additional taxa at the specific rank. Additionally, we established that the use of few diagnostic characters to differentiate between taxa within *P. pinnata sensu lato* is limiting. The most reliable characters for delimiting these taxa were obtained by a combination of vegetative and inflorescence features. The circumscription of the 25 species within *P. pinnata sensu lato* allows for easier identification and conservation within the genus *Psoralea*.

4.6 Taxonomic treatment

Generally, members of the *Psoralea pinnata* complex are characterised by the presence of resinous hyaline sunken or raised secretory cavities (drying black) covering the entire plant, but mostly on the pinnate foliage. Flowers are fascicled or rarely solitary, strongly to faintly scented to unscented. Inflorescence capitate, spicate or racemeose, axillary or terminal. All taxa have the presence of a unique, cup-shaped structure at the base of each flower pedicel, formed by the fusion and intercalary growth of three to four successive bracts, termed the cupulum. The calyx is very variable in incision and pubescence but all have 5 lobes. All the species are uni-ovulate with an ovate, indehiscent pod concealed within the calyx. Fruits are papery, reticulate, finely pubescent, stipitate. Seed reniform, black or brown.

4.6.1. Key to taxa within the Psoralea pinnata species complex

1a. Small to large shrubs, up to 3 m tall..2

2a. Suffrutices, without an erect woody stem, less than 2 m tall; branching at or near the base ....……..3

2b. Small to large shrubs, with an erect woody stem, 2−3 m tall; branching above the base...8

3a. Variable number of leaflets produced at different stages of growth; flowers reddish purple ....**1. *P. rhizotoma***

3b. Constant number of leaflets produced at all stages of growth; flowers blue, mauve or purple...4

4a. Peduncle shorter than the subtending leaf; leaves only 5-foliolate 5

4b. Peduncle equal in length or longer than the subtending leaf; leaves 5−15 foliolate........6

5a. Old seasonal shoots bare, rough; leaves tightly packed, fleshy; leaflets with raised glands; stipule acuminate, persistent, becoming prominent and woody when leaves are shed....... ....**2. *P. vanberkelae***

5b. Old seasonal shoots leafy, smooth; leaves spreading, not fleshy; leaflets without raised glands; stipules subulate, caducous, not prominent ....**3. *P. azuroides***

6a. Leaves 11−15-foliolate, leaflets sparsely pubescent; peduncle, cupulum and calyx pubescent with patent black hairs; flowers deep blue, front of standard petal with white striations ....….**4. *P. semota***

6b. Leaves 5−7-foliolate, leaflets glabrous; peduncle, cupulum and calyx glabrous; flowers deep purple or mauve, front of standard petal without white striations…........................7

7a. Procumbent shrub with rigid branches; surface of mature leaflets shiny; leaflets clasping the shoot; flowers deep purple; endemic to Western Cape Province….........**5. *P. laevigata***

7b. Decumbent or procumbent shrub with flexuous branches; surface of mature leaflets dull; leaflets spreading; flowers mauve; endemic to NE Eastern Cape and South KwaZulu-Natal...... ....**6. *P. abbottii***

8a. Resprouter (coppicing after burns)..9

9a. Leaves 13−15(17)-foliolate; peduncle equal or longer than of the subtending leaf, 30−50 mm long; plants strongly scented…………………………….................**7. *P. suaveolens***

9b. Leaves (3)5−7(9)-foliolate; peduncle shorter than the subtending leaf, less than 30 mm long; plants weakly scented or unscented…… 10

10a. Old seasonal shoots bare, rough; leaves tightly packed; leaflets with raised glands; stipules persistent, becoming woody..11

10b. Old seasonal shoots leafy, smooth; leaves spreading; leaflets without raised glands; stipules caducous..........………………………..12

11a. Plants sparsely branched or stems unbranched; up to 3 m tall, bearing clusters of seasonal flowering shoots; stipules 3–5×3 mm; calyx equal or more than 7 mm long...**8. *P. kougaensis***

11b. Plants with many rigid branches on stems; not more than 2 m tall, bearing disperse seasonal flowering shoots; stipules 0.7–2×1−1.5 mm; calyx up to 7 mm long…**9. *P. muirii***

12a. Leaves with 3−5(7) leaflets; three upper most leaflets digitately inserted; endemic to Swartberg mountains, Western Cape......... .... **10. *P. sordida***

12b. Leaves with 5−9 leaflets; three upper most leaflets pinnately inserted; endemic to Eastern Cape, southern coast and midlands of KwaZulu-Natal, Mpumalanga, Swaziland and southern Mozambique........... 13

13a. Plants up to 3 m tall with erect leafy stems and short seasonal shoots; stipules subulate, swollen, recurved, woody and persistent on old shoots; calyx variously hairy, dark green or purplish; flowers mauve, standard petal with large white V-shaped nectar guide, veins purple; wing petals mauve…… .... **11. *P. glabra***

13b. Plants up to 2 m tall with many rigid branchless stems bearing clusters of seasonal flowering shoots radiating either terminally from stems or along older shoots giving a broom-like appearance; stipules triangular, flat, erect to semi-patent, not prominent or persistent on old shoots; calyx glabrous, glaucous or pale green; flowers white or greenish white; standard petal without large white V-shaped nectar guide, veins either translucent or pale violet; wing petals white..... ....**12. *P. margaretiflora***

8b. Reseeder (killed by fire)...14

14a. Seasonal shoots glabrous or glabrescent...15

15a. Peduncle filiform and flexuous, more than 45 mm long; flowers white to mauve ....**13. *P. imminens***

15b. Peduncle stout and rigid, up to 45 mm long; flowers blue to purple………………... 16

16a. Leaflets flat, linear-oblong to linear-lanceolate, 2.5−5(6) mm wide, base cuneate, apex apiculate............... ....**14. *P. latifolia***

16b. Leaflets cylindrical, linear lanceolate, 0.2−1.5 mm wide, apex and base acute ……. 17

17a. Leaves 5-foliolate, leaflets curving; flowers 17−18 mm long; inflorescences borne vertically on short vernal shoots arising off horizontal branches; calyces 10−12 mm long …..………… **15. *P. speciosa***

17b. Leaves more than 5-foliolate, leaflets straight or curving; flowers 13−16 mm long; inflorescences borne on standard shoots; calyces less than 10 mm long........................ 18

18a. Leaves 9−11-foliolate……….. 19

18b. Leaves 5−7 (9)-foliolate.. 20

19a. Leaflets up to 50 mm long, flexuous and filiform; inflorescences congested; peduncle shorter than the subtending leaf, 6–15 mm long; calyx 8−9 mm long, papery, shaggy white-haired; flowers 15−16 mm long, bright blue, strongly sweet scented; extends from Wolseley and Romansrivier southwards to Somerset West and Sir Lowry's Pass then westwards towards Hermanus and Caledon from Tulbagh to Riviersonderend..... ....**16. *P. ivumba***

19b. Leaflets up to 35 mm long, stout and rigid; inflorescences lax, peduncle equal or longer than the subtending leaf, 17–30 mm long; calyx 7–8 mm long, thick, nigro-hirsute; flowers 12–15 mm long, dark blue to dark purple, weakly scented or odourless; extends from Mossel Bay to Tsitsikamma Mountains...**17**. ***P. affinis***

20a. Small to large showy shrub forming an umbrella shape; peduncles with black hairs, 25–35 mm long; flower hyacinth blue; cupulum overlapping the calyx; calyx 7−8 mm long, covered with a mixture of finely black and white hairs, lobes equally developed; endemic between Stilbaai and Gouritz … ....**18. *P. brilliantissima***

20b. Small to large robust shrub not forming an umbrella shape; peduncles glabrous, 30−45 mm long; flower mauve to purple; cupulum not overlapping calyx; calyx 5−6 mm long, glabrous, lobes sub-equally developed; endemic to Swartberg mountains …**19. *P. elegans***

14b. Seasonal shoot pubescent...21

21a. Shoot densely pubescent; leaves clustered at the end of bare branches on old shoots, 9−13-foliolate, basal pair of pinnae attached or separate at the node; inflorescence lax; calyx tube more than or equals to the length of the lobe; endemic to South Langeberg to Tsitsikamma Mountains...**20. *P. floccosa***

21b. Shoot sparsely pubescent; leaves distributed along the length of an old shoots, 7−11-foliolate, basal pair of pinnae separate at the node; inflorescence congested; calyx tube less than the length of the lobe; endemic to Cederberg mountains.................... 21. ***P. intonsa***

1b. Large shrubs to trees, more than 3 m tall...22

22a. Leaves (13)15–19-foliolate; leaflets less than 25 mm long; flowers white, strongly scented………………...**22. *P. odoratissima***

22b. Leaves 7–9(11)-foliolate; leaflets more than 25 mm long; flowers deep blue or blue or mauve to pale blue, weakly scented or unscented …...23

23a. Leaflets up to 50 mm long; peduncle up to 60 mm long; flower deep blue to purple; calyx deeply pubescent with deep black hairs ....**23. *P. arborea***

23b. Leaflets less than 50 mm long; peduncle absent or less than 60 mm long; flower pale mauve, blue or pale blue; calyx sparsely pubescent with black or white hairs or both .. 24

24a. Leaflets flat, linear, 20–45(50) mm long, 1.5−2.5 mm wide, base cuneate, apex apiculate; peduncle equal or longer than the subtending leaflet, 25–45 mm long; inflorescences lax, exposed above the subtending leaf; flowers blue; calyx with fine silvery hairs; endemic to mountains between George to Humansdorp.....**24. *P. montana.***

24b. Leaflets cylindrical, linear lanceolate, 15–35 mm long, 0.5−1.5 mm wide, base rounded, apex acute; peduncle absent or shorter than the subtending leaf, 2–3 mm long; inflorescences congested, hidden within the subtending leaf; flowers pale mauve or pale blue; calyx with a mixture of black and white hairs; endemic to Cape Peninsula to Kogelberg Mountains………………………………………………………… **25. *P. pinnata***

4.6.2. Descriptions of the species

4.6.2.1. *Psoralea rhizotoma* C.H. Stirt. & Muasya *sp. nov.* Type: South Africa, Eastern Cape, Umtata (3128): Mhlahlani Forest Reserve, Transkei (-BC), 24 Jan 1986, *Stirton C.H. 11140* (K (K000392587), holo.!; PRE!, iso.).

Erect multi-stemmed suffrutex to 1.5 m tall and ± 1 m wide. *Stems* many, semi-prostrate along river banks but ascending to erect in grassland, branching at or near the base, branches lax, longitudinally-striated and slightly angular, flexuous. *Seasonal shoots* smooth, glabrous, bright green. *Leaves* 7−9-foliolate, imparipinnate, pinnae reducing in size and number from base of seasonal shoot to flowering end until disappearing completely, patent, petiolate; leaflets 14–30(40)×0.5–2 mm, linear lanceolate, basal pair longest, terminal leaflet second longest, uppermost leaflets digitately to slightly pinnately inserted, glabrous; petioles 4–6 mm long, shorter than terminal leaflet; rachis 5−10 mm long; stipules 2−3×1.5−2 mm, persistent, patent, shorter than petiole, drying black. *Inflorescences* axillary, borne in most axils of seasonal shoots, fasciculate, pseudo-spicate, lax, 1-flowered, 1−6 per axil, pedunculate, pedicellate; peduncles 30–35 mm long, filiform, flexuous, longer than the subtending leaf; cupulum 1.5 mm long, glabrous, bilobed, one of the lobes cleft, lobes equally developed, triangular; pedicels 2–3 mm long. *Flowers* 11−14 mm long, reddish purple. *Calyx* 5−6 mm long; tube 3–4×3.5 mm, glabrous, shiny, greenish; lobes purplish, strongly ridged, shorter than the tube, equally developed, oblong-ovate, sub-acute, vexillar lobes fused one quarter their length; both tube and lobes glabrous to pubescent on the outer surface, but with stubby black hairs within the lobes; margins ciliate. *Standard petal* 11–12×8–9 mm, ovate-oblong, deep purple with large white M-shaped nectar guide subtending a short dark purple patch in the crotch, veins of blade darker, appendages present, auricles well-developed, large and swollen, apex rounded; claw 3–4 mm long. *Wing petals* 10–11×3–3.5 mm, white to pale purple, blade overlapping, capping the keel, folded and puckered along its mid-line; sculpturing present, basal, comprised of 2–3 rows of 9–12 transcostal lamellae; claw 3.5–4.5 mm long. *Keel* 9–10×3.5–4 mm, white, apically suffused with dark violet-purple; claw 4–5 mm long. *Androecium* 11 mm long, tenth stamen free, fenestrate. *Pistil* 11 mm long, stipe 1 mm long, ovary 2 mm long, sparsely glandular near apex, style 5 mm long, straight then up flexed, swollen at point of flexure, height of curvature 3 mm, stipitate, penicillate. *Fruits* oblong, 3–4×2.5–3.0 mm. *Seeds* solitary, 2–3 x 2 mm, dark brown to black (Figure 6).

4.6.2.1.1. Diagnostic characters

Psoralea rhizotoma is easily distinguished by its rhizomatous rootstock, decumbent to ascending floppy flexuous branching habit, long (30–35 mm) peduncles, shiny greenish purple glabrous calyces, striking reddish purple flowers with standard petal usually flushed violet and with a large white M-shaped nectar guide and a purple flash in the cleft. It is similar to *P. glabra* in growth habit but differs in its multi-stemmed suffrutex habit growing up to 1.5 m tall and ± 1 m wide (versus an erect or sub-erect rigid resprouting shrub to 3 m tall); with variable numbers of leaflets on flowering shoots (versus same number of leaflets); inflorescences axillary, lax, distributed along the length of seasonal shoots with variously 1−6 flowers per axil (versus axillary in upper axils of seasonal shoots, congested with 1–3 flowers per axil); flowers reddish purple (versus flowers mauve to blue), standard blade ovate-oblong (versus obovate), and peduncles 30−35 mm long, filiform and flexuous (versus 4−11 mm long, stout and rigid).

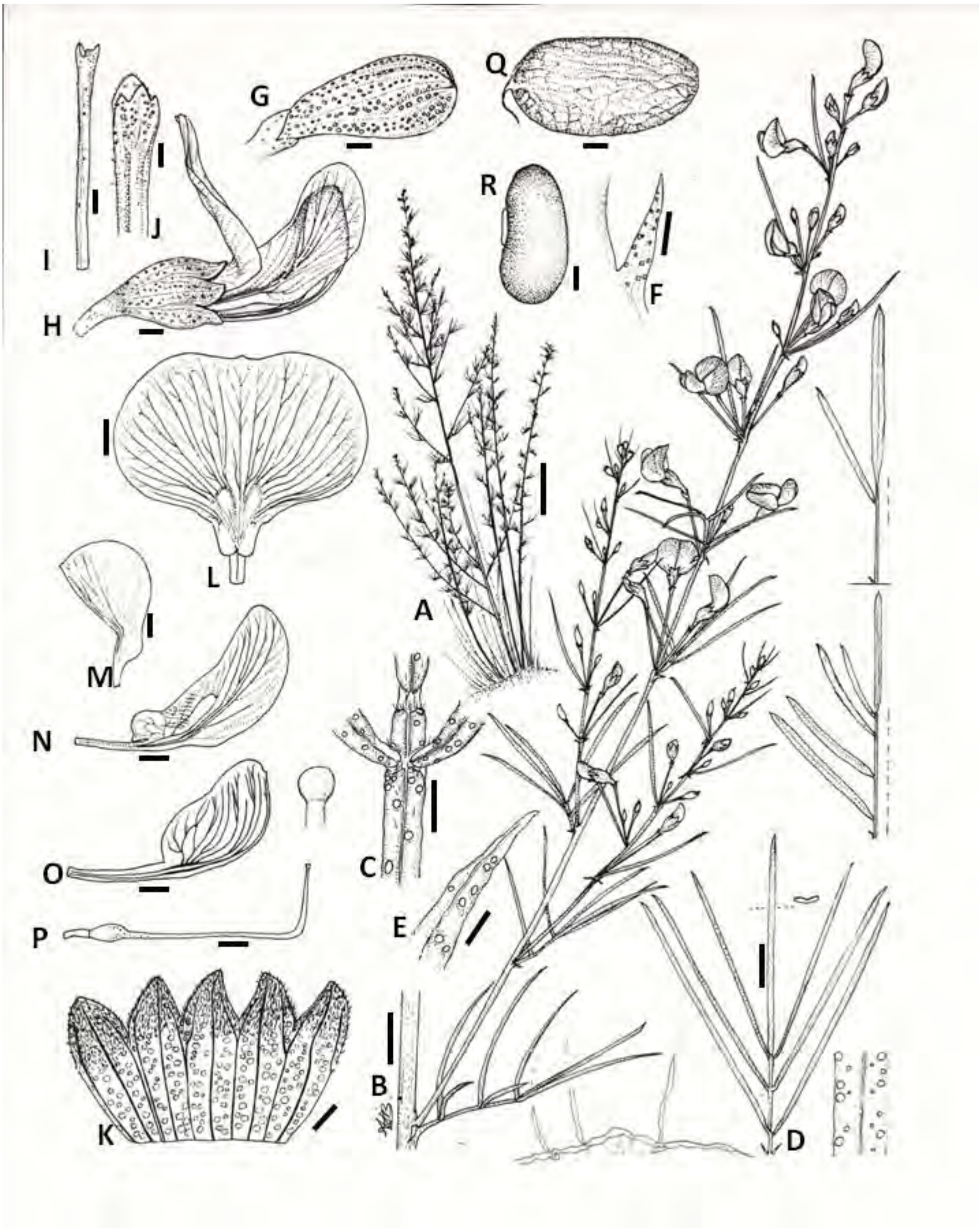


Figure 6. Vegetative and reproductive morphology of *P. rhizotoma* (A) habit; (B) flowering shoot; (C) leaf insertion; (D) 7-foliolate leaf (E) leaflet; (F) stipule (G) bud; (H) flower; (I) peduncle terminating with a cupulum; (J) cupulum; (K) calyx opened out with vexillary lobe to the right; (L) front view of standard petal; (M) side view of standard petal; (N) wing petal; (O) keel; (P) pistil; (Q) fruit; (R) seed. Voucher *Stirton C.H. 11140* (NBG). Scale bars: A−F, 1 cm; G–R, 1 mm. Artist: Angela Beaumont.

4.6.2.1.2. Distribution and habitat

Psoralea rhizotoma has a wide distribution and extends from the Eastern Cape Province to the Drakensberg Mountains in KwaZulu-Natal and Mpumalanga provinces (Figure 7). It occurs in montane grassland and along stream sides and riverbanks between 1500−1600 m elevations. It is found growing mainly in Pondoland-Ugu Sandstone Coastal Sourveld, Amathole Montane Grassland, Midlands Mistbelt Grassland, Drakensberg Foothill Moist Grassland, Tsomo Grassland, Northern Zululand Sourveld and Bhisho Thornveld vegetation types (CB 4, Gd 1, Gs 9, Gs 10, Gs 15, SVl 22, and SVs 7) (Mucina *et al.* 2006b; Mucina *et al.* 2006c; Rutherford *et al.* 2006).

4.6.2.1.3. Etymology

The specific epithet *rhizotoma* is derived from the Modern Latin *rhizoma* (root) and from the Classical Greek word *rhizōma*. *Rhizoma* is an adjective referring to the nature of the rootstock of this species.

4.6.2.1.4. Conservation status

Psoralea rhizotoma has been given a status of least concern (LC) after evaluating it against the five IUCN criteria of threat (IUCN 2012a, 2012b). It did not qualify for any of the threat categories as it is widely distributed and usually locally abundant where it occurs. As many localities are in protected areas, there are no immediate threats, but outside these areas, it is threatened by the destruction of grassland by overgrazing and ploughing.

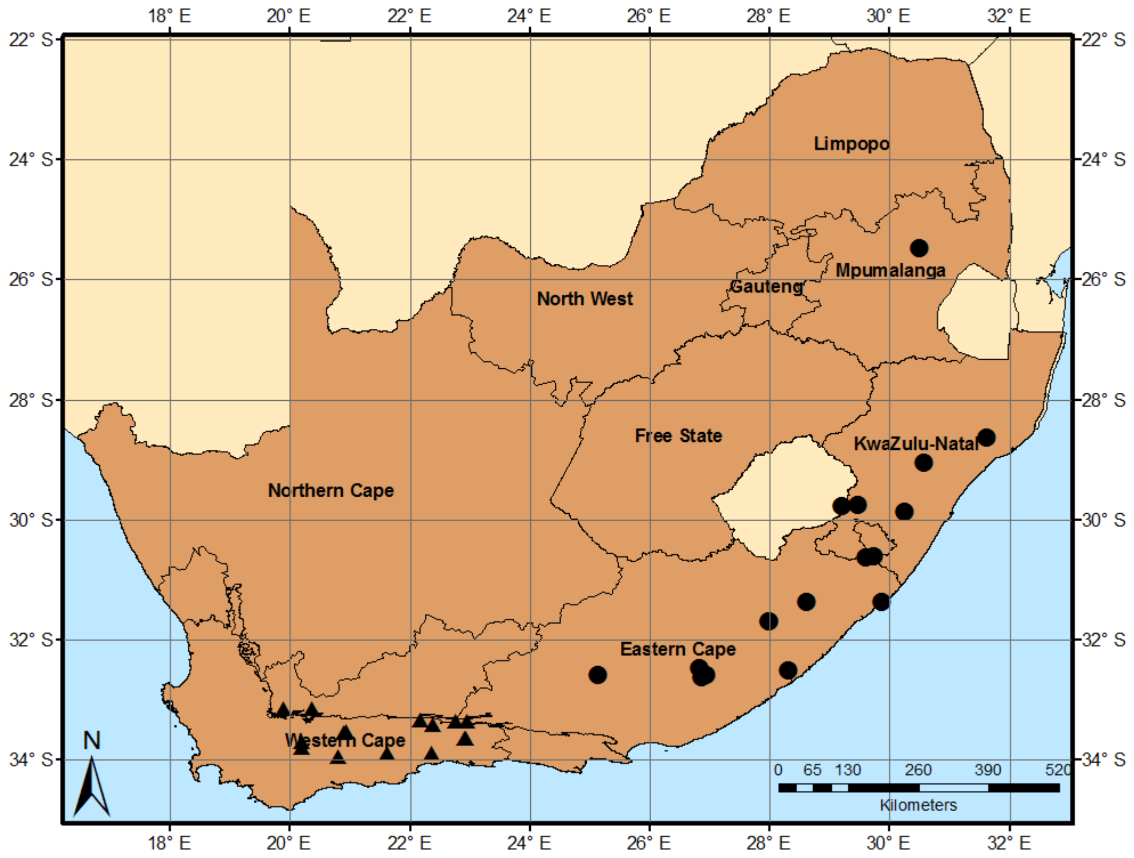


Figure 7. Known distribution of *P. rhizotoma* (circles) and *P. azuroides* (triangles).

4.6.2.1.5. Specimens studied

South Africa. MPUMALANGA: **2530 (Lydenburg):** Buffelskloof Nature Reserve (-AA), 30 Nov 2012, *Muasya A.M. & Stirton C.H. 7076* (BOL); Buffelskloof Nature Reserve (-DA), 12 Mar 2003, *Nkuna L.A. & van Wyk B. 1512* (PRE, PRU). KWAZULU-NATAL: **2831 (Nkandla):** Nkandla Forest (-CA), 19 Jun 1956, *Edwards T. 1467* (PRE). **2929 (Underberg)**: Giant's Castle Nature Reserve (-AD), 18 Jan 1965, *Trauseld W.R. 363* (PRE); Sani Pass Hotel, Sani Pass (-CB), 5 Feb 1982, *Bamps P.J.R. 7160* (PRE); Glengariff Farm, Mahaqwa Mountain (-CD), 12 Jan 2008, *Muasya A.M. & Stirton C.H. 3659* (BOL). **2930 (Pietermaritzburg):** 2 Jan 1929, *Wylie J. s*.n. (K); Byrne-Cunningham's Castle (-CC), 15 Jan 1985, *Nicholas A. & Neave D. 2100* (K). EASTERN CAPE: **3029 (Transkei)**: Farm Lynn Avis, 11 m on Ixopo-Donnybrook road (-CC), 29 Jan 1964, *Crewe R. 27* (K, NU); Kokstad (-CC), 22 Feb 1972, *Strey R.G. 10760* (K, PRE); eastern lower slopes of Mt. Ngeli (-DC), 17 Dec 1981, *Stirton C.H. 10430* (PRE); Ingeli Mountain, East Griqualand, Mar 1883, *Tyson W. 448* (K). **3128 (Umtata):** Mhlalani Forest Reserve (-BC), 24 Jan 1986, *Stirton C.H. 11140* (K, PRE). **3129 (Port St. Johns)**: St. Bernards, East Griqualand (-BD), 25 Dec 1952, *Barker W.J. 8003* (NBG, PRE). **3223 (Willowmore)**: Palmietfontein, Camdeboo mountain, Aberdeen District (-BD), 9 Feb 1960, *Acocks J.P.H. 21044* (PRE). **3226 (Fort Beaufort)**:

Cavers, Bedford (-CA), 22 Dec 2015, *Muasya A.M. 7292* (BOL); Hogsback, Stockenstrom (-BD), 15 Dec 1940, *Barker W.J. 894* (NBG). **3227 (Stutterheim)**: Evelyn Valley, Stutterheim Division (-CB), *Leighton F.M. 2693* (PRE); Gaika's Kop, Amatole Mountains (-DB), 16 Jan 1990, *Stirton C.H. 12492* (PRE). **3228 (Butterworth)**: Kentani (-CB), 1902, *Pegler A. 255* (PRE).

4.6.2.2. *Psoralea vanberkelae* C.H.Stirt., A.Bello & Muasya, in PhytoKeys 44: 102 (2015). Type: South Africa. Western Cape, Knysna (3423); Robberg Coastal Corridor, Fynbos Private Nature Reserve (-AB), 18 Oct 2013, *van Berkel N. 1118* (BOL, holo.!; GRA!, K!, NBG!, SCHG!, PRE!, iso.).

Sprawling and mounding suffrutex to 60 cm tall and up to 1.5 m wide, resprouter. *Stems* 1–10, branching in upper parts; branches erect, rough, grey, mostly bare except for upper parts. *Seasonal shoots* rough, blackish, hairy. *Leaves* tightly parked, 5-foliolate, petiolate, fleshy; basal leaves of seasonal shoots smallest, patent to semi-erect, surface bumpy, glabrous, glands raised, hyaline but drying reddish brown to black; leaflets 10−11×0.5 mm, linear oblong, glandular, flat on adaxial surface with a distinct furrow, basal pair equal to or slightly shorter than the terminal leaflet, three uppermost digitately inserted; petioles 2–3 mm long; rachis 7 mm long, grooved; stipules 2×1 mm, acuminate, fused, joined by a bridge of tissue, glabrescent, teeth broadly triangular, apex acute, fleshy, persistent, becoming prominent and woody when leaves are shed, hairy, hairs short and stubby, densely covered with large raised glands. *Inflorescences* axillary in upper nodes of short seasonal shoots, borne solitary per axil, pedunculate, pedicellate; peduncle short, 2 mm long, hairy; peduncle bracts paired, minute; cupulum 1 mm long, pale green, trifid, shortly triangular, lobes equal, black-haired, covered in large glands, drying reddish brown; pedicel 2 mm long. *Flowers* 10−11 mm long, pale to intense mauve to blue. *Calyx* 6 mm long, tube 4×4 mm, ribbed; lobes slightly subequal, shorter than tube, 2 mm long, pale green, sparsely covered in small black flat hairs and densely encrusted with mixed sized glands on outside, margins of lobes densely black ciliate, inside of lobes densely stubby black-haired, vexillar lobes scarcely fused above tube. *Standard petal* 9−10×7−8 mm, very broadly ovate, reflexed to 90 degrees, apex rounded, mauve but dark purple in central area above the M-shaped white nectar "guide", venation purple, callosities above the claw absent; claw 2−3 mm long, flattened, erect. *Wing petals* 6−7×3−4 mm, longer than keel petals, strongly folded once along middle, slightly billowy near apex, held parallel to keel, strongly auriculate, sculpturing present, upper basal comprising 4−5 transcostal lamellae; claw 4−5 mm long. *Keel* 5−6×3−4 mm; claw up to 5 mm long. *Androecium* 7 mm long, tenth stamen free, sheath split abaxially, fenestrate, nectarial ring present, 0.3 mm high. *Pistil* 7 mm long, ovary 2 mm long, stipitate, glabrous but sparsely covered in curved stalked glands across sides, stigma penicillate. *Fruits* oblong 5×3 mm, papery, rugose, reticulate, brown. *Seeds* solitary, 4×2.5 mm, oblong-elliptic, khaki with black mottles and flecks, hilum central (see Figure 2, Plate 2 in Bello *et al.* 2015).

4.6.2.2.1. Diagnostic characters

Psoralea vanberkelae is a recently described member of the *P. pinnata* complex. Like *P. muirii* it has distinctive raised glands on its clasping, tightly packed leaves; persistent stipules which becomes woody on old seasonal shoots; bare and rough old stems and branches; and a resprouting mode of regeneration after fires. It however differs from *P. muirii* in its shorter stature of a sprawling, mounding and colonial suffrutex habit to 60 cm tall and up to 1.5 m wide (versus erect shrub to 1.5(−2) m tall, 0.5 m wide); glabrous, fleshy, and congested 5-foliolate leaves (versus glabrescent, non-fleshy, 5−7-foliolate, flexible leaves); and many, pale to intense mauve to blue flowers in congested pseudo-inflorescences (versus mauve, fewer, less congested pseudo-inflorescence flowers).

4.6.2.2.2. Distribution and habitat

Psoralea vanberkelae is a narrow endemic known from only a few hundred individuals in a small area of the Robberg Coastal Corridor between George and Knysna (see Figure 3 in Bello *et al.* 2015). It occurs in South Outeniqua Sandstone Fynbos (FFs19, Rebelo *et al.* 2006) in a mixture of Eastern Fynbos and Renosterveld. It grows in full sun on thin sandy soils overlying Peninsula formation quartzite on gentle slopes. As the area was subjected to a controlled burn in April 2008 the plants are becoming moribund.

4.6.2.2.3. Etymology

The specific epithet *vanberkelae* honours Ms. Nicky van Berkel, a C.R.E.W. volunteer and iSpotter ("Nicky"; http://www.ispot.org.za/user/10095), who brought this species to our attention and sent us reference material and photographs. Like many plant enthusiasts from C.R.E.W. she plays a valuable role in establishing the conservation status of plants in her area. The plant is a beautiful flagship species for a very threatened habitat.

4.6.2.2.4. Conservation status

Psoralea vanberkelae is locally abundant in its habitat and the main population is protected by private ownership (Fynbos Private Nature Reserve). It is, however, restricted to a narrow range of distribution (area less than 20 km^2). The coastal stretches where the plants occur are all on private land with limited access. The cliff edges rise sharply from the sea and their escarpments are not easy to access. We therefore assess this species to be Vulnerable under the South African Red List categories and criteria (VU D2, von Staden *et al.* 2009, IUCN 3.1, 2012a, 2012b).

4.6.2.2.5. Specimens studied

South Africa. WESTERN CAPE: **Knysna (3423):** Robberg Coastal Corridor, Fynbos Private Nature Reserve (-AB), 18 Oct 2013, *van Berkel N. 1118* (BOL, GRA, NBG, SCHG, PRE); 14 Feb 2014, *Bello A., Stirton C.H. & Muasya A.M. 53* (BOL).

4.5.2.3. *Psoralea azuroides* C.H. Stirt. *sp. nov.*, Stirton & Schutte in Goldblatt & Manning Strelitzia 9: 505 (2000); *Psoralea* sp. 20, Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Western Cape, Oudtshoorn (3322): Northern slopes of Outeniqua mountains (-CD), 12 Nov 1986, *Stirton C.H., Žantovská J. & Vlok J.H.J. 11574* (K!, holo.).

A compact suffrutex forming dense clumps, 0.2−0.8 m tall. *Stems* erect, 10−60, branching at the base, arching in upper parts, rigid, brown with white storied lenticels. *Seasonal shoots* smooth, glabrous, yellowish green; older shoots blackish purple. *Leaves* 5−foliolate, imparipinnate, distributed along the branches, held tightly to the stem, terete, spreading to patent, petiolate; leaflets 15–22×0.3–0.7 mm, glandular, linear lanceolate, terminal leaflet shortest, basal pair of leaflets longest, three uppermost digitately inserted; petioles 6–7 mm long, shorter than terminal leaflet; rachis 5–10 mm long; stipules 1−2×0.5−1.5 mm, shorter than petiole, narrowly triangular, glabrous outside, finely hairy inside, subulate. *Inflorescences* axillary, borne in upper axils of seasonal shoots without leafy extension, one-flower per axil, fasciculate, 1−5 per shoot, pseudo-capitate, lax, pedunculate, pedicellate; peduncles stout and rigid, 12–14 mm long; cupulum bilobed, setose or linear, 2 mm long, glabrous, terminal lobe longest, abaxial lobe with cleft, pedicels 1–4 mm long. *Flowers* 13–15 (17) mm long, each subtended by 2 minute basal hairy bracts on either side, deep azure blue to mauve. *Calyx* 6–7 mm long, tube 3–4×3.7 mm, glabrous or villoso-pubescent, ribbed, with dense flat glands, constant in size; lobes sub-equally developed, black-fringed, vexillar lobe longest, shorter than the tube, all triangular, acute, vexillar lobes fused for 1 mm above the tube; inner face of lobes densely black pubescent, glabrous outside. *Standard petal* 12–13×10–12 mm, very broadly ovate, very slightly emarginate, azure blue to mauve with a central purple flash fading towards the apex, veins darker; auriculate, appendages well-developed, swollen; claw 2–3 mm long. *Wing petals* 12–13×3–5 mm, cultrate, same colour as the standard, upper basal comprised of 2–3 rows of 6–7 transcostal lamellae bulging along mid-line; claw 2–3 mm long. *Keel* 9–10×3–4.5 mm, apex obtuse; claw 4–5 mm long. *Androecium* 11 mm long, tenth stamen lightly adherent to right hand base of sheath margin, scarcely fenestrate. *Pistil* 8 mm long, ovary 1.5 mm long, stipitate, upper end covered in short curved club-shaped glands; style 5 mm long, straight, then curved, height of curvature of 3 mm, widest at point of flexure, stigma capitate, penicillate. *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* solitary, 2–4×2 mm, dark brown to black (Figure 8).

4.6.2.3.1. Diagnostic characters

Psoralea azuroides is characterised by its compact generally mound-like habit to about 0.8 m tall and 1 m wide, yellowish green seasonal shoots borne on the upper portion of brownish branches with small white storied lenticels; stiff, rigid spreading leaves, and the most intense azure blue (more rarely mauve) flowers. It is similar to *P. laevigata* in growth form but differs in its dull, stiff, spreading leaves (versus shiny, smooth, claw-like clasping leaves); narrowly clasping semi-patent stipules (versus patent, broadly spreading stipules); shorter peduncles, 12−14 mm long (versus longer peduncles, 23−25 mm long); 1-flower per axil (1−3-flowers per axil); dull azure blue to mauve flowers (versus shiny, deep purple flowers); and standard petals with large swollen appendages (versus standard petals with poorly developed appendages). The sympatric species *P. laevigata* and *P. speciosa* may be confused with *P. azuroides* vegetatively in the field as all have short 5(7)-foliolate leaves of almost the same length but they differ in their habit and leaf details. *Psoralea laevigata* is an ascending to erect multi-stemmed suffrutex up to 1 m tall with very shiny, smooth, slippery, claw-like clasping leaflets. *Psoralea speciosa* is a large, robust, reseeding shrub up to 2.5 m tall with 1−3 short arcuate stems giving rise to short vertical vernal flowering. *Psoralea laevigata* and *P. speciosa* both have deep purple flowers with a large white M-shaped patch just above the appendages (absent in *P. azuroides*).

4.6.2.3.2. Distribution and habitat

Psoralea azuroides is endemic to the Langeberg and Outeniqua Mountains in the Western Cape Province (Figure 7). It occurs in mountain fynbos on dry slopes at elevations between 260−1130 m, being particularly prevalent on Tschando shale bands. It is restricted to the South Sonderend Sandstone Fynbos, South Outeniqua Sandstone Fynbos, North Outeniqua Sandstone Fynbos, South Kammanassie Sandstone Fynbos and Eastern Rûens Shale

Renosterveld vegetation types (FFs 14, FFs 16, FFs 19, FFs 26 and FRs 13) (Rebelo *et al.* 2006; Rutherford *et al.* 2006).

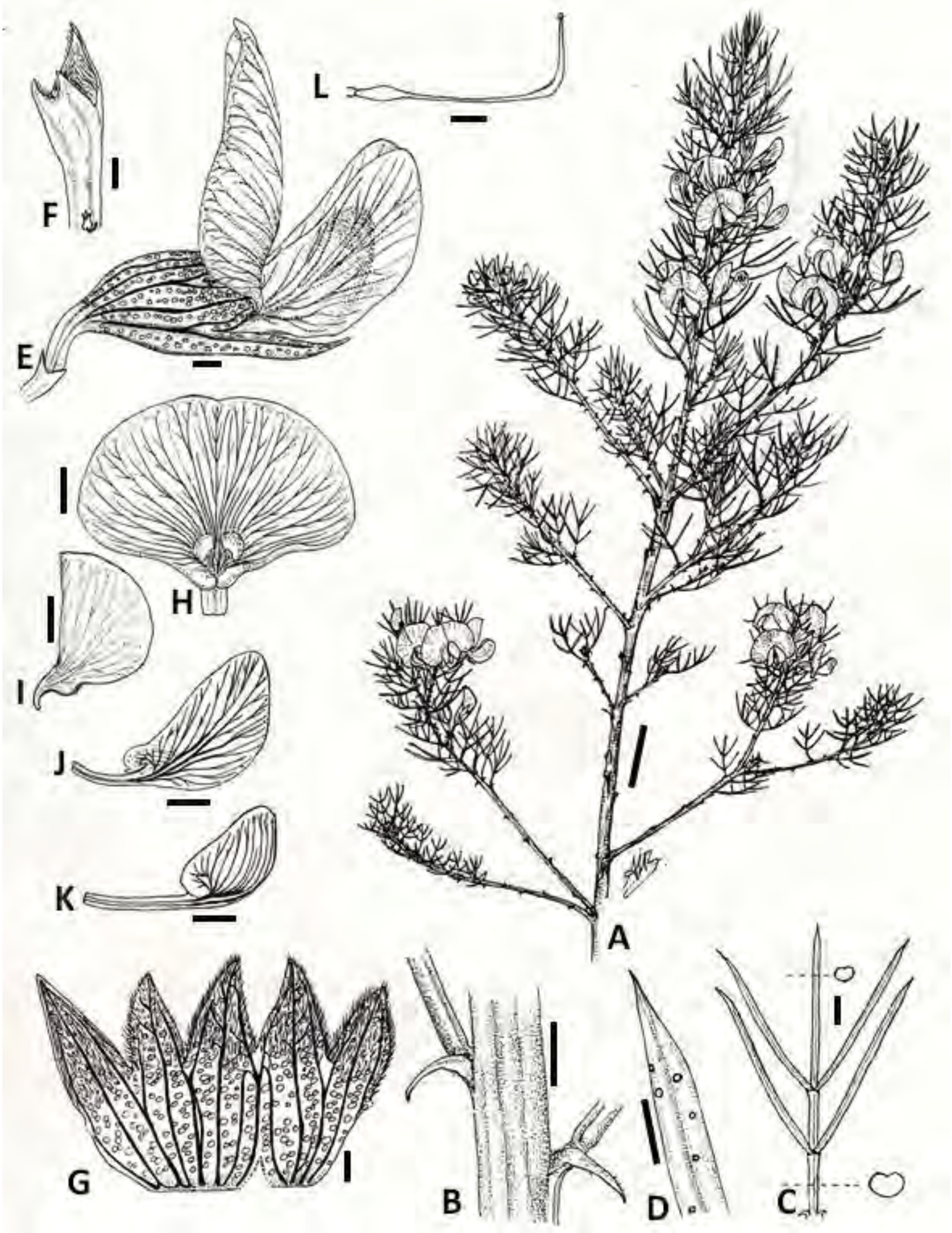


Figure 8. Vegetative and reproductive morphology of *P. azuroides* (A) flowering shoot; (B) stipule on a twig; (C) 5-foliolate leaf; (D) leaf blade showing glands; (E) flower; (F) cupulum; (G) calyx opened out with vexillar lobes to the right; (H) front view of the standard petal showing swollen appendages; (I) side view of the standard petal; (J) wing petal; (K) keel; (L) pistil. Voucher *C.H., Stirton, J. Žantovská & J.H.J. Vlok 11574* (PRE). Scale bars: A, B, D 1 cm; C, E–L, 1 mm. Artist: Angela Beaumont.

4.6.2.3.3. Etymology

The specific epithet *azuroides* is derived from Middle English *asur* and Anglo-French *azeure* and alludes to the distinctive deep blue flower colour of this species.

4.6.2.3.4. Conservation status

Psoralea azuroides has a broad range of distribution and it is safe where it occurs. We therefore assessed it as Least Concern (LC, von Staden *et al.* 2009, IUCN 3.1, 2012a, 2012b).

4.6.2.3.5. Specimens studied

South Africa. WESTERN CAPE: **3320 (Montagu)**: Cogmanskloof (-CC), 8 Oct 1922, *Tredgold D.K. 410* (PRE); Tradouw Pass (-DC), 26 Oct 1986, *Stirton C.H. & Žantovská J.Z. 11255* (K); Voormansbosch (-DC), Oct, *Zeyher C.L.P. 238* (MO); 4 km west of Helderfontein hut in Bosmansbos Wilderness area along trail on north side of ridge, Moerasrivier, Langeberg (-DD), 1 Jan 2006, *Helme N.A. 3760* (NBG). **3321 (Ladismith)**: Berfontein, close to Witblitz paadjie, west slopes of Witteberg, above Witelsrivier, Langeberg (-DC), 12 Dec 1990, *Mc Donald 2042* (NBG). **3322 (Oudtshoorn)**: Near summit, Robinson's Pass (-AC), 1 May 1933, *Salter T.M. 3272* (K); 27 Jan 2013, *Muasya A.M. & Stirton C.H. 6896* (BOL); upper southern slopes of Ruitersberg, Outeniqua mountains, (-CC), 30 Nov 1986, *Vlok J.H.J. 1733* (PRE); Between tributaries of Perdeberg, Ruitersbos Forest Reserve (-CC), *Thompson M.F. 2701* (K, NBG); top of Outeniqua Pass, George Division (-CC), 19 Oct 1953, *Lewis G.J. 3704* (PRE); Four Ways Cross (-CC), 3 Jan 2008, *Muasya A.M. & Stirton C.H. 4610* (BOL); Hopsvallei Farm, Tweewatersberg, Outeniqua Mountains (-CD), *Muasya A.M. & Stirton C.H. 5070, 5072* (BOL); Robinson's Pass, Outeniqua Mountains (-CC), May 1936, *Laughton F.S. 1473* (PRE); 28 Sep 1960, *Middlemost G. J. 2112* (NBG); 2 Jan 2008, *Muasya A.M. & Stirton C.H. 3612* (BOL, PRE); 2 May 1974, *Goldblatt P. 1778* (MO, PRE); Jonkersberg top of Robinsons' Pass (-AC), 25 Jan 1986, *Vlok J.H.J. 1378* (PRE); above the Outeniqua Pass, George Division (-CD), 22 Jul 1962, *Taylor H.C. 3437* (NBG, PRE); Noordekant van kruin van Outeniqua pas, noord van George (-CD), 13 Apr 1985, *Grobbelaar N. 2948* (PRE); Cradock's Berg (-CD), 11 Aug 1814, *Burchell W.J. 5939* (L); upper slopes of Groot Doring River catchment next to Forestry track, North slopes of Outeniqua Mountains (-CD), 12 Dec 1986, *Stirton C.H. 11574* (PRE); Mannetjiesberg, Kammanassie, Uniondale District (-DB), 1 Feb 1941, *Esterhuysen E. 4709,* 3 Nov 1941, *6499* (BOL). **3323 (Willowmore):** 5 m from Avontuur to Prince Alfred's Pass (-CC), 12 Feb 1964, *Wells M.J. 2848* (K, PRE); Prince Alfred's Pass, 16 Feb 2014, *Stirton C.H., Bello A. &*

Muasya A.M. 13869 (BOL). **Without precise locality:** S-SW of Langeberg, 21 Jan 1969, *Marsh J.A. 1105* (NBG, PRC); C.B.S., *Zeyher 862* (M**).**

4.6.2.4. *Psoralea semota* C.H. Stirt. *sp. nov.* *Psoralea sp.* 30, Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Western Cape, Caledon (3419): Schilpadkop, Riviersonderend mountains (-BA), 2 Jan 1953, *Esterhuysen E. 21045* (PRE, holo.!; BOL!, iso.).

Multi-stemmed mounding suffrutex up to 1 m tall and 1.5 m wide. *Stems* semi-erect to ascending, many, branching at or near the base, spreading laterally and sprawling or arching, rigid. *Seasonal shoots* pubescent, dark green. *Leaves* (9)11–13(15)-foliolate, imparipinnate, distributed along the branches (at higher elevations old branches bare and leaves tightly clustered at tips), patent, petiolate, sparsely pubescent, mucronate; leaflets 8–15×0.5–1.5 mm, linear to linear lanceolate; terminal leaflets shortest, basal pair longest, three uppermost pinnately inserted, upper surface of mature leaflets shiny, tips sinuous, glabrescent; petioles 1–2 mm long, shorter than terminal leaflet; rachis 5−7 mm long; stipules 1–2×0.5–1 mm persistent, becoming woody with age, shorter or equal in length to petiole, covered with black hairs. *Inflorescences* axillary, borne along the length of seasonal shoots, pseudo-spicate, lax, one flower per axil, fasciculate, 1−6 flowers per shoot, pedunculate, pedicellate; peduncles flexible, 30–40 mm long, longer than the subtending leaf, densely clothed with patent black hairs; cupulum shallowly bilobed, overlapping the base of the calyx, densely covered with patent black hairs, lobes equally developed, narrowly triangular; pedicels 1–2 mm long, clothed with black hairs. *Flowers* 12−13 mm long, deep blue. *Calyx* 5–6 mm long, tube 3–4×4 mm, covered with bristly black hairs both within and outside; lobes sub-equally developed, lower lobe slightly longer, lateral straight, triangular, acute, broader than the other four lobes, vexillar lobes fused for up to one third their length, inner face of the lobes covered with black bristle-like hairs. *Standard petal* 9–10×6–8 mm, elliptic, apex acute, deep blue with white striations, dark blue on the margins with nectar guide comprised of two narrowly triangular M-shaped patches, veins darker blue; claw 3–4 mm long. *Wing petals* 8–9×3–3.5 mm, white with purplish violet fading at the tip; claw 3–4 mm long. *Keel* 9×3–3.5 mm wide; claw 4–5 mm long. *Androecium* 10 mm long, scarcely fenestrate. *Pistil* 8 mm long, stipitate; ovary 1 mm long, glabrous, sparsely covered in club-shaped glands; style 6 mm long, straight, then curved, glabrous, height of curvature 2.5 mm long, thickened at point of flexure. *Fruits* unknown. *Seeds* unknown (Figure 9).

4.6.2.4.1. Diagnostic characters

Psoralea semota is similar to *P. affinis* in occupying the same habitat from which it differs in being a compact sprawling to erect multi-stemmed suffrutex <1 m tall and up to 1.5 m wide (versus erect single-stemmed reseeding shrub to 3 m tall), with pubescent leaves (versus sparsely pubescent or glabrous leaves), densely black haired cupula, and pedicels (versus glabrous), peduncles longer than subtending leaflets (versus peduncles nearly as long as subtending leaflets), and flowers borne along the length of seasonal flowering shoot (versus flowers borne at the apex of seasonal flowering shoot). *Psoralea semota* is also similar to *P. suaveolens* in having more than 5 pairs of leaflets and elongated peduncles. They however differ in that *P. semota* is a compact resprouting suffrutex (up to 1.5 m wide and 1 m tall) whose leaves, peduncles, cupulum and calyx are covered dense black hairs; whereas *Psoralea suaveolens* is a tall multi-stemmed resprouting shrub up to 2 m tall with glabrous leaves and flowers. At higher elevations *P. semota* becomes a low spreading decumbent shrub with mostly bare stems with the leaves tightly clustered at the ends of branches. More fieldwork is needed to distinguish the Langeberg and Riviersonderend populations as they may represent distinct taxa. We have not studied this rare species in the field.

4.6.2.4.2. Distribution and habitat

Psoralea semota is a high montane species and occurs on South and South East slopes in well-drained sandy soil in mountain fynbos between 1000–1330 m above sea level. It extends from the Hex River Mountains to the Langeberg and Riviersonderend mountains in the Western Cape Province of South Africa (Figure 10). It is distributed entirely in protected areas and has been recorded in Breede Shale Fynbos, North Hex Sandstone Fynbos, North Sonderend Sandstone Fynbos, South Sonderend Sandstone Fynbos, South Langeberg Sandstone Fynbos, Western Altimontane Sandstone Fynbos and Western Little Karoo (FFh 4, FFs 7, FFs 13, FFs 14, FFs 16, FFs 30 and SKv 8) (Rebelo *et al.* 2006; Rutherford *et al.* 2006).

4.6.2.4.3. Etymology

The specific epithet *semota* is derived from the Latin adjective *semotus –a -um* (a. remote, distant, and b. distinct from, similar to) and alludes to both its rare and remote occurrence and its distinctness from and similarity to *P. affinis*.

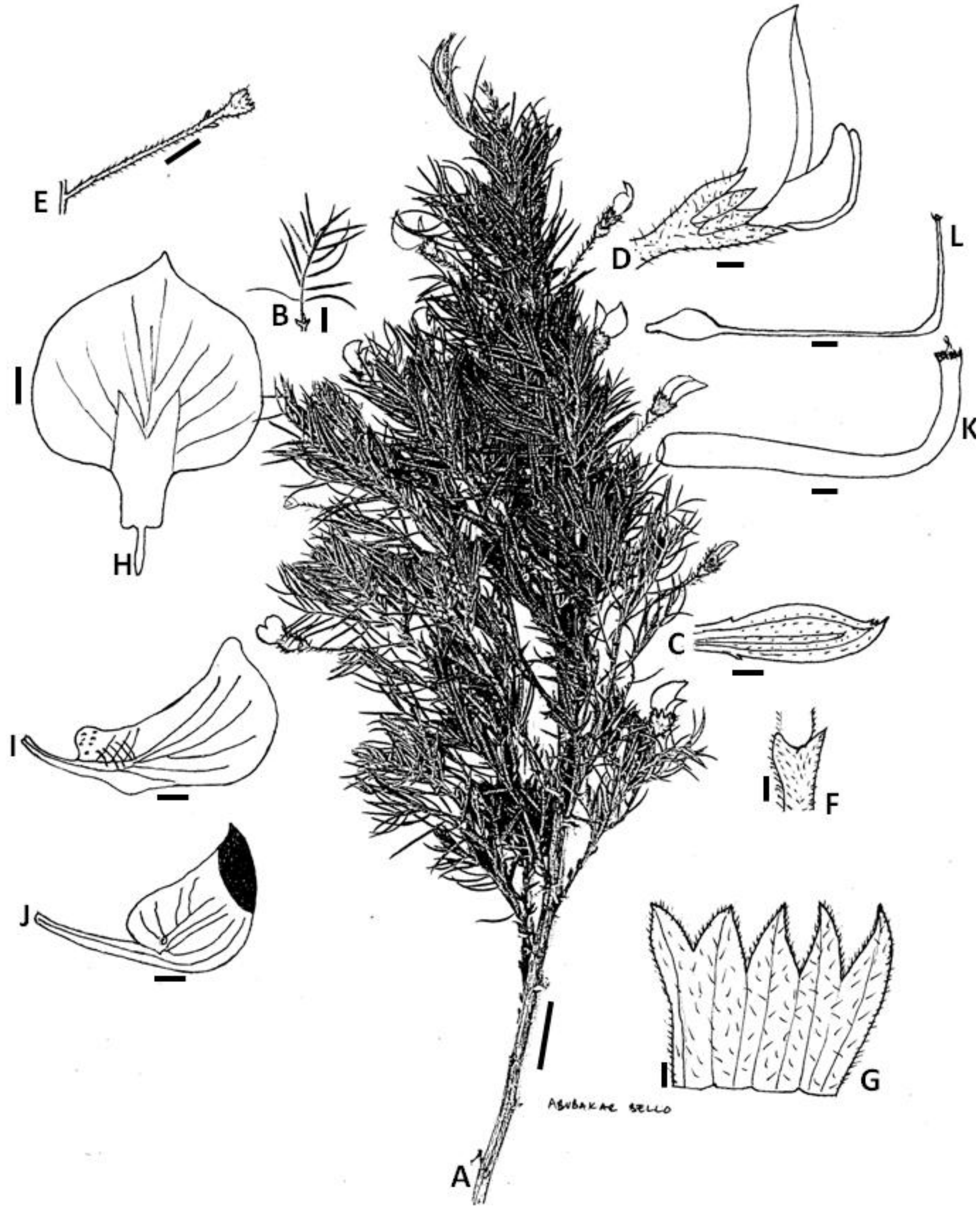


Figure 9. Vegetative and reproductive morphology of *P. semota* (A) flowering shoot; (B) 13-foliolate leaf (C) bud; (D) flower; (E) peduncle; (F) cupulum; (G) calyx opened out with vexillary lobes to the right; (H) front view of standard petal; (I) wing petal; (J) keel; (K) androecium; (L) pistil. Voucher *E. Esterhuysen 21045* (PRE). Scale bars: A−B, 1 cm; C–L, 1 mm. Artist: Abubakar Bello.

4.6.2.4.4. Conservation status

Psoralea semota is a poorly known and collected species. The known populations are small and localised. It occurs entirely within protected areas at high elevations but given its small populations it is prone to stochastic events such as too frequent fires. It is therefore assessed as vulnerable (VD2) according to IUCN criteria of threat (IUCN 2012a).

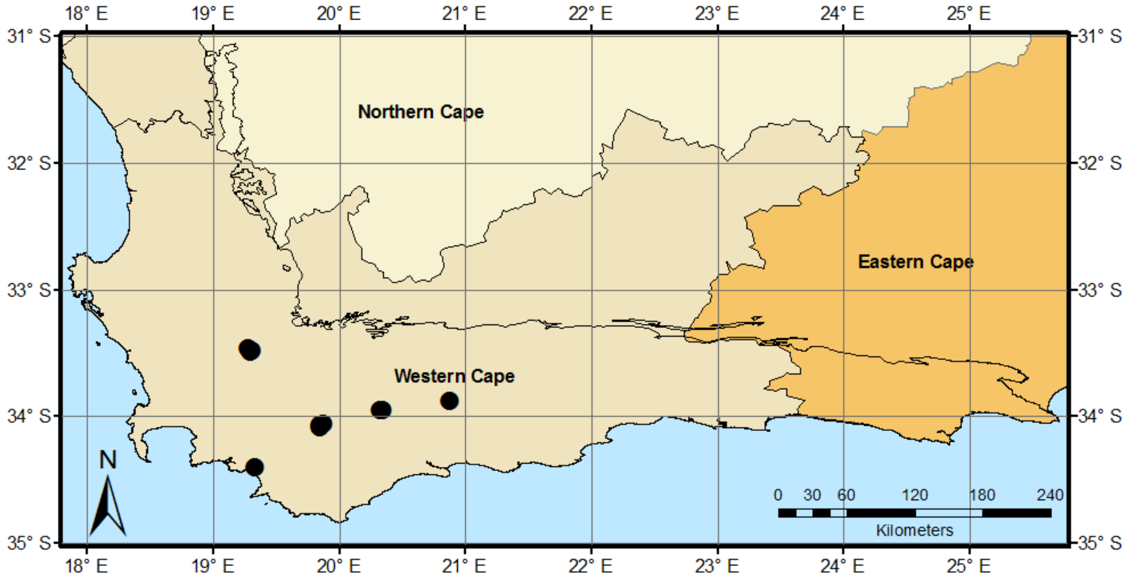


Figure 10. Known distribution of *P. semota*.

4.6.2.4.5. Specimens studied

South Africa. WESTERN CAPE: **3319 (Worcester)**: Mosterts Hoek Twins, Worcester, South western region (-AD), 8 Jan 1944, *Esterhuysen E. 9817* (BOL); Streamside, Waaihoek Peak (-AD), 24 Jan 1949, *Esterhuysen E. 15131* (BOL); Western slopes of Waaihoek Peak (-AD), 13 Jan 1954, *Esterhuysen E. 22631* (K); 13 Feb 1981, *35634* (K, PRE); 13 Feb 1981, *Esterhuysen E. 22631* (BOL). **3320 (Montagu)**: Upper southern slopes, half way between Protea Valley and Leeurivierberg, Vensterbank, Swellendam, Langeberg (-CD), 15 Jan 2006, *Helme N.A. 3802* (NBG); Upper South East slopes of Leeurivierberg, Swellendam, Langeberg (-CD), 15 Jan 2006, *Helme N.A. 3803* (NBG); Saddle at western extremity of Repeaterkop ridge, Grootvadersbosch State Forest, Langeberg (-DD), 27 Nov 1987, *McDonald D.J. 1514* (NBG). **3419 (Caledon)**: Slopes of Mosterthoek Peak, above Waboomsrivier (-AD), 18 Dec 1985, *Goldblatt P. 7482* (PRE); Schilpadkop, Riviersonderend Mountains (-BA), 2 Jun 1953, *Esterhuysen E. 21045* (BOL, PRE); On old scree slope below ridge, west side of Pilaarkop Peak (-BA), 17 Nov 1965, *31409* (BOL); Pilaarkop, Riviersonderend Mountains, near Lindeshof (-BA), 21 Nov 1981, *35708* (K, PRE); Along Die Rif Path, about 2 km South East of Happy Valley Peak, 10 km East of Greyton (-BA), 7 Nov 2001, *Helme N.A. 2323* (NBG).

4.6.2.5. *Psoralea laevigata* L.f., Suppl. Pl.:339 (1782); Poir. in Lam., Encycl. 5: 691 (1804); DC., Prodr. 216 (1825); G. Don, Gen. hist. 2: 201 (1832); Eckl. & Zeyh., Enum.: 224 (1836); Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012); *Psoralea laevigata* sensu Sesse & Moc., Pl. Nov. Hisp.: 120 (1889). Type: South Africa, Western Cape "Habitat ad Cap. Bonae Spei", *Thunberg* s.n. (Not found). Neotype: In humidis (altit. III). Montagu (3320): Prope "Puspasvalley" (Swellendam), (-CD), Oct, *Ecklon C. F. & Zeyher K. L. P. 1518* (MO!, neo.; OXF!, PRC!, isoneo.). [Note: After repeated searches for the type of this species, which is easily distinguished from the protologue, we have chosen an Ecklon & Zeyher collection to act as Neotype].

An ascending to erect suffrutex up to 1 m tall and 2(3) m wide; resprouter, forming dense clumps. *Stems* ascending to erect, many, branching at or near the base. *Seasonal shoots* smooth, slippery, glabrous, shiny, green. *Leaves* 5(7)-foliolate, imparipinnate, distributed along the branches, patent, petiolate; leaflets 7–15×0.4–0.6 mm, linear lanceolate, stiff, claw like, upper surface glabrous and shiny, lower surface glabrous, terminal leaflet shorter, basal pair longest, three uppermost digitately inserted; petioles 3–4 mm long, shorter than terminal leaflets; rachis 5−10 mm long; stipules 2–3×1.2–1.5 mm, persistent, subulate, linear, glabrous, shorter than the petiole. *Inflorescences* axillary, borne along the length of seasonal shoots without leafy extension, pseudo-spicate, lax, 1–3 flowers per axil, pedunculate, pedicellate; peduncles stout and rigid, 23–25 mm long, longer than the subtending leaf; cupulum bilobed, glabrous, near the apex, lobes unequally developed, one of the lobes scarcely developed, triangular; *Flowers* 10−12 mm long, deep purple; pedicels 2–4 mm long. *Calyx* 5−6 mm long, tube 2–4×3.5 mm, ribbed, purple toward the lobes, veins purplish; lobes equally developed, shorter than the calyx tube, glabrous, lobes narrowly triangular, acute, densely glandular. *Standard petal* 9–10×13–15 mm, very broadly ovate, purple with large deep purple nectar flash at the crotch above the well-developed appendages, veins darker than blade; claw 3–4 mm long. *Wing petals* 9–11×3–4 mm, same colour as the standard but white towards the base; claw 3–4 mm long. *Keel* 9–10×2–3 mm; whitish with purple patch on inner apex; claw 4–5 mm long. *Androecium* 9 mm long, tenth stamen free, fenestrate, nectarial ring present, 0.3 mm high. *Pistil* 9 mm long, ovary 1.5 mm long, stipitate, glabrous but sparsely covered near distal end in curved stalked glands, style 7 mm long straight, then curved upwards, thickened at point of flexure, height of curvature 2 mm, stigma penicillate. *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* solitary, 2–4 x 2 mm, dark brown to black (Figure 11).

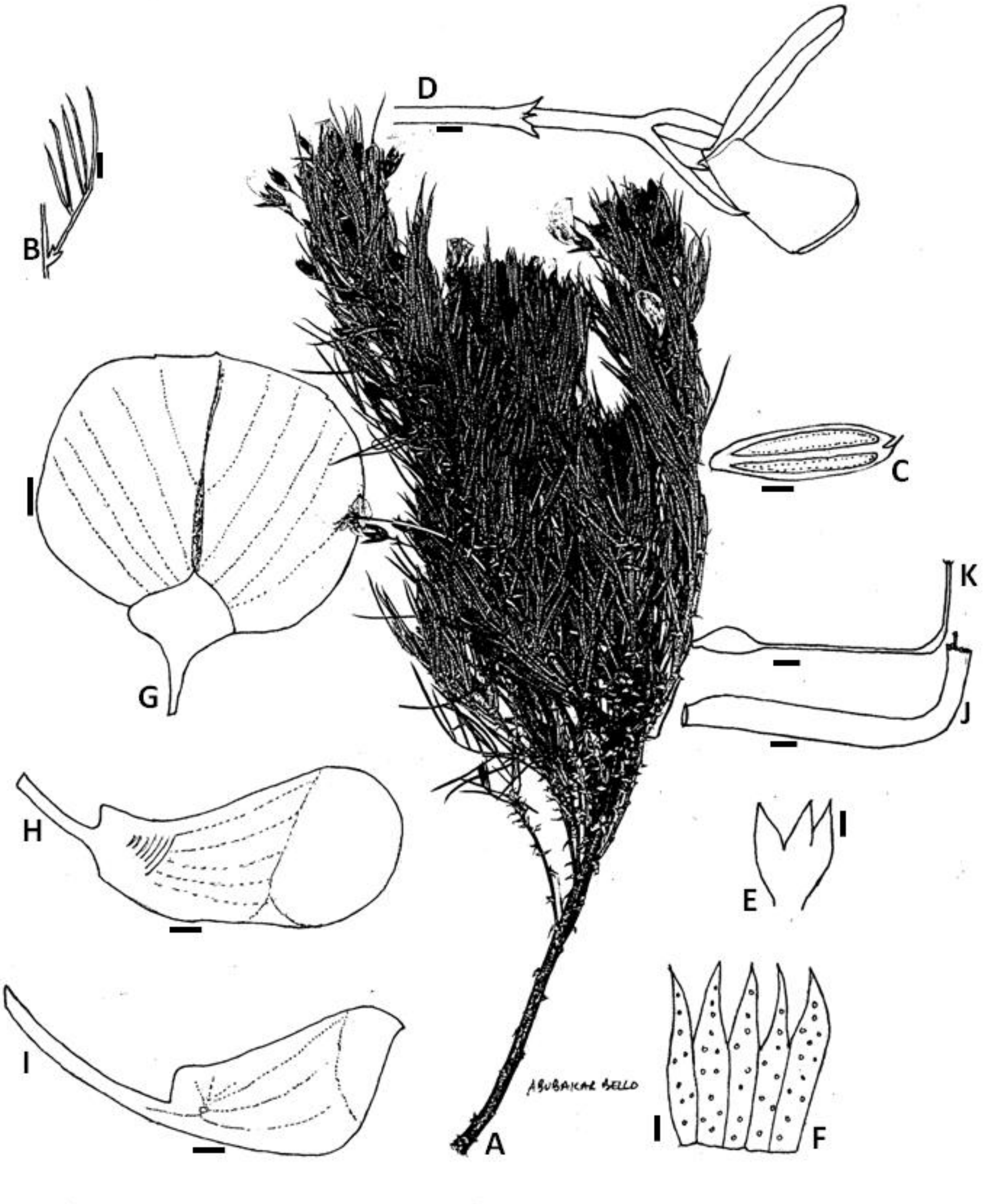


Figure 11. Vegetative and reproductive morphology of *P. laevigata* (A) flowering shoot; (B) 5-foliolate claw like leaf; (C) bud; (D) flower; (E) cupulum; (F) calyx opened out with vexillary lobe to the right; (G) front view of standard petal; (H) wing petal; (I) keel; (J) androecium; (K) pistil. Voucher *A. Bello 119* (BOL). Scale bars: A, 1 cm; B–K, 1 mm. Artist: Abubakar Bello.

4.6.2.5.1. Diagnostic characters

Psoralea laevigata is superficially similar to *P. azuroides* and shares its short 5-foliolate, leaflets. It differs however in its ascending to erect stems, growing up to 1×2(3) m (versus smaller erect plants 0.2−0.8×1 m); blackish stems and branches (versus brown with white lenticels); 7–15×0.4–0.6 mm, (versus 15–22×0.3–0.7 mm), subulate to linear, 2–3 mm long stipules (versus recurved narrowly triangular, 1–2 mm long stipules); inflorescences borne along flowering shoots (versus inflorescences in upper parts of flowering shoots); 1–3 flowers per axil (versus 1 flower per axil); deep purple standard with a darker central flash above the appendages (versus mauve to deep azure blue standard with a large white M-shaped patch above the appendages); irregularly lobed cupulum situated at the apex of a 23–25 mm long peduncle (versus a bilobed cupulum with adaxial cleft and situated at the apex of a 13–14 mm long peduncle); and glabrous margins of calyx lobes (versus black ciliate margins).

4.6.2.5.2. Distribution and habitat

Psoralea laevigata is a montane species and occurs on the lower to mid southern slopes and plateaus in mountain fynbos at elevations between 200−1000 m. It is restricted to the southern slopes of the Langeberg Mountains and is not sympatric with *P. azuroides* (Figure 12). It appears to be restricted to South Outeniqua Sandstone Fynbos (FFs 16) (Mucina *et al.* 2006a; Rebelo *et al.* 2006).

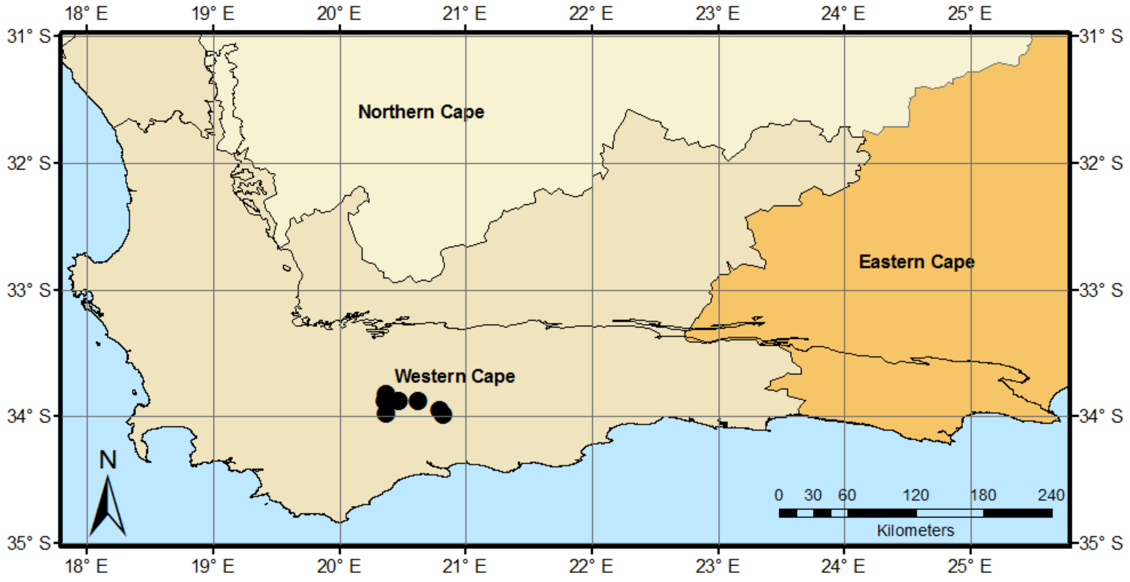


Figure 12. Known distribution of *P. laevigata*.

4.6.2.5.3. Etymology

The specific epithet *laevigata* is derived from the Latin adjective *laevigatus –a –um* (having a smooth or slippery surface) and is an appropriate descriptor of the leaves and stems of this species.

4.6.2.5.4. Conservation status

Psoralea laevigata has been evaluated as Rare (von Staden *et al.* 2009) due to its low density and occurrence as occasional individuals and small colonies. It has protection over most of its distribution range and the best explored localities are the Marloth Nature and Swellendam Forest Reserves but populations are small. This status Rare is retained here as the conditions have not changed. Although striking in stature on emerging early after fire, the species is rarely recorded or collected in the mature state as it is very difficult to distinguish in mature fynbos. The leaves of coppicing plants are large and bright green whereas leaves on old woody plants are much reduced, darker green and more rigid. Young and old plants look very different.

4.6.2.5.5. Specimens studied

South Africa. WESTERN CAPE: **3320 (Montagu)**: Swellendam, in the kloof, above the town (-CD), 1 Apr 1926, *Smith C.A. 2740* (PRE); Swellendam Hillside (-CD), Jan 1938, *Thorns F.W. s.n.* (NBG); Marloth Nature Reserve, Swellendam (-CD), 17 Jun 1969, *Haynes R.A. H157* (PRE, NBG); 21 Jan 1963, *Merwe P.V.D. 1321* (PRE, NBG); path to Twelve o'clock Peak, Marloth Reserve (-CD), 4 Dec 1987, *Goldblatt P. 8704* (PRE); Swellendam Forest Reserve, southern slopes of Langeberg (-CD), 27 Jan 1969, *Marsh J.A. 1105* (PRE, NBG); near River, Wamakersbos, Swellendam Forestry Station (-DC), 31 Oct 1980, *Van Wyk C.M. 339* (NBG); slopes of the Dwarsberg, Boosmansbos Wilderness Area (-DC), 13 Dec 2007, *Muasya A.M. & Stirton C.H.* 3457 (BOL).

4.6.2.6. *Psoralea abbottii* C.H.Stirt. in Kew Bulletin, 50, 3: 619 (1995). Type: South Africa, KwaZulu-Natal, Port Edward (3130): Umtamvuna Nature Reserve, Vultures Nek (-AA), 26 Dec 1985, *Abbott A. 2895* (NU!, holo.; NH!, PCE!, PRU!, iso.).

Decumbent resprouting suffrutex. *Stems* 1– many, trailing, flexuous, branching at or near the base, up to 1.5 m long. *Seasonal shoots* dark green, smooth, glabrous. *Leaves* (3) 5−7-foliolate, petiolate, stipulate, patent or spreading, distributed along the branches; leaflets 15–25×0.3−0.5 mm, linear, smooth, glabrous, dull dark green, terminal leaflet shortest, basal pair longest, three uppermost digitately inserted; petioles 8–8.3 mm long, shorter than terminal leaflet; rachis 10–15 mm; stipules 1.5–2.5 mm long, persistent, glabrous, setose, patent. *Inflorescences* axillary, borne along the length of seasonal shoots without leafy extension, one-flowered, lax, pedunculate, pedicellate; peduncle 14–15 mm long, subtended by two small free unequal bracts; cupulum terminal on peduncle, trilobed, 1.5–2 mm long, lobes sub-equal, adaxial largest, all acuminate, internal bracts present, solitary, 0.8 mm long; pedicel 4–5 mm long. *Flowers* 13–14 mm long, mauve with dark purple. *Calyx* 6–7 mm long, tube 3.5–4×4 mm; lobes more-or-less equal, shorter than the tube, sometimes tinged purple, triangular, carinal slightly broader, vexillary lobes slightly fused beyond the tube, inner face black, densely pubescent, glabrous outside, shiny. *Standard petal* 11–12×11–12 mm, very broadly ovate, apex truncate, slightly auriculate; mauve with a dark purple nectar guide above the small appendages; claw 4–5 mm long. *Wing petals* 13–14×3 mm, cultrate and upcurving; claw 4–5 mm long, much longer but slightly narrower than keel, auriculate, sculpturing present, upper basal, comprised of 8–12 transcostal arches. *Keel* 9–10×3–5 mm, apex acuminate; claw 5 mm long. *Androecium* 9 mm long, sheath split adaxially, tenth stamen lightly adherent to the right hand base of the sheath margin, fenestrate. *Pistil* 9 mm long; ovary 1.5 mm long, stipitate; style 6 mm long, straight, then up curved at end, point of flexure thickened, height of curvature 3 mm long; stigma capitate, penicillate. *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* solitary, 2–4 mm long, dark brown to black (for illustrations see Stirton, 1995−Figure 1).

4.6.2.6.1. Diagnostic characters

Psoralea abbottii is unique in *Psoralea* being the only pinnate-leaved species with a flexuous sprawling habit. This, together with its long peduncles (14–15 mm long), 1-flowered inflorescences, attenuated flower buds and acuminate cupulum, sets it apart from other species like *P. glabra* to which it is similar in flower colour. *Psoralea glabra* by contrast, is an erect woody shrub up to 3 m tall (versus suffrutex non-woody up to 1.5 m), 1–3-flowered inflorescences, short flower buds, and acute cupulum (versus 1-flowered, long flower buds).

4.6.2.6.2. Distribution and habitat

Psoralea abbottii grows along the margins of swamp forest, and in open grassland adjacent to marshes draining into swamp forest. It is distributed from Engcobo in the Eastern Cape Province northwards to Weza and the Ingeli Mountains in KwaZulu-Natal Province (Figure 13). It occurs in Midlands Mistbelt Grassland, Drakensberg Foothill Moist Grassland, KwaZulu-Natal Coastal Belt, Pondoland-Ugu Sandstone Coastal Sourveld and Bhisho Thornveld vegetation types (CB 3, CB 4, GS 6, GS 10 and SVs 7) (Mucina *et al.* 2006b, Mucina *et al.* 2006c, Rutherford *et al.* 2006).

4.6.2.6.3. Etymology

The specific epithet *abbottii* honours the late Mr Tony Abbott in recognition of his significant contributions to the conservation of the Pondoland flora, his passion for hunting down unusual plants, and also for founding Plant Life, a Magazine for amateur botanists and horticulturalists. He also founded the Umtamvuna River Trust, which helped to establish the Ngeli Nature Reserve that was threatened by bauxite mining, and worked closely on conservation with traditional healers in his region.

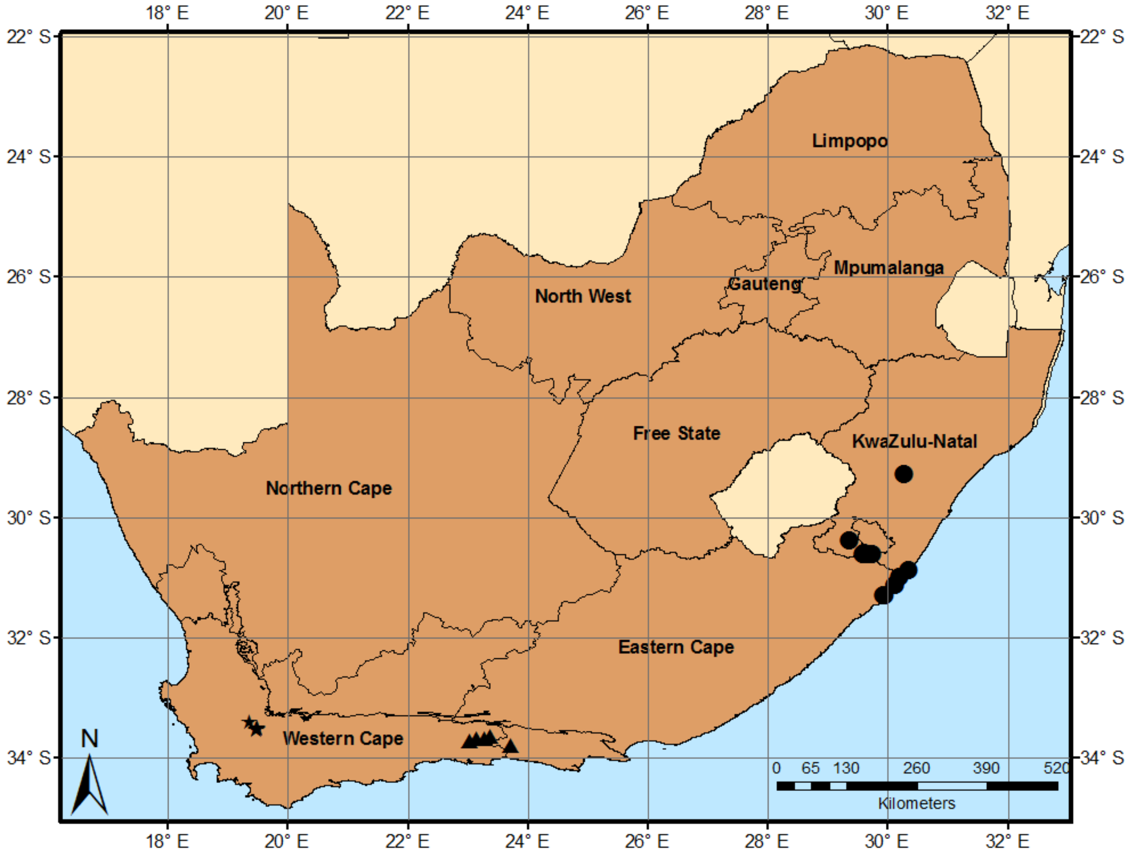


Figure 13. Known distribution of *P. abbottii* (circles), *P. suaveolens* (stars) and *P. kougaensis* (triangles).

4.6.2.6.4. Conservation status

Psoralea abbottii is a rare, cryptic and little collected species as it is very difficult to find when it is not in flower. It is known from fewer than 10 known localities (Extent of Occurrence (EOO) below 8000 km^2) and its population is declining because of expanding crop cultivation, too frequent fires, and overgrazing (Victor and van Wyk, 2005). These impacts still pertain. It is red listed as vulnerable (VU B1 ab (i, ii, iii, iv, v) according to the South African Red data list (version 2014.1; Victor & Van Wyk in Raimondo *et al.* 2009).

4.6.2.6.5. Specimens studied

South Africa. KWAZULU-NATAL: **2930 (Pietermaritzburg)**: Gilboa Karkloof mountains (-AD), 12 Feb 1986, *Edwards T. 110* (PRE); Cunningham's Castle, above Byrne village (-CC), 15 Jan 1985, *Nicholas A.* and *Neave D. 2100* (PRE). **3029 (Kokstad)**: Eastern slope of

Ngeli Mountains (-BD), *Whitworth J.H. s.n* (NBG); Mar 1972, *Nicholson H. 1222* (PRE); 5 Mar, *Strey 10921* (K, NH); 1 km west of Kwa Shwili, Ngeli Nature Reserve (-DA), 24 Nov 1994, *Williams R. 1193* (PRE); 1883, *Tyson 448* (K); Ngeli Mountains foot hills, Weza State Forest (-DA), 19 Nov 1986, *Van Wyk A.E. 7482* (K, PCE, PRE, PRU); Weza Forest, Alfred District (-DA), 2 Jan 1957, *Taylor H.C. 5305* (NBG); 2 Jan 1966, *Strey R.G. 6366* (K, PRE). **3030 (Port Edward)**: Western Heights, Umtamvuna Nature Reserve (-AA), 12 May 1983, *Abbott A. 1110* (NH, PCE, PRU); Umtamvuna Nature Reserve (-AA), 26 Apr 1984, *Abbott A. 1930* (NH, PCE, PRU); 13 Jan 1990, *Abbott A. 4921* (NH, NU, PCE, PRE, PRU); 26 Jan 1986, *Abbott A. 8841.1* (PCE); Glenlyons, Oribi Gorge (-AA), 10 Oct 2011, *Muasya A.M. & Stirton C.H. 6151* (BOL); Vulture's Nek (-AA), 26 Dec 1985, *Abbott A. 2895* (K, NU); 19 Mar 2006, *Abbott A. 8396* (PCE, PRU); Palm Beach, 12 km north of Port Edward (-CD), 5 Jan 1981, *Germishuizen G. 1756* (PRE). EASTERN CAPE: **3127 (Lady Frere):** Hills near Encobo (-DC), 13 Oct 1961, *Esterhuysen E.* (BOL, K). **3129 (Port St. Johns)**: Along first stream, west of entrance road, Mkambati Nature Reserve (-BD), 4 Mar 2007, *Helme N.A. 4588* (NBG); 26 Jan 1986, *Stirton C.H. 11147* (K).

4.6.2.7. *Psoralea suaveolens* C.H.Stirt., A. Bello & Muasya sp. nov. Type: South Africa, Western Cape, Worcester (3319): Citrus Farm Reservoir, Kanetvlei, near De Doorns, Hex River Mountains (-AD), 26 Dec 2007, *Muasya A.M. & Stirton C.H. 3531*(BOL, holo!.; PRE!, iso.).

Slender shrub up to 2 m tall, colonial, resprouter. *Stems* single, erect, young stems greenish yellow with storied and scattered white lenticels, later turning brown with a rough striated surface; short lateral branches numerous at seedling and sapling stages but wither away with strong branches forming mainly in the upper portions; plants strongly scented. *Seasonal shoots* glabrescent, green with purple striae. *Leaves* 13−15(17)-foliolate, imparipinnate, distributed along the branches, patent, petiolate, grooved; leaflets 13–20×0.5–1.0 mm, linear (saplings with narrowly lanceolate leaflets), apiculate, terminal leaflet shortest, basal pair longest, three uppermost pinnately inserted; petioles 5–7 mm long, shorter than terminal leaflet; rachis 20−25 mm long; stipules 4–5×0.5–1 mm, persistent, shorter than petiole, free, glabrous, rapidly senescent, ovate-lanceolate, acuminate, spreading then recurving. *Inflorescences* axillary, borne along the length of seasonal shoots, pseudo-spicate, lax, 1-flower per axil, pedunculate, pedicellate; peduncles filiform and flexuous, 30–50 mm long, exerted from leaves; cupulum bilobed, 1 mm long, lobes equally developed, setose, scarious; pedicels 3–4 mm long. *Flowers* 12−14 mm long, mauve and white. *Calyx* 5−6 mm long, tube

4–5×4 mm, glabrous or with sparse white hairs, yellowish green, smooth, shiny, purple at abaxial part of the margins; lobes sub-equally developed, glabrescent, longer than the tube, lower lobe slightly longer, lateral and vexillar lobes acute, falcate, narrowly triangular, lower lobe acute, sparsely glandular, glands constant in size. *Standard petal* 10–11×8–9 mm, broadly elliptic, pale mauve in upper part fading to white in lower part and sides, veins darker in front, back purplish; claw 3–4 mm long. *Wing petals* 8–10×3–4 mm, white, held erect exposing keel; sculpturing present, upper surface comprised of 3−4 rows of 9−10 transcostal lamellae near the auricle; claw 2–3 mm long. *Keel* 5–6×3–4 mm, white, suffused on inner face apically with dark violet-purple patch; claw 3–4 mm long. *Androecium* 8 mm long, tenth stamen lightly attached, fenestrate. *Pistil* 8 mm long, ovary 2 mm, style straight, 4 mm long then upcurved, thickened at the point of flexure, height of curvature 3.5 mm long. *Fruits* oblong, 4–5×2.5–3.0 mm wide. *Seeds* solitary, 2–4×2 mm, dark brown to black (Figure 14).

4.6.2.7.1. Diagnostic characters

Similar to *P. odoratissima* in that they have the highest number of leaflets in the genus (9−15(17)-foliolate) and both have strongly scented flowers, and long rachises and petioles but differs in being a slender single-stemmed shrub to 2 m tall (versus large shrub to small tree 4–6 m tall); with trunk usually less than 10 cm in diameter (versus trunk 10−20 cm in diameter); seasonal shoots glabrescent (versus covered in lax long white hairs); 9−15(17)-foliolate leaves (versus(13)15–19-foliolate), narrower, 0.5–1.0 mm wide leaflets (versus 0.5–2.0 mm wide); basal pair of leaflets longest (versus basal pair same length as laterals); peduncle 30–50 mm long, (versus peduncle 3–6 mm long), and flowers mauve and white, exerted beyond subtending leaves (versus flowers white and same length as subtending leaves). *Psoralea suaveolens* and *P. odoratissima* form a natural group and are distinct from the other species in the *P. pinnata* complex.

4.6.2.7.2. Distribution and habitat

Psoralea suaveolens is a rare species known only from the lower valleys of Hex River Mountains in the Worcester region of the Western Cape Province (Figure 13). It occurs in seepages and along streamsides in mountain fynbos at 450–1000 m elevation. It is restricted to the South Hex Sandstone Fynbos vegetation type (FFs 8) (Mucina *et al.* 2006a). It produces occasional spontaneous hybrids with *P. usitata* in the lower mountain valleys of the Kanetvlei area. The flowers are visited by small Megachilid bees.

4.6.2.7.3. Etymology

The specific epithet is derived from the Latin word *suaveolens* (fragrant, odorous, sweet-smelling) and alludes to the sweet-smelling flowers.

4.6.2.7.4. Conservation status

Psoralea suaveolens is known from only a few, more remote and undisturbed stream and riverside localities in the De Doorns region and Hugosberg. Most of these are adjacent to cultivated vineyards and citrus plantations and are threatened by regular flash flooding and invasive species (*Acacia longifolia* (Andrews) Willd. and *Sesbania punicea* (Cav.) Benth.). The southern slopes of the Hex River Mountains are still poorly explored so other populations may still be discovered. It is highly likely though that its former distribution was destroyed mostly by intensive agriculture in the Hex River valley. Based on the IUCN categories (IUCN, 2012a, 2012b), we assess it as near threatened (NT D2).

4.6.2.7.5. Specimens studied

South Africa. WESTERN CAPE: **3319 (Worcester)**: Citrus Farm Reservoir, Kanetvlei, near De Doorns, Hex River Mountains (-AD), 26 Mar 2009, *Muasya A.M. & Stirton C.H. 4477* (BOL); 28 Nov 2009, *Muasya A.M. & Stirton C.H. 4975* (BOL); 21 Feb 2011, *Muasya A.M. & Stirton C.H. 5935* (BOL); Zandrifskloof, Hex River Mountains (-AD), 11 Oct 2009, *Muasya A.M. & Stirton C.H. 4074* (BOL); 28 Nov 2009, *Muasya A.M. & Stirton C.H. 4987* (BOL); Near upper reservoir, Kanetvlei, Hex River mountains (-AD), 22 Sep 2010, *Muasya A.M. & Stirton C.H. 5511* (BOL); 29 Feb 2009, *Muasya A.M. & Stirton C.H. 5929* (BOL); Western slopes of Middagkransberge (-CC), 3 Dec 1981, *Stirton C.H. 10094* (K); 3 Dec 1981, *Stirton C.H.* 10095 (K).

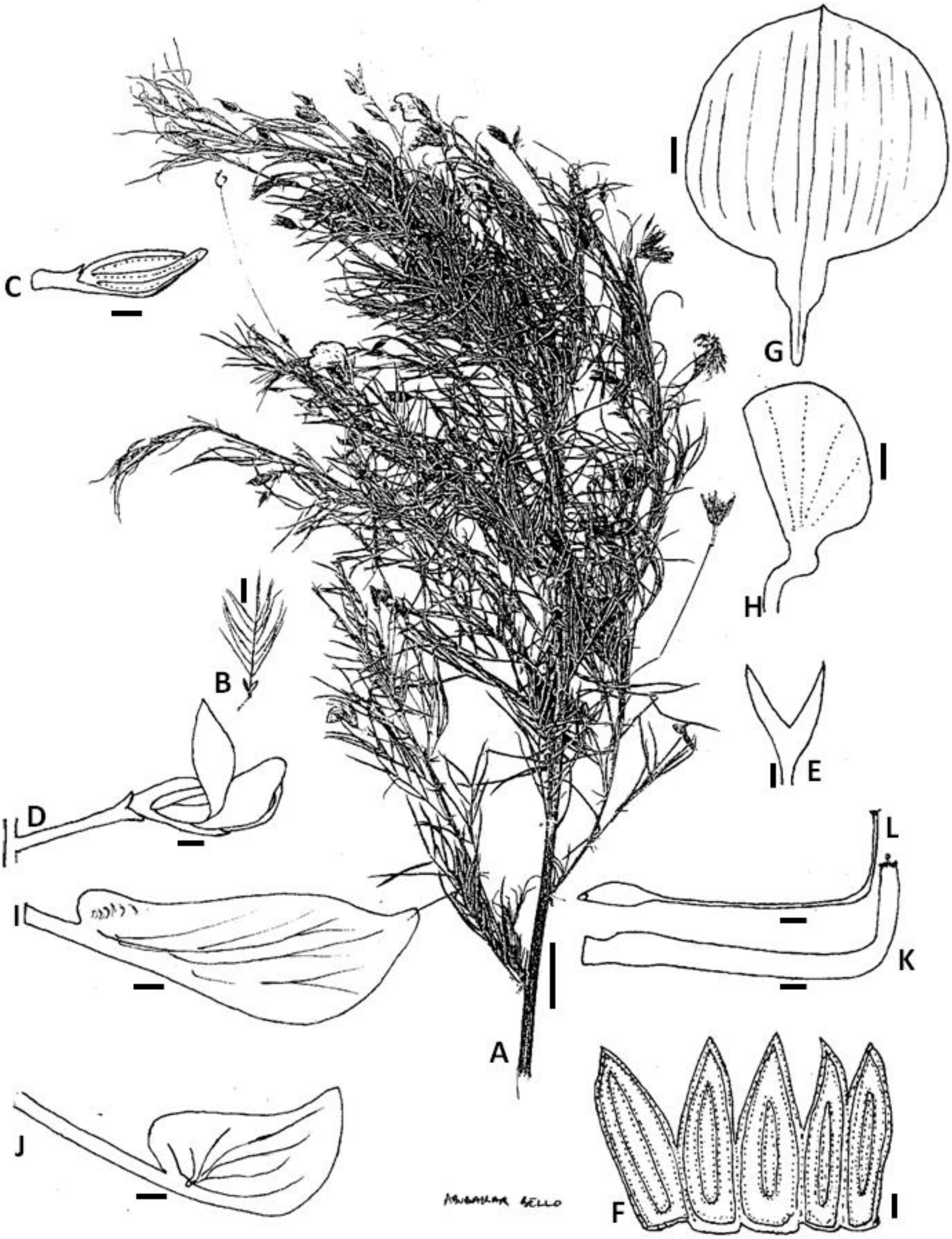


Figure 14. Vegetative and reproductive morphology of *P. suaveolens* (A) flowering shoot; (B) 5-foliolate claw like leaf; (C) bud; (D) flower; (E) cupulum; (F) calyx opened out with vexillary lobes to the right; (G) front view of standard petal; (H) side view of standard petal; (I) wing petal; (J) keel; (K) androecium; (L) pistil. Voucher *A.M. Muasya & C.H. Stirton 5511* (BOL). Scale bars: A, 1 cm; B–L, 1 mm. Artist: Abubakar Bello.

4.6.2.8. *Psoralea kougaensis* C.H. Stirt., Muasya & A. Bello *sp. nov.* Type: South Africa, Western Cape, Willowmore (3323): Uniondale, ridge north side of Tsitsikamma mountains, near Joubertina (-DB), Mar 1954, *Esterhuysen E. 22821* (K!, holo., BOL!, WAG!; iso.).

Erect columnar, multi-stemmed, unbranched or few-branched shrub up to 3 m tall, resprouter, forming dense clumps. *Stems* erect, many, rigid, usually unbranched except for seasonal shoots which are shed the following season, greyish-brown with fine blackish lattice and fissures; as plants age the older stems produce water shoots from near the base, *Seasonal shoots* greenish black, bristly pubescent, glandular. *Leaves* 5(7)-foliolate, imparipinnate, crowded at the end of bare branches on older stems, or distributed along short branches on young shoots, clasping the shoots, with prominent raised glands half the width of leaf, becoming warty when dried, glabrous to sparsely hairy, petiolate; leaflets 15–30×0.2–0.4 mm, linear lanceolate, terminal leaflet shortest, basal pair of leaflets longest, laterals slightly longer than the terminal, three uppermost pinnately inserted; petioles 2.5–3 mm long; rachis 6−9 mm long, channelled on upper side, rounded on lower side; stipules 3–5×3 mm, persistent, prominent, longer than petiole, narrowly triangular, glabrous, fused for one third their length, stiff, cartilaginous. *Inflorescences* axillary, borne in upper axils of seasonal shoots with leafy extension, pseudo-capitate, congested but fasciculate, 3–5 flowers per axil, pedunculate, pedicel absent; peduncles stout and rigid, 2–3 mm long, shorter than the subtending leaf; cupulum trilobed, glabrous or bristly pubescent with black hairs, lobes triangular, 1−1.5 mm long. *Flowers* 12−13 mm long, white to mauve, hidden within foliage. *Calyx* 7−8 mm long, tube 4–6×4 mm; lobes equally developed, shorter than the calyx tube, glabrous or sparsely pubescent, carinal lobe slightly wider and less hairy, densely glandular, glands constant in size. *Standard petal* 9–11×8.5–9 mm, very broadly ovate, dark mauve fading towards margins with a broad triangular purple central patch above the swollen appendages from which arises a very dark erect flash tapering to the apex; claw 2–3 mm long. *Wing petals* 8–10×3–3.5 mm, white, tips sometimes pale lavender, sculpturing present, upper basal, lower basal, and upper left central, comprised of 2–3 rows of 10–12 transcostal lamellae; claw 2–3 mm long. *Keel* 7–10×4–5 mm, white but apically suffused with dark violet-purple on inner apex; claw 2–3 mm long. *Androecium* 9 mm long, tenth stamen shortly united with staminal sheath adaxially, scarcely fenestrate. *Pistil* 8 mm long, stipitate, ovary 1 mm long, glabrous, sparsely covered in club-shaped glands, style glabrous, straight for 5 mm then curved upwards, thickened at point of flexure, height of curvature 2 mm; *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* one, 2–4×2 mm, dark brown to black (Figure 15).

4.6.2.8.1. Diagnostic characters

Similar to *P. speciosa* in having large calyces, but differs in being an erect, columnar, multi-stemmed, branchless or few-branched resprouting shrub (versus arching large multi-branched, reseeding shrub), with 5(7)-foliolate, tightly packed, distinctly warty leaves (versus 5-foliolate papillose, more lax and usually with curving leaves), glabrous, narrowly triangular, prominent persistent stipules, (versus puberulent, triangular, large caducous stipules), white to mauve flowers with a large triangular purple central nectar guide above the swollen appendages from which arises a very dark erect flash tapering to the apex (versus deep or indigo-blue flowers with large white M-shaped nectar guide and no central purple flash) and glabrous or sparsely pubescent calyces (versus glabrous but fringed with dark hairs). A distinctive feature of *P. kougaensis* occurs in the fruiting stage when the seeds become exposed prior to shedding because the calyx lobes dry out and splay leaving the seeds sitting exposed on top. This also occurs in the unrelated *P. odoratissima.*

4.6.2.8.2. Distribution and habitat

Psoralea kougaensis is a montane species first discovered in 1975 and extends from the Swartberg Mountains to as far east as the Potjiesberg and Kammanasie mountains in the Western Cape Province (Figure 13). It occurs in Mountain fynbos on dry exposed slopes between 300−830 m. It appears to be restricted to the North Outeniqua Sandstone Fynbos and Kouga Sandstone Fynbos (FFs 26, FFs 27) vegetation types (Rebelo *et al.* 2006).

4.6.2.8.3. Etymology

The specific epithet *kougaensis* refers to the general Kouga region and mountains where the species occurs.

4.6.2.8.4. Conservation status

Psoralea kougaensis is a habitat specialist and does not appear to be under any form of threat. It occurs predominantly in more remote inaccessible areas. In one or two localities, however, it has begun to spread along road verges as a result of road works. It is assessed as Rare in line with the South African criteria of rarity (von Staden *et al.* 2009).

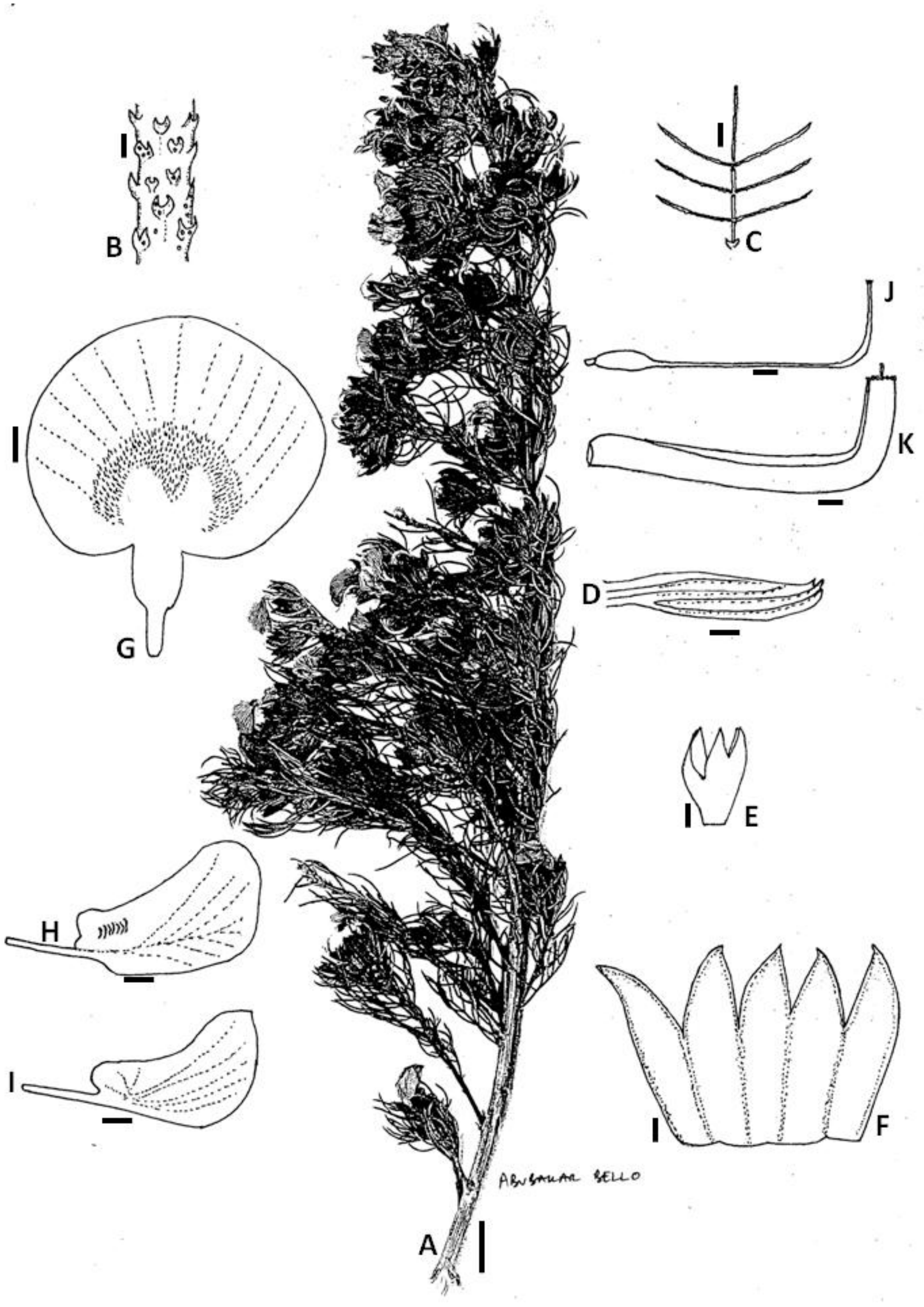


Figure 15. Vegetative and reproductive morphology of *P. kougaensis* (A) flowering shoot; (B) 7-foliolate leaf; (C) bud; (D) flower; (E) cupulum; (F) calyx opened out vexillary lobes to the right; (G) front view of standard petal; (H) wing petal; (I) keel; (J) pistil; (K) androecium. Voucher *M. Dludlu 45* (BOL). Scale bars: A, 1 cm; B–K, 1 mm. Artist: Abubakar Bello.

4.6.2.8.5. Specimens studied

South Africa. WESTERN CAPE: **3322 (Oudtshoorn)**: Swartberg mountains, mid-northern slope of Blesberg, near forestry track (-BC), 15 Dec 1986, *Vlok J.H.J. 1779* (PRE); George, Jonkersberg (-DD), Oct 1926, *Phillips J.F.V. 1084* (PRE); Ratelkop (-DD), Oct 1998, *Baard J. 294* (PRE). **3323 (Willowmore)**: Potjiesberg Pass, 3 km from Uniondale to George (-CA), 20 Dec 2009, *Muasya A.M & Stirton C.H. 5275* (BOL); Uniondale, lower northern slope of Potjiesrivier heights (-CA), 26 Oct 1986, *Vlok J.H.J. 1675* (PRE, NU); slopes of Braamberg/Witberg, on road to Saptou, on De Hoop farm (-CB), 29 Sep 1980, *Snijman D. 356* (PRE, NBG); Donkerhoek se Nek, Prince Alfred's Pass (-DD), 27 Sep 1978, *Hugo L. 1334* (PRE); Joubertina (-DB), 10 Sep 1975, *Manson J.A. 105* (PRE, NBG); Joubertina on R62 Westwards, 13 Km to Xkagodi, on Kouga River (-DD), 19 Sep 2000, *Nienaber E.P. EN907* (PRE). EASTERN CAPE: **3324 (Steytlerville):** Suuranys Pass (-CC), 1 Oct 1984, *Stirton C.H. 10925* (K).

4.6.2.9. *Psoralea muirii* C.H. Stirt. & Muasya *sp. nov.* Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 506 (2000); *Psoralea* sp. 16, Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 574 (2012). Type: South Africa, Western Cape, Riversdale (3421): Aasvogelberg, near Albertinia (-BA), 19 Nov 1914, *Muir J. 1850* (PRE!, holo.; BOL!, iso.).

Robust resprouting shrub up to 1.5(2) m tall. *Stems* many, sub-erect to erect, multi-branched from near the base, rigid, rough, mostly bare except for leafy upper parts, branching at or near the base; young stems green to blackish and finely pubescent, older stems glabrous, grey with dull white storied lenticels giving a striped appearance, sapling shoots diassapear with age; stipule scars prominent. *Seasonal shoots* rough, blackish, hairy. *Leaves* 5−7-foliolate, imparipinnate, crowded at the ends of bare branches, erect, petiolate, with raised glands; leaflets 9–11×0.3–0.4 mm, linear lanceolate, glabrous, densely glandular, raised and prominent, wartlike; terminal leaflet shortest or about the same length as the laterals, basal pair longest, apex of leaflets acute, three uppermost pinnately inserted; petioles 2–3 mm long; rachis 10 mm long; stipules 0.7–2×1–1.5 mm, poorly developed but persistent, shorter than the petiole, glabrous. *Inflorescences* axillary, borne in uppermost 5–9 axils of seasonal shoots, pseudo-capitate, congested, 1 (2) flowers per axil, pedunculate, pedicels absent; peduncles stout and rigid, 3–4 mm long; cupulum trilobed, glabrous, lobes poorly developed. *Flowers* 12−14 mm long, mauve. *Calyx* 5–7 mm long, tube 3–4×3.5 mm; lobes equally developed, shorter than the calyx tube, oblong-ovate, sub-acute or rounded at the apex, margins ciliate; glabrous, with dense glands, variable in size, equally distributed across the tube and lobes; ribs present, distinctly thickened. *Standard petal* 10×9 mm, sub-orbicular, pale to dark mauve with purplish veins and a small central violet nectar guide, auricles well-developed, apex emarginate; claw 2–3 mm long. *Wing petals* 10–14×4 mm, cultrate, mauve; auricles well-developed, sculpturing present, upper basal and upper left central comprised of 7–9 transcostal lamellae; claw 3–4 mm long. *Keel* 11–12×3–4 mm; claw 5–7 mm long. *Androecium* 10 mm long, tenth stamen slightly attached, fenestrate. *Pistil* 11 mm long, ovary 2 mm long, glabrous but with a few small stalked glands; style 6 mm long, straight, then upcurved, swollen beyond point of flexure, height of curvature 3 mm long. *Fruits* oblong, 4–5×2.5–3.0 mm, coarsely reticulate, membranous, black. *Seeds* solitary, 2–4×2 mm, blackish grey (Figure 16).

4.6.2.9.1. Diagnostic characters

Similar to *P. pinnata* in appearance from which it differs in being a robust sub-erect to erect resprouting shrub to 1.5(2) m tall (versus erect reseeding shrub to small tree up to 5 m tall), 5−7-foliolate with linear lanceolate prominently warty leaves (versus 7−9-foliolate linear villoso-pubescent smooth leaves), stipules 0.7−2.0 mm long, glabrous (versus 3−4 mm long, hairy), mauve flowers borne in upper 5−9 axils of seasonal flowering shoots (versus mauve or pale blue flowers borne along the length of seasonal flowering shoots). The coastal species *P. muirii* can be confused when dried with the montane *P. kougaensis* as both have prominent warty leaf glands which, though similar in the fresh state, shrivel differently when dried. They are allopatric.

4.6.2.9.2. Distribution and habitat

Psoralea muirii is a lowland fynbos species occurring mainly between Albertinia and Still Bay and less frequently towards the Gouritz River (Figure 17). It favours limestone soils between 350–400 m elevations. *Psoralea muirii* is found in the Albertinia Sandstone Fynbos, Canca Limestone Fynbos, and Kouga Grassy Sandstone Fynbos vegetation types (FFd 9, FFl 3 and FFs 28) (Rebelo *et al.* 2006).

4.6.2.9.3. Etymology

Psoralea muirii is named in honour the Scottish Physician John Muir (1874–1947) who published the first flora of the Riversdale region (Ph. D. Edinburgh University). He was also an expert on coastal drift seeds and a keen collector of sea shells. He received an Honorary D.Sc. from Stellenbosch University in 1929 for this work. He also published articles on common names of local plants and animals, genealogy and local folklore of the region. He was the first person to collect *P. muirii*.

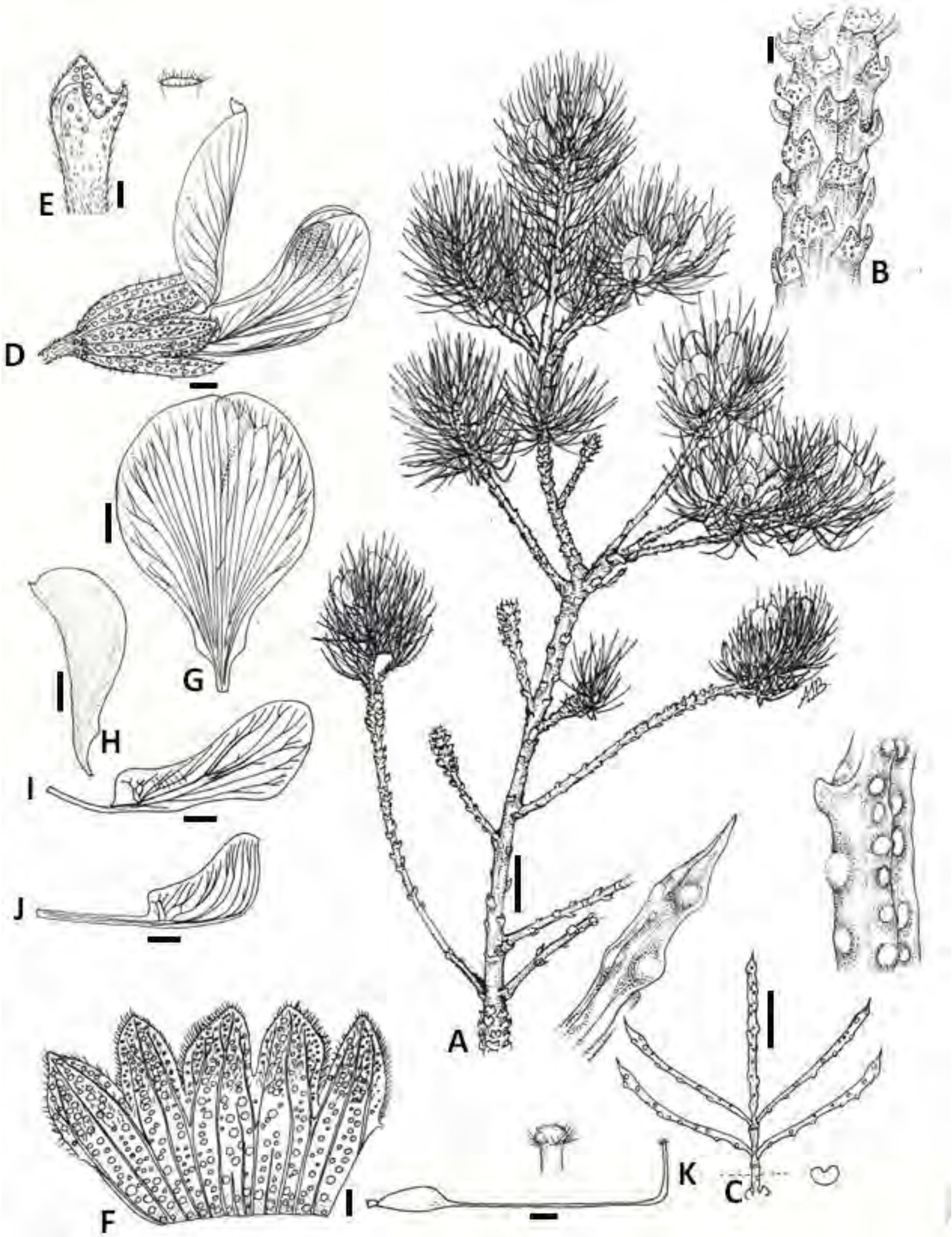


Figure 16. Vegetative and reproductive morphology of *P. muirii* (A) flowering shoot; (B) scales on the stem; (C) 5-foliolate leaf; (D) flower; (E) cupulum; (F) calyx opened out with vexillary lobes to the right; (G) front view of standard petal; (H) side view of standard petal; (I) wing petal; (J) keel; (K) pistil. Voucher *J. Muir 1850* (BOL). Scale bars: A–C, 1 cm; D–K, 1 mm. Artist: Angela Beaumont.

4.6.2.9.4. Conservation status

Psoralea muirii is assessed as endangered (EN B1 ab (iii, v)). Its extent of occurrence (EOO) is 620 km². It is known from only seven locations and is declining due to invasive inroads from Australian Acacias and habitat degradation from bush dragging to prepare fields for harvesting of Restionaceae for thatch (Stirton *et al.* 2008).

4.6.2.9.5. Specimens studied

South Africa. WESTERN CAPE: **3421 (Riversdale)**: Hectorskraal (-AD), 25 Aug 1978, *Hugo L. 1247* (NBG, PRE); 3 km from Stilbaai to Gouritzmond road (-AD), 26 Oct 2008, *Muasya A.M. & Stirton C.H. 4181* (BOL); Between Botterkloof and Blikhuis Farms on Stilbaai to Gouritz road (-AD), 30 Nov 2009, *Muasya A.M. & Stirton C.H. 5031, 5034* (BOL); Aasvogelberg (-BA), 19 Nov 1914, *Muir J. 1850* (BOL, PRE); Turnoff to Rhein's Coastal Nature Reserve on Stilbaai to Gouritzmond road (-CD), 30 Nov 2009, *Muasya A.M. & Stirton C.H. 5036* (BOL); Rhein's Coastal Nature Reserve (-CD), 21 Oct 2014, *Bello A. & Stirton C.H. 143* (BOL).

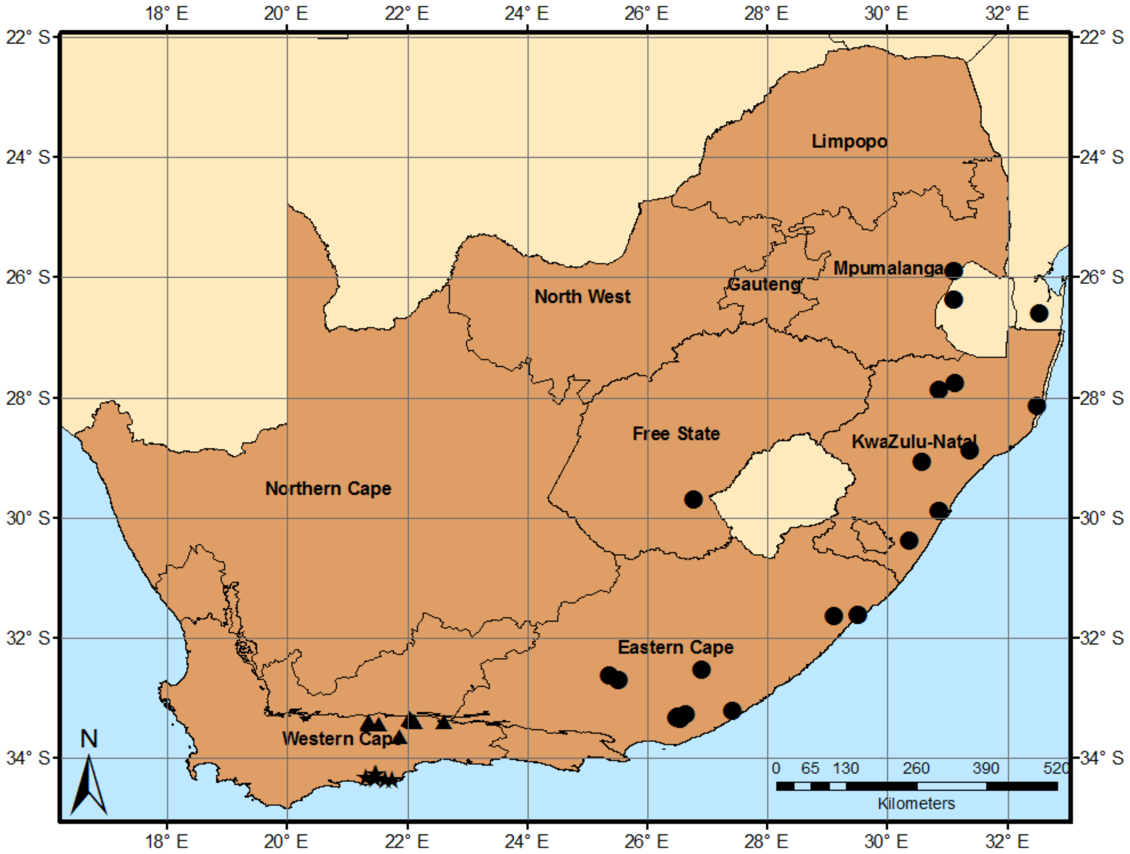


Figure 17. Known distribution of *P. muirii* (stars), *P. sordida* (triangles) and *P. glabra* (circles).

4.6.2.10. *Psoralea sordida* C.H. Stirt. & Muasya *sp. nov.*, Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); *Psoralea* sp. 29, Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Western Cape, Ladismith (3321): Swartberg Mountains (-BD), 11 Nov 1986, *Stirton C.H. & Žantovská 11572* (K!, holo.).

Small much-branched decumbent compact post-fire shrub thereafter reducing with age to 1−2 lanky stems up to 2.5 m tall. *Stems* erect, virgate, branching in upper parts, grey with large scattered white lenticels; branches stiff, erect, brown. *Seasonal shoots* densely canescent, dull dark green becoming blackish, then glabrous with age, broom-like, appressed to main stem. *Leaves* (3)5(7)-foliolate, imparipinnate, stiff, filiform, distributed along the branches, semi-erect to spreading, petiolate, smooth, glabrous, channelled on upper surface; leaflets 20–23×0.2–0.3 mm, linear lanceolate, terminal leaflet longest, basal pair second longest, three uppermost digitately inserted; petioles 4–6 mm long, shorter than terminal leaflets; rachis 5−10 mm long; stipules 3.0−3.5×1 mm, persistent, shorter than petiole, setose, glabrous. *Inflorescences* axillary, borne in most axils of seasonal shoots, pseudo-spicate, lax, 3 flowers per axil, sessile or pedunculate, pedicellate; peduncles stout and rigid, 2–4 mm long; cupulum trilobed, glabrous or with a few hairs along the margins, lobes unequally developed, lower lobe longest; pedicels 1(2) mm long. *Flowers* 11−12 mm long, dirty whitish to pale mauve, each subtended by two separate uneven bracts. *Calyx* 6–7 mm long, tube 3–4×4 mm, glabrous, shiny, green to blackish purple; lobes unequally developed, shorter than the tube, broadly triangular, carinal somewhat narrower, vexillar lobes fused for half their length, inner face with stubby black hairs. *Standard petal* 10×10 mm, broadly ovate, apex truncate, dirty whitish to pale mauve with a prominent splaying central purple nectar guide, venation coloured purple; claw 3–4 mm long. *Wing petals* 10–11×3.5–4.5 mm, mauve; sculpturing present, upper surface comprised of 3−4 rows of 9−10 transcostal lamellae near the auricle; claw 4–5 mm long. *Keel* much shorter than the wing petals, 6–7×3 mm, white to pale mauve, apically suffused with dark violet-purple on inner face; claw 4–5 mm long. *Androecium* 7 mm long, tenth stamen lightly attached adaxially, fenestrate. *Pistil* 7 mm long, stipe 1 mm long, ovary 1.5 mm, style 5 mm long straight then curved, thickened at the point of flexure, height of curvature 3.5 mm long. *Fruits* oblong, 3–4×2.5–3.0 mm. *Seeds* solitary, 2–3×2 mm, dark brown to black (Figure 18).

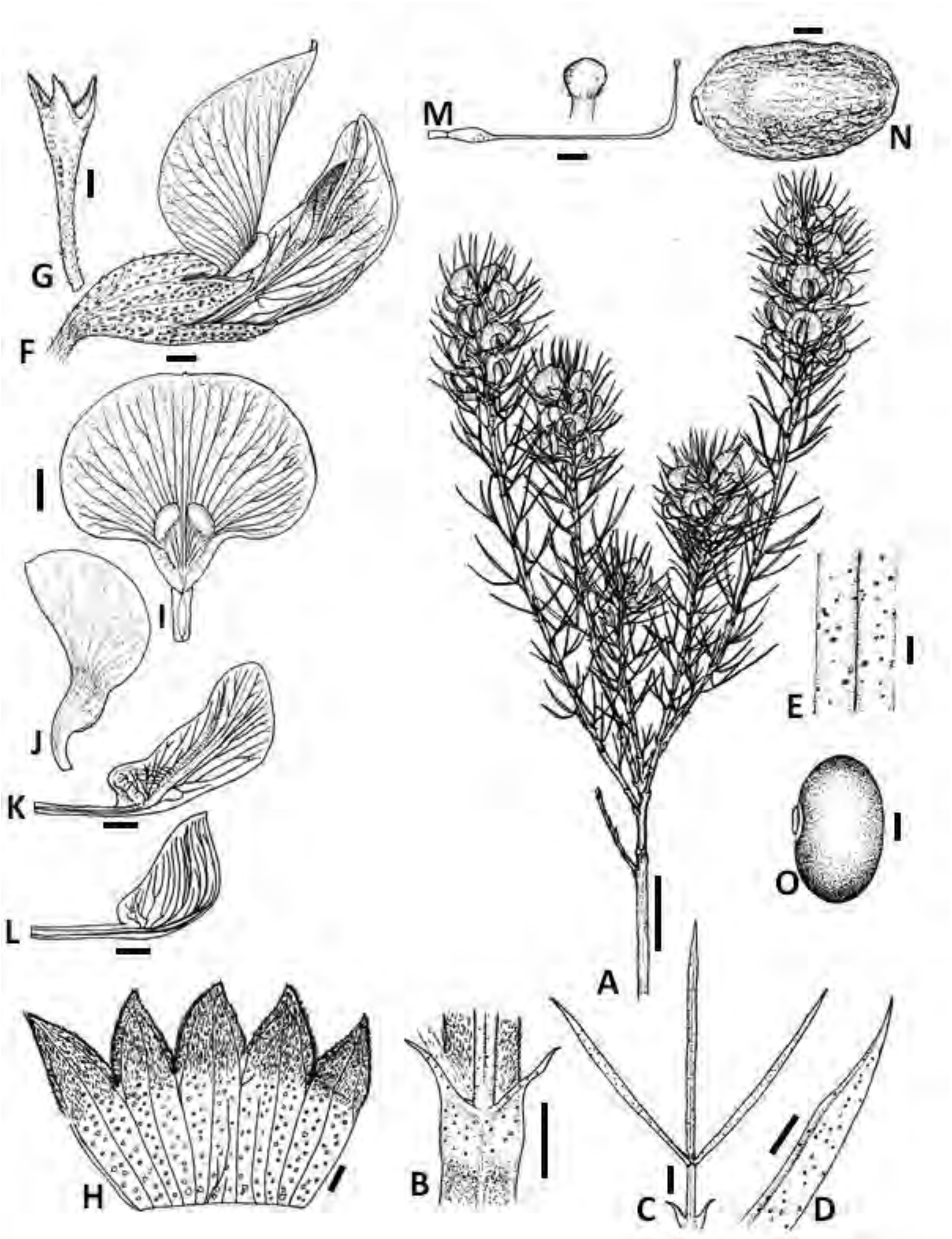


Figure 18. Vegetative and reproductive morphology of *P. sordida* (A) flowering shoot; (B) a twig showing the stipules; (C) 3-foliolate leaf; (D) leaflet; (E); (F) flower; (G) peduncle terminating with a cupulum; (H) calyx opened out with vexillary lobes to the right; (I) front view of standard petal; (J) side view of standard petal; (K) wing petal; (L) keel; (M) pistil; (N) fruit; (O) seed. Voucher *C.H. Stirton & J. Žantovská 11527* (PRE). Scale bars: A, B, D, 1 cm; C, E–O, 1 mm. Artist: Angela Beaumont.

4.6.2.10.1. Diagnostic characters

Psoralea sordida is the only member of the *P. pinnata* complex which exhibits 3-foliolate leaves in the upper parts of flowering shoots. It is similar to *P. pinnata* in having sessile or sub-sessile peduncles, but differs in habit, being initially (post fire) a small much-branched resprouting shrub but reducing with age to 1−2 lank stems up to 2.5 m tall (versus a single-stemmed large much-branched reseeding shrub to small tree up to 5 m tall), with glabrous 3–5(7)-foliolate leaves with terminal leaflet longest (versus villoso-pubescent 7–9-foliolate leaves with the terminal leaflet shortest), whitish to pale mauve standard with a prominent splaying central purple nectar guide and purple veins (versus white with suffused mauve towards the centre and a narrow central purple flash and hyaline veins); and cupulum and calyx glabrous (versus hairy). Like *P. diturnerae* A.Bello, C.H.Stirt & Muasya (Bello *et al.* 2015a), it spends its first year as a compact mounded densely branched shrub after which the lowest branches abscise and 1(2) stems then arise giving the appearance, after a few years, of a tall willowy shrub with many branches each bearing clusters of short seasonal flowering shoots that radiate either terminally from the stem or along older shoots giving the plant a scruffy broom-like appearance. Unusually in the complex it exhibits both reseeding and resprouting modes of regeneration after fire. Seed set is high and seedling recruitment is good but once established plants then coppice regularly after fires. As a result, the species can form large extensive colonies.

4.6.2.10.2. Distribution and habitat

Psoralea sordida is a montane species restricted to the Swartberg, Langeberg and Outeniqua Mountains (Figure 17). It occurs in mountain fynbos mainly on southern slopes at elevations between 460–2000 m. It is recorded from North Swartberg Sandstone Fynbos, South Swartberg Sandstone Fynbos and Uniondale Shale Renosterveld (FFs 23, FFs 24 and FRs 15) (Rebelo *et al.* 2006).

4.6.2.10.3. Etymology

This species is derived from the Latin adjective *sordidus -a -um* (*sordeo*, dirty, filthy) and alludes to its untidy appearance.

4.6.2.10.4. Conservation status

Psoralea sordida forms large colonies and as much of its range is in protected areas it is not threatened. We therefore designate it NT. However, in the Swartberg mountains it forms a large introgressive hybrid swarm with *P. forbesii* (sp. nov. ined.) making it very difficult to accurately identify plants from this region and this needs to be borne in mind when naming plants from the area as many variants occur and persist as respouters, especially along roads.

4.6.2.10.5. Specimens studied

South Africa. WESTERN CAPE: **3321 (Ladismith)**: Seweweekspoort, Swartberg Forest Reserve, 18 miles (29 km) from Ladismith (-AD), 7 May 1963, *Taylor H.C. 4724* (K, NBG); Huisrivierkloof (-AD), 4 Feb 1986, *Geldenhuys C.J. 1081* (K); Seweweekspoort, 72 km from Laingsburg to Calitzdorp (-AD), 20 Aug 2012, *Stirton C.H.* and *Muasya A.M. 13627* (BOL); Bobuffelsfontein, Klein Swartberg (-AD), 12 Dec 1981, *Stirton C.H. 10292* (K); Little Karoo, Gamka Nature Reserve (-DB), 8 Dec 1987, *Erasmus R. 142* (NBG); north side of Garcia's Pass, Riversdale (-CC), 5 Aug 1951, *Middlemost G.J. 1703* (NBG); Mossel Bay, Cloetesberg (-DD), 26 Jun 1987, *Linder H.P. 4157* (BOL). **3322 (Oudtshoorn)**: Swartberg Pass, Oudtshoorn side, near summit (-AC), 5 Nov 1986, *Fellingham A. 1235* (NBG); Swartberg Pass (-AC), 7 Aug 1949, *Morris S.D. 214* (NBG); lower South West slopes of Swartberg Pass (-AC), 4 Nov 1970, *Strauss M.E. 40* (NBG); Swartberg Pass (-AC), 19 Aug 1986, *Van Wyk C.M. 2666* (PRE); Seweweekspoort, Ladismith district (–AC), 9 May 1981, *Brusse F.A. 3548* (PRE); half way up Swartberg Pass (-AC), 19 Feb 2009, *Muasya A.M. & Stirton C.H. 4445* (BOL); Top of Swartberg Pass (-AC), 22 Feb 2011, *Muasya A.M. & Stirton C.H. 5965 1&2* (BOL); mid-northern slopes of Blesberg, next to forestry track, Swartberg mountains (-BC), 15 Dec 1986, *Vlok J.H.J. 1780* (PRE).

4.6.2.11. *Psoralea glabra* E.Mey., Comm. Pl. Afr. Austr. 1: 83 (1836); Walpers, Repert. Bot. Syst. 1: 656 (1842); Lock, Leg. Afr. Check-list: 458 (1989); Schmidt, Lotter & McCleland: Jacana Media: 216–218 (2002). Type: South Africa, Eastern Cape, "Witbergen, am Fuss der Berge, bei Rietvlei, Bamboeshoek, Bamboosspruit en Wilgerboschsprui", *Drége* s.n. (MO!, lecto. here designated; S!, isolecto.). Meyer had a very broad concept for his species. He designated four syntypes. His syntype a) is chosen as the lectotype as it is typical of the species across its range and best fits the protologue. Syntype b) is a different species from the Camdeboosberg and has been recently described as *P. margaretiflora* C.H.Stirt. & V.R.Clark (Stirton *et al.* 2011). Syntype c) has not been found and finally Syntype d) is an allopatric species from the Western Cape identified in this paper as *P. intonsa* C.H. Stirt., Muasya & A. Bello.

Psoralea pinnata L. var. *glabra* (E. Mey.) Harv. in Harv. & Sond., Fl. Cap. 2: 145 (1862); Burtt Davy, Fl. Pl. Ferns Transvaal, pt. 2: 375 (1932). Type as above.

Slender woody resprouter up to 3 m tall. *Stems* few to many, branching at or near the base, spreading to erect and arching, rigid, grey but turning brown when old, plants become quite leggy with age with foliage only in the upper parts; branches striate, glabrous or with a few short hairs on youngest parts. *Seasonal shoots* short, green, glabrous or with a few short hairs on youngest parts. *Leaves* 5–9-foliolate, imparipinnate, distributed along the seasonal branches, patent, petiolate; leaflets 9–15×0.5–1.5 mm, linear, apiculate, glabrous, gland-dotted, terminal leaflet shortest, basal pair variably longer, equal to or shorter than terminal, laterals about the same length of the terminal, three uppermost commonly pinnately (or more rarely digitately) inserted, all glabrous; petioles 3–4 mm long, shorter than the terminal leaflet; rachis 5–10 mm long; stipules 1.3–2.2×0.5–1.3 mm, persistent, subulate, swollen, recurved, margins reflexed, becoming woody on old branches, fused for half their length, glabrous. *Inflorescences* axillary, borne in upper axils of seasonal shoots with or without leafy extension, pseudo-capitate or pseudo-spicate, lax, 1–3 flowers per axil, pedunculate, pedicellate; peduncles stout and rigid, shorter to longer than the subtending leaf, 4–11 mm long; cupulum bilobed or trilobed, glabrous, 2.5 mm long; pedicel 2–3 mm long. *Flowers* 10−13 mm long, mauve to pale mauve. *Calyx* 3−5 mm long, tube 3–4×3.5 mm, glabrous to sparsely hairy, densely glandular; lobes sub-equally developed, all acuminate, emarginate; tube and lobes sometimes densely dark pubescent within and appearing ciliate. *Standard petal* 10–11×8–10 mm, obovate, mauve to pale mauve but may appear whitish blue in some light, veins purple; claw 2–3 mm long. *Wing petals* 10–11×3–4 mm, mauve to almost white (especially northern populations), narrowly oblong-oblanceolate; fused to but longer than the keel; sculpturing present, basal, comprised of 2–3 rows of 8–10 transcostal lamellae, claw 2–3 mm long. *Keel* 8–10×2.5–3.5 mm, dark purple on inner face at apex; claw 2−3 mm long. *Androecium* 10 mm long, tenth stamen free, fenestrate. *Pistil* 9 mm long, ovary 1.3 mm long, style 6 mm long, straight then curved upwards, swollen at point of flexure, height of curvature 2.5 mm long, sparsely covered with club-shaped glands. *Fruits* oblong, 3–4×2.5–3.0 mm papery, wrinkled, remaining enclosed in the desiccated calyx. *Seeds* solitary, 3–3.7×2–2.5 mm, dark brown to black (Figure 19).

4.6.2.11.1. Diagnostic characters

Psoralea glabra is similar to *P. margaretiflora* in growth habit (both resprouters) and *P. latifolia* in height (both grow up to 3 m tall) with which it is partially overlapping in the Sneeuberg Centre of Floristic Endemism. It can be distinguished from these species by a combination of its resprouting habit (versus reseeding in *P. latifolia*), narrower leaflets, <1 mm wide (versus 2.5−5 mm wide leaflets in *P. latifolia* and up to 1.5 mm in *P.*

margaretiflora), glabrous shoots (versus glabrous to glabrescent in *P. latifolia*), mauve to pale mauve flowers (versus white to greenish white in *P. margaretiflora* and mauve to blue in *P. latifolia*), smaller 3–5 mm long calyces (versus 5–6 mm in *P. margaretiflora* and 7–8 mm in *P. latifolia*), shorter 4–11 mm long peduncles (versus longer 15−17 mm in *P. margaretiflora* and 15–25 mm in *P. latifolia*) and 2–3 mm long pedicels (versus 1–1.5 mm in *P. margaretiflora* and 2–4 mm in *P. latifolia*).

4.6.2.11.2. Distribution and habitat

Psoralea glabra inhabits damp valleys and seepages in mountain fynbos and temperate grasslands at an elevation of 30−2300 m. It extends from the Eastern Cape up to the southern coast and midlands of KwaZulu-Natal into Mpumalanga and eastern Free State in South Africa and then into Swaziland and southern Mozambique (Figure 17). It falls within the Great Fish Thicket, KwaZulu-Natal Coastal Belt, Transkei Coastal Belt, Amathole Montane Grassland, Stinkfonteinberge Quartzite Fynbos, Midlands Mistbelt Grassland, Ngongoni Veld and Bhisho Thornveld vegetation types (AT 11, CB 3, CB 5, FFq 6, Gd1, GS 9, SVs 4, and SVs 7) (Hoare *et al.* 2006; Rutherford *et al.* 2006; Rebelo *et al.* 2006; Mucina *et al.* 2006a).

4.6.2.11.3. Etymology

The specific epithet *glabra* is derived from the Latin *glabrus –a –um* (hairless or smooth) and refers to the glabrous nature of this species. It is said to be called "isithuziso sezulu" in Zulu (*Heywood 253*).

4.6.2.11.4. Conservation status

Psoralea glabra has an automated status of least concern (LC) (Raimondo *et al.* 2009). In this study, it has been assessed based on the five IUCN (IUCN 2012a, 2012b; von Staden *et al.* 2009) criteria and does not qualify for any of the categories of threat. Although it has a wide range of distribution and is not threatened, overall a number of localities outside protected areas are threatened by expanding rural settlements, coastal ribbon development, and this need to be monitored in future.

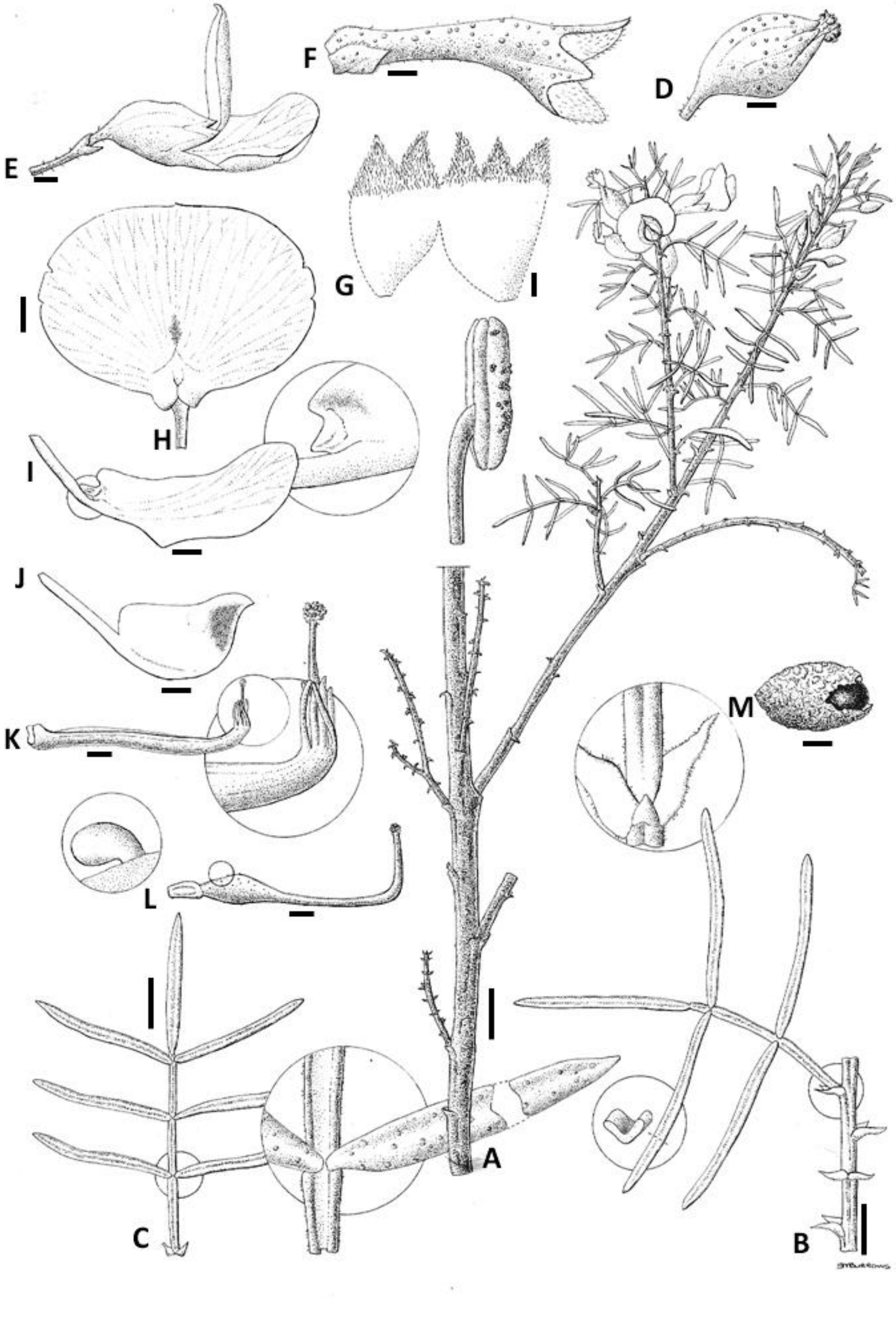


Figure 19. Vegetative and reproductive morphology of *P. glabra* (A) flowering shoot; (B) a twig showing the stipules; (C) 7-foliolate leaf; (D) bud; (E) flower; (F) peduncle terminating with a cupulum; (G) calyx opened out; (H) standard petal; (I) wing petal; (J) keel; (K) androecium; (L) pistil; (M) fruit. Voucher *Drége* s.n. (MO). Scale bars: A−C 1 cm; D–M, 1 mm. Artist: Sandi Burrows.

4.6.2.11.3. Specimens studied

South Africa. MPUMALANGA: **2531(Komatipoort)**: Vooruitzicht 374JU, Malondweni, Songimvelo Game Reserve (-CC), *Balkwill K., McCallum D.A. & Reddy R.A. 12000* (J, K); Barberton (-CC), *Locke s.n.* (PRE); South of Shiyalongubo Dam, Ugutugulo, River Gorge, West of Gorge (-CD), 8 Aug 2002, *McMurtry D. 10723* (PRE); Ida Doyer Nature Reserve, 20 m SE of Barberton (-CD), 14 Sep 1971, *Werger M. 1447* (PRE). FREE STATE: **2926 (Bloemfontein):** Leppan's Farm, 2 m south of Collingham (-DB), 23 Mar 1947, *Story R. 2160* (GRA, PRE). KWAZULU-NATAL: **2731 (Louwsberg):** 7 m west of Ngome (-CD), 12 Apr 1956, *Codd L.E.W. 9583* (L, PRE); 7 m W of Ngome, 12 Apr 1956, *Codd L.E.W. 9583* (K, PRE). **2831 (Nkandla)**: Farm "Misty Acres", Eshowe (-CD), 17 Apr 1975, *Law P. 36* (NBG). **2829 (Harrismith):** Estcourt (-DB), 14 Feb 1907, *Medley Wood J. 10261* (NH). **2830 (Dundee):** Qudeni Forest (-DB), 11 Dec 1988, *Stirton C.H. 11843* (NU). **2831 (Shongweni)**: Nkandla forest camping site (-CA), 2 Apr 1986, *Jordaan M. 682* (PRE); Nkandla Forest Reserve (-CA), 4 Apr 1986, *Van Wyk A.E 7344* (PRE); 26 Mar 1956, *Edwards D. 1328* (PRE); 5 m from Mbongolwane (-CC), 10 May 1962, *Strey R.G. 42*05 (PRE); Ngoye Forest Reserve (-DD), 22 Apr 1966, *Venter H. 2342* (PRE); Feb 1904, *Medley Wood J. 9314* (PRE); Umlalazi Nature Reserve (-DD), 30 Nov 1983, *Buthelezi C. 347* (NH). **2832 (Mtubabtuba):** Lake St. Lucia (-AD), 1 Feb 1975, *Taylor R.A. 291* (K, NU); Lake St. Lucia (-AD), 20 Jul 1978, *Pooley E. 2109* (K, NU). **2930 (Pietermaritzburg):** Colbourne Farm (-AB), 10 Jul 1993, *Vos W. & McGregor T. 572* (K); Greytown (-BA), Jan 1930, *Doidge E.M. s.n.* (PRE); Greytown (-BA), Apr 1930, *Wylie J. s.n.* (BOL, PRE); Ahrens (-BA), Apr 1943, *Fisher B.S. 466* (PRE); Ozwatini (-BA), 23 Jun 1967, *Strey R.G. 7510* (PRE); Ismont (-DD), 14 Sep 1969, *Strey R.G. 9054* (PRE); Richmond (-CD), 30 Apr 1964, *Bayliss R.D.A. 2196* (NBG); 11 Apr 1905, *Medley Wood J. 9852* (PRE); Ndwedwe (-DD), 23 Jun 1967, *Strey R.G. 7510* (K, PRE); Botha's Hill (-DD), 29 Apr 1892, *Medley Wood J. s.n.* (GRA, NH); Hillcrest (-DD), May 1917, *Thode H.J. 3458* (NBG); Near Port Natal (Durban) (-DD), 1864, *Dr. Sutherland s.n.* (K, TCD); Prope Port Natal (-DD), 1851, *Plant R.W. 39* (K, TCD). **2931 (Stanger):** 5 m from Mbongolwane, 10 May 1962, *Strey R.G. 4205* (K, PRE). EASTERN CAPE **3029 (Kokstad):** Kokstad (-CB), 30 Apr 1971, *Baylis R.D. 4693* (MO); Ntsizwa (-CD), 22 Feb 1972, *Strey R.G. 10760* (PRE); Mt. Ayliff, Tabankulu (-CD), 14 Apr 1935, *Jacot Guillarmod A. s.n.* (PRE). **3030 (Port Shepstone)**: Trienedan, Umgayeflat (-AD), 21 Apr 2008, *Rudatis A.G.H.13* (NBG); Illovo (-CD), 4 May 1997, *Medley Wood J. 6411* (PRE); Beacon Hill (-CD), 3 Apr 1966, *Strey R.G. 6524* (PRE); Black Trail, Oribi Gorge (-CD), Feb 1973, *Davidson L.E. 2427* (J, PRE); Ellismere (-CD), 5 Dec 1916, *Rudatis H. 1024* (PRE, Z); Dumisa (-CD), 24 Apr 1908, *Rudatis 359* (L); 21 Apr 1908, *Rudatis H.*

s.n. (K); Vernon Crookes Nature Reserve (-CB), 17 Mar 1989, *Ward C.J. 9797* (PRE); Uvongo (-CD), 29 Apr 1956, *Whellan 1050* (PRE); Kranskloof 8100, Paddock (-CD), 13 Dec 1988, *Heywood M. 253* (PRE); Melon Drive, Palm Beach (-CD), 10 Jan 2008, *Muasya A. M. & Stirton C.H. 3646* (BOL); Mpenjati resort, Old Main road (-CD), 21Apr 1986, *Abbott A. 3103* (NU); Palm Beach (-CD), 6 Nov 1980, *Stirton C.H. 8093* (K); 5 Jan 1981, *Germishuizen G. 1756* (K, PRE). **3130 (Port Edward):** Port Edward (-AA), Jan 2000, *Edwards T. & Potgieter C. 1819* (K, NU); Pont Outpost (-AA), 23 Feb 1986, *Abbott A. 3023* (NU); Umtamvuna Nature Reserve (-CA), 19 Mar 2006, *Abbott A. 8395* (PRU); Clearwater Farm, 31 Dec 1982, *Abbott A. 674* (PRU); Mpenjati resort, Old Main road 21 Apr 1986, *Abbott A. 3103* (NU). EASTERN CAPE: **3128 (Umtata):** Hills near Umtata (-DB), 20 Jan 1895, *Schlechter R. 6337* (Z, PRE). **3129 (Port St. Johns)**: Magwa Falls (-BC), 22 Apr 1955, *Lewis s.n.* (PRE, SAM); Magwa Store (-BC), 11 May 1969, *Strey R.G. 8551* (PRE); Misty Mount, Libodi, Transkei (-CA), 21 Jun 1965, *Sole G.Z. 2* (NBG); Port St. Johns, Jul 1889, *Flanagan H.G. 178* (PRE).**3225 (Somerset East)**: ± 18 km West of Somerset East, East of Bruintjieshoogte, Farm 63, North of Bergvlei (-CA), 25 Feb 2007, *Helme N.A. 4583* (NBG); Glen Avon Falls, Naudesrivier, Glen Avon Farm, Somerset East Division (-CA), 4 Nov 2008, *Clark V.R. & Andrews A. 75* (BOL, GRA); Strydomsberg, Zunga Catchment (-CA), 8 Apr 1974, *Scharf H.T. 1233* (PRE); Edge of Boschberg Escarpment, Glen Avon Farm (-CA), 7 Nov 2008, *Clark V.R. & Andrews A. 340* (BOL, GRA); Boschberg Nature Reserve (-CA), 5 Dec 2008, *Clark V.R., Daniels R.J., Fabricius M., & Le Roux J.M. 265* (GRA). **3226 (Fort Beaufort)**: Top of Katberg Pass, Stockenstroom (-BC), 10 Mar 1947, *Storey R. 2086* (PRE); 23 Dec 1980, *Germishuizen G. 1572* (K, PRE); Rabbits's Bush (-BC), 23 Jul 2005, *Nevhutalu L.S. & Nkuna L.A. 2000* (K); Rabbit's Hill, 29 Aug 1971, *Bayliss R.D. 4758* (MO); Grahamstown (-BC), 27 Sep 1984, *Stirton C.H. 10774* (K); Dohne Peak (-BC), 5 Jul 1942, *Acocks J.P.H. 8957* (PRE). **3227 (Stutterheim)**: Fort Cunynghame (-AD), 1897, *Sim T.R. 1208* (PRE); Windvogelberg near Cathcart (-CA), Apr 1962, *Roberts B.R. 1770* (PRE); Cathcart (-CA), 26 Feb 1894, *Kuntze O. s.n.* (K, NY); Keiskammashoek (-CA), Apr 1944, *Goulimis C. s.n.* (BOL); Southernmost peak of "Three Hogs" (-CA), 5 Dec 1994, *Dold T. 1493* (GRA); Pirie, King Williams Town (-CD), 24 Jan 1936, *Taylor L.E. 1755* (PRE). **3325 (Port Elizabeth)**: 20.2 km, vanaf Coerneystasie of Zuurbergpas of pad na Somerset-Oos (-BC), 12 Dec 1984, *Grobbelaar N. 2905* (PRE); Zuurberg Sanatorium (-BC), 7 Apr 1934, *Long F.R.1244* (K, PRE). **3326 (Grahamstown)**: Signal Hill, Mountain Drive, Grahamstown (-AD), Grahamstown Nature Reserve (-AD), 3 Feb 1955, *Comins D.M. 976* (GRA, PRE); Aug 1923, *Roger F.A.* 28632 (K); 1865, *MacOwan P. s.n.* (K); *Hooper Cole 2188* (K); 20 Jul 1970, *Melwith R. 70/334* (K); 8 Jun 1976, *Booi 54* (GRA, PRE); 23 Jun 1950, *Karsten s.n.*

(BOL, PRE); Modderlaagde River (-AD), *Zeyher 903* (TCD); Between Stones Hill and Howieson's Poort (-AD), Aug 1931, *Rennie J.R. & B.R. 63* (GRA); Faraway farm, 3 km E. of Coldspring (-AD), 20 May 1989, *Jacot Guillarmod A. 9547* (GRA); Coldspring (-AD), *Gower L. s.n.* (PRE); Stones Hill (-BC), May 1940, *Britten L.L. 7* (BOL); Grahamstown (-BC), 27 Sep 1984, *Stirton C.H. 10774* (K, NU); Sep 1918, *Van Dam s.n.* (GRA); 23 Jul 1950, *Karsten M.C. s.n.* (BOL, PRE); Jun 1925, *Dyer R.A. 50* (PRE); Jan 1917, *Breyer 16861* (PRE); May 1915, *Bury s.n.* (PRE); Collingwood Kloof (-BC), 6 Jun 1948, *Leighton F.M. 3079* (BOL, SAM). **3327 (East London):** East London (-BB), 1888, *Thode H.J. 6670* (NBG).

Swaziland. **2531 (Kiepersol)**: Hhogho, Kings Forest, Bulembu, Swaziland (-CC), 13 Aug 2002, *Dlamini G.M. A2336* (PRE); 2.5 m from Havelock Asbestos Mine (-CC), 22 Apr 1953, *Codd L.E.W. 7835* (PRE); Kings Bush, Havelock (-CC), Mar 1961, *Miller O.B. 5414* (K, PRE, SRGH); Siteki, near Army post (-CC), 24 May 1956, *Compton R.H. s.n.* (K). **2621 (Mbabane)**: Mbabane, edge of Forest (-AC), Jul 1951, *Compton R.H. 22733* a&b (NBG); 17 Feb 1961, *Dlamini G. s.n.* (PRE); Mbezluzi falls, Mbabane (-AC), 9 Apr 1955, *Compton R.H. 25080* (NBG); Duiker's Bush, Swaziland, 24 May 1956, *Compton R.H. s.n.* (K).

Mozambique. **2632 (Bela Vista)**: 4 km along the road to MaputoVlei (-BC), 3 Jan 1980, *de Koning J. 7895* (K, LMU, NBG, UEM). **Without precise locality:** Faku's Territory, Mpondoland, Jul 1864, *Dr. Sutherland s.n.* (K, TCD); Between Blauuwkrans and Coldstream, *Pocock A. s.n.* (Z-00083944); Pondoland, *Bachman F. 570* (B, Z-00083938); Natal, 1801, *Sanderson 364* (TCD); Fynn's Bush, 17 Feb 1970, *Strey R.G. 9631* (PRE); Bloemhof, Apr 1910, *Thode H.J.* 4494 (PRE); Albany, *Williamson s.n.* (TCD); Natal, 1882, *Medley Wood J. 84* (BOL).

4.6.2.12. *Psoralea margaretiflora* C.H.Stirt. & V.R.Clark in PhytoKeys 5: 32 (2011). Type: South Africa, Eastern Cape, Somerset East (3225): Graaff-Reinet Dist., Farm 360, Petersburg, Asante Sana Private Game Reserve (-AC), Mar 2008, *Clark V.R. & Crause I. 4* (GRA, holo. !; BOL!, K!, NBG!, NSW!, PRE!, S!, iso.).

Erect resprouter up to 2 m tall, forming dense clumps. *Stems* erect, 1−30, branching at or near the base, greyish-brown with numerous white storied lenticels, bare except for bright green, glabrous, glandular, seasonal shoots in the upper axils, radiating either terminally from stems or along older shoots giving a broom-like appearance. *Seasonal shoots* produced seasonally on old stems, leafy along entire length. *Leaves* 7-foliolate at base of each seasonal shoot, 5-foliolate thereafter, leaf size variable, larger on water shoots from the rootstock, distributed evenly along the seasonal branches, glabrous, petiolate, patent; leaflets (15)18–25 (33)×(0.3) 0.5 –1(1.5) mm, linear lanceolate, basal pair longest, mid-pair shortest, and terminal leaflets second longest; three uppermost pinnately (rarely digitately) inserted; shiny, glabrous, dark green; apex acuminate, base rounded; petioles 4–5 mm long, shorter than terminal leaflet; rachis 7–15 mm long; stipules 2–3(4)×2 mm, persistent, shorter than petiole, triangular, glabrous, rigid, semi-patent; rapidly senescent on flowering shoots. *Inflorescences* axillary, borne in most axils of seasonal shoots, pseudo-spicate, lax, 1–3(5) flowers per axil, pedunculate, pedicellate; peduncles stout and rigid, (10) 15−17 mm long, equal in length to the subtending leaf; cupulum trilobed, glabrous, lobes unequally developed, lower lobe longest, acuminate, upper two lobes fused for half their length, yellowish, rapidly senescent, 1.0−1.2 mm long; pedicels 1–1.5 mm long. *Flowers* 10−12 mm long, white to greenish white. *Calyx* 5–6 mm long, tube 3–4×4 mm, ribbed, glandular, pale green, glabrous on outside; lobes equally developed, same length as the tube, triangular, all 3 mm long, carinal lobe cucullate at apex, finely black-haired on inner face of lobes. *Standard petal* 9–10×7–8 mm, broadly elliptic, greenish white, nectar flash purple, trifid above the strongly developed auricles; claw 2–3 mm long. *Wing petals* 9–10×3–4 mm, white, locked into keel but not fused, petal sculpturing present, upper basal, comprising 7–8 transcostal parallel lamellae; claw 2–3 mm long. *Keel* 7–9×4–5 mm, apex deep purple; purple on inner face of apex; claw 5 mm long. *Androecium* 9 mm long, tenth stamen free, sheath split adaxially, fenestrate. *Pistil* 9 mm long, ovary 1.5 mm long, stipitate, glabrous, style straight for 6 mm long, then curved upwards, thickened at point of flexure, height of curvature 2 mm, erect, penicillate. *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* solitary, 2–4×2 mm, dark brown to black (for illustrations see Stirton *et al.* 2011−Figure 1).

4.6.2.12.1. Diagnostic characters

Psoralea margaretiflora is characterised by its tall habit of many stiff bare stems bearing numerous short seasonal shoots, burst-branching from the apex, 5(–7)-pinnate leaves, and its unique greenish white flowers with a small trifid purple nectar guide and translucent veins. It resembles *P. glabra* in growth habit, but differs in its flat, erect to semi-patent, triangular stipules not prominent or persistent on old shoots (versus subulate, reflexed, and becoming woody on old branches), basal leaflets shortest (versus terminal leaflet shortest), peduncles (10)15–17 mm long (versus 4–11 mm long), white to greenish white flowers (versus pale to dark mauve); calyx yellowish green with lobes equally developed (versus pale green and subequally developed), broadly elliptic standard petals with translucent vein (versus obovate with purple veins), and white wing petals (versus purple). It has been confused in its area of distribution with the single stemmed reseeder *P. oligophylla* Eckl. & Zeyh. which has a lax, virgates, spreading habit with numerous long glaucous seasonal shoots, 1–3-foliolate, glaucous leaves, numerous white flowers, and standard petals with a purple ring surrounding a bright yellow nectar guide. Stirton *et al.* (2011) provides a detailed key to separate *P. margaretiflora*, *P. latifolia, P. glabra* and *P. oligophylla*.

4.6.2.12.2. Distribution and habitat

Psoralea margaretiflora is endemic to the lower and mid-escarpment slopes of the Sneeuberg in the Graaff-Reinet District, being concentrated on the Kamdebooberge, Koudeveldberge and Toorberg in the west, and from the Nardousberg to Aasvoëlkrans (behind Pearston) in the east. As pointed out by Stirton *et al.* (2011), the vegetation types inhabited by this species are difficult to classify. It occurs variously in Karoo Escarpment Grassland and "Afromontane Grassland" verging into Mountain Fynbos, but also occurs in closed *Otholobium macradenium* shrubland at elevations of 1200–1800 m (Figure 20). The plant grows primarily on rich turf soils and colluvium associated with dolerite. It is mainly associated with the following vegetation types: Karoo Escarpment Grassland and Upper Karoo Hard veld (Gh 1 and NKU 2) (Mucina *et al.* 2006a; Mucina *et al.* 2006b). On the eastern, wetter Boschberg end of the Sneeuberg it is replaced by *P. glabra*. In the summer months the plants are drenched daily in the mornings by thick mountain mist.

4.6.2.12.3. Etymology

The species epithet *margaretiflora* is derived from the Latin words *margaritaceus* -a –um (pearl-like) and *flos* (flower) and alludes to the striking appearance of its pearl-like flowers as seen in early morning mountain mist.

4.6.2.12.4. Conservation status

Psoralea margaretiflora is assessed as least concern (LC). It is a range-restricted species (EOO 2385 km^2), but locally abundant on middle and lower slopes of the Sneeuberg Range and is not threatened (Clark and Raimondo 2014).

4.6.2.12.5. Specimens studied

South Africa. EASTERN CAPE: **3223 (Rietbron)**: Kamdebooberg, Farm 9B, "Sleeping giant", (-BD), 23 Jan 2011, *Clark V.R., Stirton C.H.* and *Weston P. 2* (BOL, GRA, K, NSW, PRE); South east of slopes of Goewermentsberg, Farm Oaklands 104 (-BD), 25 Jan 2011, *Clark V.R., Stirton C.H.* and *Weston P. 91* (BOL, GRA, K, NSW, PRE); Farm Oaklands 104 (-BD), 4 Apr 2008, *Clark V.R.* and *Crause I. 140* (BOL); Farm Oaklands 104 (-BD), Dec 2008, *Clark V.R.* and *Cloete C.1* (BOL, GRA, K, NSW, PRE); Farm Oaklands 104 (-BD), 19 Dec 2008, *Clark V.R., Stirton C.H.* and *Weston P. 1* (BOL); Camdeboo Mountains, Farm Palmietfontein (-BD), 9 Feb 1960, *Acocks J.P.H. 21044* (K, PRE). **3224 (Graaff Reinet):** Farm Onbedacht 294, Koudeveldberge in Sneeuberg (-AA), Dec 2007, *Clark V.R.* and *Pienaar C. 369* (GRA); Farm Onbedacht 294, eastern slopes of Koudeveldberge Sneeuberg (-AC), Nov 2006, *Clark V.R. & Te Water Naudé T. 140* (GRA). **3225 (Somerset East)**: Graaff–Reinet to Jansenville (-AC), 17 Aug 1999, *Nienarber E.P. EN557* (PRE); Petersburg, Asante Sana Private Game Reserve, Suurkloof in the Sneeuberg (-AC), Dec 2005, *Clark V.R.* and *Coombs G. 101* (GRA, PRE); Sneeuberg, Farm Buffelshoek 25, lower slopes west of Aasvoëlkrans (-AC), Nov 2007, *Clark V.R.* and *Rose M.C. 20* (GRA); Farm Stockdale 387, in upper reaches of Naudeshoekspruit valley (-AD), Oct 2006, *Clark V.R.* and *Ramdhani S. 197* (GRA). **Without Precise locality**: Belfort, Dec 1911, *Dr. Jacottet* (Z).

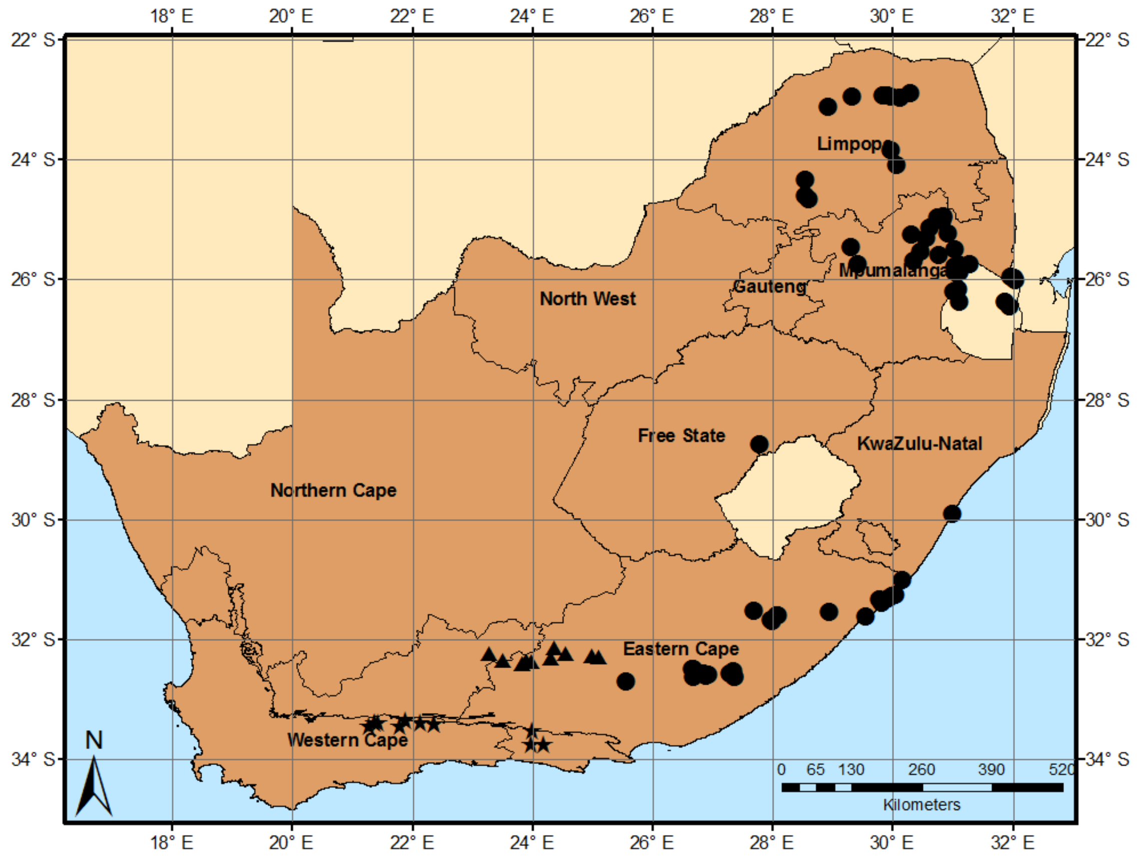


Figure 20. Known distribution of *P. margaretiflora* (triangles), *P. latifolia* (circles) and *P. imminens* (stars).

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4.6.2.13. *Psoralea imminens* C.H. Stirt. *sp. nov.*, Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); *Psoralea* sp. 25, Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Western Cape, Oudtshoorn (3322): Rus-en-Vrede Waterfall (-AD), 11 Nov 1986, *Stirton C.H.* and *Žantovská J. 11564* (K!, holo.; NU!, PRE!, iso.).

Slender shrub to small tree up to 3 m tall, reseeder. *Stems* erect, single, branching in upper portion, flexuous, yellowish-green brown, with numerous greyish white storied lenticels, becoming grey and deeply fissured with age. *Seasonal shoots* smooth, glabrous or glabrescent, willowy. *Leaves* 5–7(9)-foliolate, filiform, imparipinnate, distributed along the branches, patent, spreading, glabrous, curving, with raised glands; leaflets 24–45×0.2–0.3 mm, filiform, glabrous, terminal leaflet shorter than basal pair which are longest, laterals about the same length as the terminal, three uppermost pinnately inserted; petioles 7–9 mm long, shorter than terminal leaflet; rachis 7−15 mm long, furrowed on abaxial face; stipules 0.7–1×0.3–0.8 mm, persistent, rapidly senescent at flowering, subulate, margins incurled, glabrous. *Inflorescences* axillary, borne in upper axils of seasonal shoots with or without leafy extension, pseudo-capitate, congested, 1–3 (5) flowers per axil; pedunculate, pedicellate; peduncles more than 45 mm long, filiform, flexuous, shorter to longer than the subtending leaf; cupulum trilobed, glabrous, lobes unequally developed, lower lobe partly cleft, carinal lobe longer, 2 mm long, sparsely hairy along the margins, setose or linear, positioned two thirds up peduncle; pedicels 4–5 mm long. *Flowers* 10−12 mm long, white to pale mauve, scented; buds beaked. *Calyx* 4−5 mm long, tube 3–4×3.5 mm; lobes equally developed, shorter than the tube, finely covered with white hairs, lobes triangular, carinal lobe tooth elongated, lobes splayed at anthesis, tending to purple with age, acute, inner face of lobes densely black pubescent. *Standard petal* 9–10×7–7.5 mm, elliptic, white to mauve, auricles slightly swollen, appendages not well developed, blade oblong, nectar guide prominent, comprised of two large free central white patches with a thinnish purple flash between them; claw 2–3 mm long. *Wing petals* 8–9×3–3.5 mm, broadly cultrate, longer than keel petals, sculpturing present, comprised of 2−3 rows of 6–10 transcostal lamellae; claw 3–4 mm long. *Keel* 5–6×3–3.5 mm, apex acute; claw 2–3 mm long. *Androecium* 11 mm long, tenth stamen free, fenestrate. *Pistil* 9 mm long, ovary 2 mm long, style 6 mm long straight, then curved upwards, widest at point of flexure, height of curvature 4 mm long, stigma penicillate. *Fruits* narrowly oblong, 4–5×2.5–3.0 mm, beaked, papery, pedicels persistent after seed shed. *Seeds* solitary, 2–4×2 mm long, dark brown to black (Figure 21).

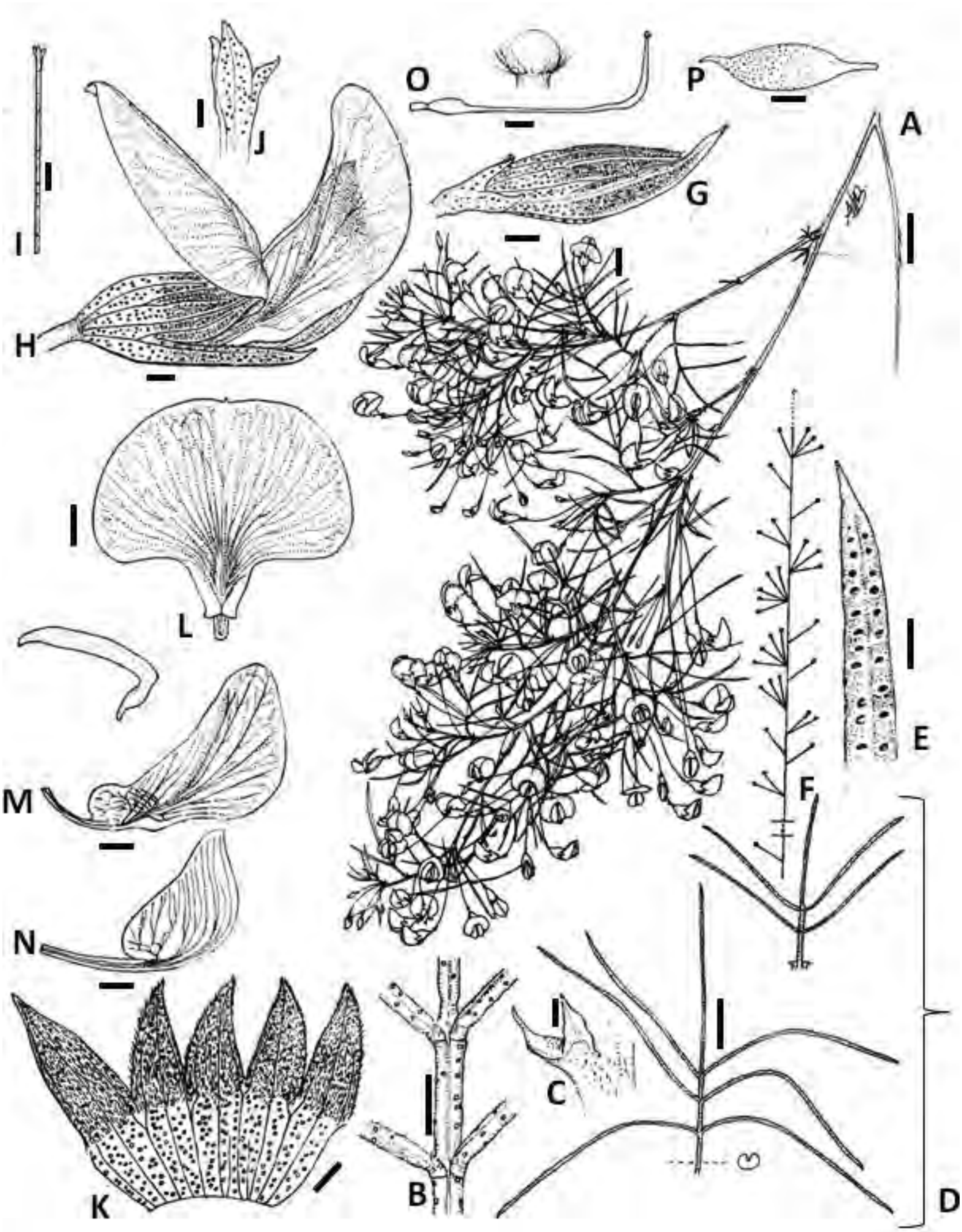

Figure 21. Vegetative and reproductive morphology of *P. imminens* (A) flowering shoot; (B) leaf insertion on a twig; (C) stipule; (D) 7 and 5-foliolate leaves; (E) leaf blade showing glands; (F); (G) bud; (H) flower; (I) peduncle; (J) cupulum; (K) calyx opened out with vexillary lobes to the right; (L) standard petal; (M) wing petal; (N) keel; (O) androecium; (P) pistil. Voucher *C.H. Stirton* and *J. Žantovská 11564* (K, PRE). Scale bars: A−F 1 cm; G–P, 1 mm. Artist: Angela Beaumont.

4.6.2.13.1. Diagnostic characters

Psoralea imminens is distinctive in the *P. pinnata* complex with its long filiform leaflets and peduncles, small cupulum, and smaller calyces. It differs from *P. pinnata* in being a smaller, less branched shrub up to 3 m tall (versus large much-branched shrub to small tree up to 5 m tall), with long, soft, filiform, spreading, glabrous, lax leaves (versus linear, rigid, villoso-pubescent, congested leaves); long filiform peduncles (versus rigid, sessile or subsessile peduncles), minute cupulum positioned two thirds up on peduncle (versus large cupulum at apex of peduncle and overlapping the large calyx), inflorescences pseudo-capitate, congested, borne in upper axils of seasonal shoots and free from subtending leaves (versus pseudo-spicate inflorescences borne in short terminal shootlets along the length of the seasonal flowering shoots and hidden within the subtending leaves). It is also similar to *P. elegans* in growth form but differs in its greenish brown stems with storied white lenticels, becoming grey and deeply fissured with age (versus smooth blackish brown with scattered white lenticels), filiform leaflets (versus linear lanceolate leaflets), stipules rapidly senescent at flowering (versus green at flowering), 1−3 white to pale mauve flowers per axil, exposed well above the subtending leaves (versus (2)3–6 mauve flowers per axil, hidden within leaves), and flower buds with a long extended lower calyx lobe (versus no extended lower calyx lobe).

4.6.2.13.2. Distribution and habitat

Psoralea imminens is a species that hugs streams and seepages along valley bottoms and ranges from the Calitzdorp region of the Klein Swartberg Mountains in the west through the Groot Swartberg Mountains to the Baviaanskloof Wilderness area in the east (Figure 20). It is found in North Swartberg Sandstone Fynbos, Kouga Sandstone Fynbos, Kango Limestone Renosterveld and Swartberg Shale Renosterveld vegetation types (FFs 23, FFs 27, FRl 1 and FRs 15) (Rebelo *et al.* 2006).

4.6.2.13.3. Etymology

The specific epithet *imminens* is derived from the Latin verb *immineo* (to hang, bend, project over) and refers to the distinctive droopy nature of the branches, leaflets and pedicels.

4.6.2.13.4. Conservation status

Psoralea imminens has a wide distribution range. It does not qualify for any of the categories of threat. It is therefore assigned a least concern (LC) status (von Staden *et al.* 2009).

4.6.2.13.5. Specimens studied

South Africa. WESTERN CAPE: **3321(Ladismith)**: Kloof (-AD), 28 October 1953, *Wurts T.M. 1207* (NBG); Klein Swartberg, above Ladismith (-AD), 17 Oct 1966, *Taylor H.C. 6964* (PRE); Seweweekspoort (-AD), 5 Nov 1953, *Wurts T.M. 1216* (NBG); Klein Swartberg (-AD), 1 Nov 1983, *Marshall A.H. 194* (PRE); Calitzdorp, Groenfontein farm, base of Swartberg (-CB), 17 Nov 2010, *Muasya A.M. & Stirton C.H.* 5795 (BOL). **3322 (Oudtshoorn)**: Van Swartberg-Onderkant Pas, Prince Albert (-AC), 10 Nov 1987, *Geldenhuys C.J. 1338* (PRE, NBG); Swartberg Pass, Prince Albert Div. (-AC), 11 Dec 1945, *Stokoe T.P. 9330* (BOL, PRE); 13 Nov 1938, *Hafström A.H. & Acocks J.P.H. 706* (PRE); Kloof below Kliphuis Vlei, Swartberg (-AC), Dec 1943, *Stokoe T.P. 9018* (BOL, PRE); Along the Dorpsrivier, Bottom Swartberg Pass on the Prince Albert side (-AC), 22 Feb 2011, *Stirton C.H. 13265* (BOL); 3 km from Prince Albert to Swartberg Pass (-AC), 19 Feb 2009, *Muasya A.M. & Stirton C.H. 4454* (BOL); 5 km from Prince Albert to Oudtshoorn via Swartberg Pass (-AC), 3 Jan 2008, *Muasya A.M. & Stirton C.H. 3596* (BOL); Rus-en-Vrede (-AD), 28 Aug 2010, *Muasya A.M. & Stirton C.H. 5575* (BOL); 28 Aug 2010, *Muasya A.M. & Stirton C.H. 5577* (BOL). EASTERN CAPE: **3323 (Willowmore)**: Braam Rivier, Kouga mountains (-DB), 28 Oct 1949, *Esterhuysen E. 16301* (BOL, PRE); 5 km N of Studtis, Baviaanskloof (-DB), 1 Nov 1980, *Van Wyk B-E. 617* (NBG). **3324 (Steytlerville)**: Kouga Mountains, mountain track that leads up to Koue Nek (-CA), 4 Nov 2011, *Euston-Brown D. 1626* (BOL); Witwaterkloof (-DA), 28 Nov 1986, *Geldenhuys C.J. 1282* (NU, PRE).

4.6.2.14. *Psoralea latifolia* (Harv.) C.H. Stirt. *comb. nov.* *P. pinnata* L. var. *latifolia* Harv. in Harv. & Sond., Fl. Cap. 2: 144 (1862); Burtt Davy, Fl. Pl. Transv. 2: 375 (1932); Verdoorn in Fl. Pl. Afr. 26: 1029 (1947); Schmidt, Lotter & McCleland: Jacana Media, 216−218 (2002). Type: South Africa, Eastern Cape, Fort Beaufort (3226): Katberg (-BC), November 1880, *Hutton s.n.* (TCD!, lecto. here designated). [Notes: i) During an investigation of the complicated synonomy of *P. pinnata*, it was noted that William Harvey's varietal names under *P. pinnata* either referred to what we now know as several distinct species or they were a mixture of different species with no clear indication of types. The name *Psoralea pinnata* L. var. *latifolia* Harv. has been used regularly in the northern Provinces of South Africa. When he published this varietal name Harvey's concept included *Psoralea arborea* Sims (treated above) plus some pinnate flat-leaved plants from "Caffraria" and "Port Natal". When Harvey established this species, he confused 2 elements. He cited the type plate of *P. arborea* (an earlier described species) which is a Western Cape species (See under that species), a specimen cited incorrectly by Ecklon & Zehyer 1514 from the Katberg (which is designated here as the lectotype), and a reference to De Candolle's citation of *P. arborea*. As *P. arborea* and *P. latifolia* are disjunct and distinct species and as there is only one specimen cited in the protologue, we have chosen that as the lectotype; ii) Compton's (F. Swaziland p.264, 1976) *P. pinnata* belong to this species.

A small to large, slender, reseeding shrub up to 3 m tall. *Stems* erect, single, branching in upper portion, rigid, brown with storied lenticels, unusual in genus mostly covered in crustose lichens; branches pale brown, striate, hairy or glabrous. *Seasonal shoots* glabrous to glabrescent, often purplish, ridged. *Leaves* 5–7(9)-foliolate, imparipinnate, distributed along the seasonal shoots, lax, erect, petiolate; leaflets 10–50×2.5–5(6) mm, linear-oblong to linear-lanceolate, apiculate at the apex, cuneate to narrowly rounded at the base, densely gland-dotted, glabrous to sparsely pubescent, terminal leaflet about the same length to the laterals, basal pair longest, apex of terminal leaflet acute, three uppermost digitately inserted; petioles 10–15 mm long, shorter than terminal leaflet; rachis 15–20 mm long; stipules 2.5–4×2 mm, persistent, separate, thickened, sides inrolled, recurved, ovate-lanceolate, acuminate. *Inflorescences* axillary, borne in upper axils of seasonal shoots but without leafy extension, pseudo-capitate, lax, 1-flowered, fasciculate, 1–3(6) per axil, pedunculate, pedicellate; peduncles stout and rigid, 15–25 mm long, shorter than the subtending leaf, cupulum bilobed, one lobe slightly cleft, bristly pubescent with black hairs, 3 mm long, lobes equally developed, triangular; pedicels 2–4 mm long. *Flowers* 13−14 mm long, mauve to blue. *Calyx* 7–8 mm long, tube 4–5×3 mm; lobes subequally developed, shorter than the calyx tube, oblong-ovate, sub-acute or rounded at the apex, lower most lobe largest; both tube and lobes glabrous to pubescent on the outer surface, but with characteristic black hairs within the lobes, margins of lobes ciliate. *Standard petal* 13–15×12–15 mm, broadly obovate, dark mauve to blue and paler at the margins with variably coloured M-shaped nectar guide; claw 3–4 mm long. *Wing petals* 12.5–13×4–6 mm, white below and pale mauve in the upper half, sculpturing present; claw 5–7 mm long. *Keel* 11–12×4–5 mm, white, apically suffused with dark violet-purple on inner face; claw 6 mm long. *Androecium* 14 mm long, tenth stamen shortly united with staminal sheath adaxially fenestrate. *Pistil* 14 mm long, ovary 3 mm long with stipe 1 mm long, style straight for 7 mm long then curved upwards for 4 mm. *Fruits* 6×3.8 mm, ellipsoid, wrinkled. *Seed* solitary, 4.2×3 mm, dark brown, ellipsoid (Figure 22).

4.6.2.14.1. Diagnostic characters

Psoralea latifolia is a distinctive species in the *P. pinnata* complex in being the only species with broad, flat, linear-oblong to linear-lanceolate leaflets to 6 mm wide. It is similar to *P. arborea* in growth habit from which it differs in being a more slender shrub up to 3 m tall (versus large robust shrub or tree up to 10 m tall), stems brown with storied lenticels (versus greenish grey with scattered lenticels), seasonal shoots glabrous to glabrescent (versus villoso-pubescent), leaves 5–7(9)-foliolate (versus 7–9(11)-foliolate), terminal leaflet same length as laterals (versus shorter than laterals), leaflets linear-oblong to linear-lanceolate and 1.5–5(6) mm wide (versus linear to linear lanceolate and 1–2 mm wide), petioles 10–15 mm long (versus 5–8 mm long), peduncles 15–25 mm long (versus 45–60 mm), northern and eastern distribution (versus western Cape).

4.6.2.14.2. Distribution and habitat

Psoralea latifolia has the widest distribution in Southern Africa with its main distribution in the wetter summer rainfall region. In South Africa it ranges from the Eastern Cape Province to KwaZulu-Natal and into the Gauteng, Mpumalanga and Limpopo provinces but has outlier populations that occur in Swaziland and Mozambique (Figure 20). It inhabits montane and coastal grasslands, mountain fynbos, the margins of thickets in damp valleys and seepages between 50−1600 m above sea level. It has been found in Eastern Cape Escarpment Thicket, Southern Mistbelt Forest, Amathole Montane Grassland, Barberton Montane Grassland, Lydenburg Montane Grassland, Northern Escarpment Quartzite Sourveld, Drakensberg Foothill Moist Grassland, Tsomo Grassland, Machado Sweet Bushveld, Springbokvlakte Thornveld, Tshokwane-Hlane Basalt Lowveld, Legogote Sour Bushveld and Southern Lebombo Bushveld vegetation types (AT 13, FOz 3, Gd 1, Gm 17, Gm 18, Gm 23, Gs 10, Gs

15, SVcb 20, SV1 5, SVl 9 and SVl 16) (Hoare *et al.* 2006; Rutherford *et al.* 2006; Mucina *et al.* 2006b).

This species is mostly visited by large Xylocopid bees but small sunbirds have been observed visiting flowers in the Sabie area of Mpumulanga (Stirton, pers. obs.).

4.6.2.14.3. Etymology

The specific epithet *latifolia* is derived from two Latin adjectives (*latus* -a -um, meaning 'broad' or 'wide', and *folius –a -um*, meaning 'leaved'. This alludes to the broad leaflets in this species. Vernacular names of this species are Umhlonitshwa (Zulu); Muwatshe, Muvhanzhe (Venda); Bloukeur or Fonteinbos (Afrikaans); Fountainbush (English).

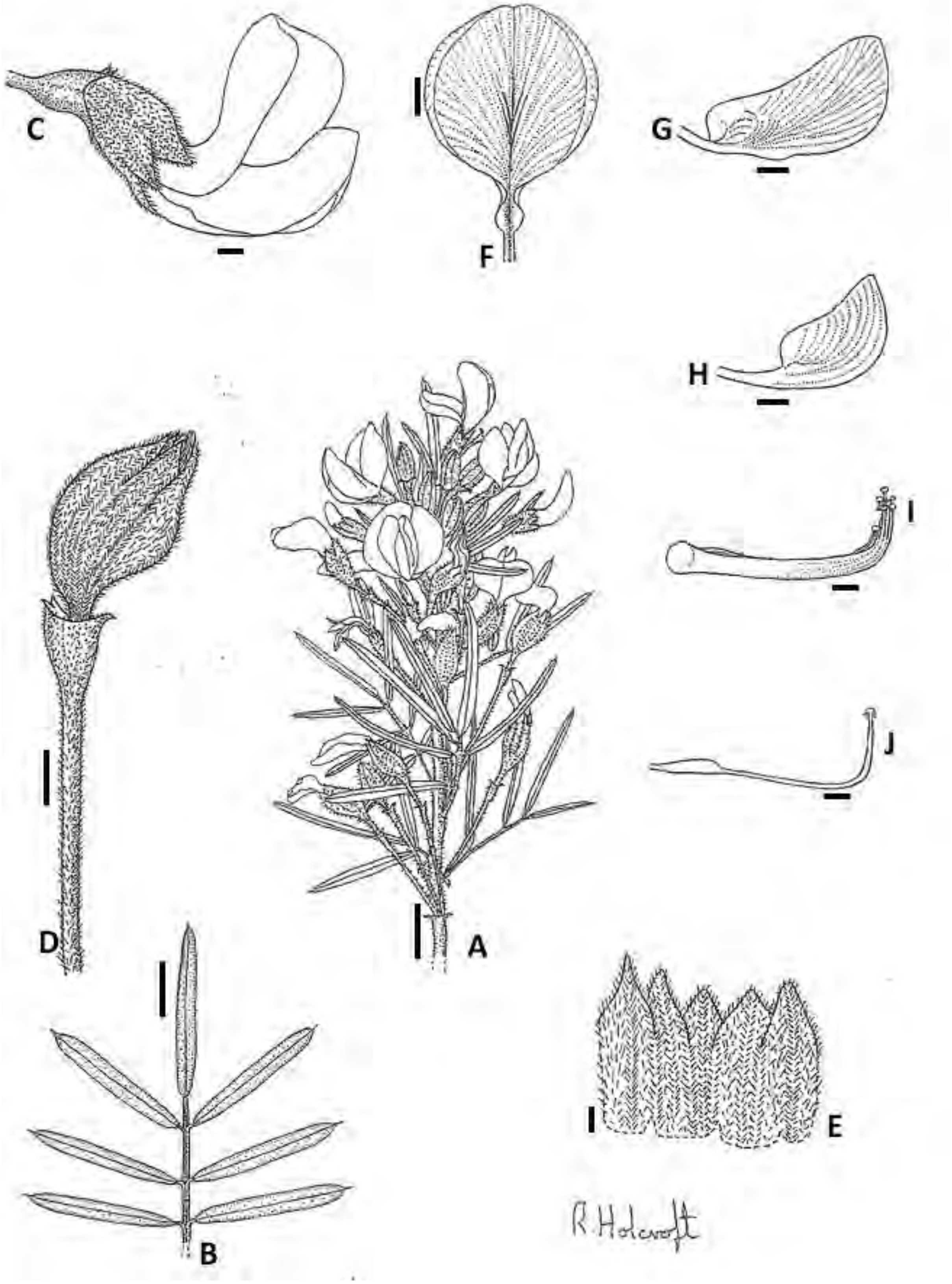

Figure 22. Vegetative and reproductive morphology of *P. latifolia* (A) flowering shoot; (B) 7-foliolate leaf (C) flower; (D) cupulum; (E) calyx opened out with vexillary lobes to the right; (F) front view of standard petal; (G) wing petal; (H) keel; (I) androecium; (J) pistil. Voucher *N. Grobbelaar 2614* (PRE). Scale bars: A, B, D, 1 cm; C, E–J, 1 mm. Artist: R. Holcroft.

4.6.2.14.4. Conservation status

Psoralea latifolia has been examined against the five IUCN criteria of threat and does not qualify for any of the categories. It has a wide distribution range and it is locally abundant in its habitat. It is therefore assigned the status of least concern (LC) (IUCN 2012a, 2012b).

4.6.2.14.5. Specimens studied

South Africa. GAUTENG: Brummeria, Pretoria, cultivated, 2 Sep 1940, *Verdoorn I.C. s.n.* (K, PRE281144). LIMPOPO: **2229 (Pontdrif)**: Scot Farm, 6 km E of Die Soutpan (-CD), 30 Jun 1978, *Scott L. 239* (BLFU); ± 12 km Northwest of Louis Trichardt on high peak of Hangklip mountain (-DD), 24 Mar 1994, *Rossouw G.L. 145* (PRE); Hangklip Peak (-DD), 21 Aug 1929, *Galpin E.E. 14005* (PRE); Wylies Poort near Verwoerd Tunnel (-DD), 25 May 1962, *Hardy D. 952* (PRE); 12 May 1932, *Pole Evans I.B. s.n.* (PRE); 5 m W of Wylies Poort (-DD), Jun 1930, *Hutchinson J. & Gillett J.B. 4403* (K, PRE). **2230 (Messina)**: Woodbush (-CC), Aug 1909, *Jenkins T.J. 7194* (PRE); Entabeni Forest Reserve (-CD), 10 Sep 1947, *Codd L.E.W. 3048* (K, PRE); Entabeni (-CD), Nov 1931, *Obermeyer A.M. 1071* (PRE); 19 Jul 1930, *Hutchinson J. & Gillett J.B. 4192* (K); 3 Jul 1971, *Weintraub D. s.n.* (PRE); Matshavhawe, on road to Khunda (-CC), 5 Sep 2006, *Mabatha F.W. 2145* (K): Thononda, near the Crown (-CD), 5 Oct 1981, *Van Wyk A.E. and Theron G.K. 4554* (PRE); Tate Vondo Forest Reserve, Sibasa Distribution (-CD), 29 Jul 1977, *Hemm G. 199* (PRE). **2329 (Pietersburg):** Soutpansberg (-BA), 27 Jul 1934, *Galpin E.E. 15144* (PRE); 23 Aug 1934, *Galpin E.E. 32218* (BOL); Top of Soutpansberg (-BA), 17 Sep 1934, *Pole Evans I.B. 3717* (K, PRE); Devil's Gulley (-BA), 17 Aug 1935, *Galpin E.E. s.n.* (K, BOL, PRE); Mountain Inn, Soutpansberg (-BB), 25 Feb 1957, *Meeuse A.D.J. 9768* (K, PRE); Louis Trichardt (-BB), 23 May 1927, *Young R.G.N. s.n.* (PRE); Farm Rustfontein, 9 m E of Louis Trichardt (-BB), 6 Jul 1955, *Schlieben H.J. 7696* (K, PRE). **2428 (Nylstroom):** Naboomspruit (now Mookgophong) (-BC), Apr 1932, *Murray s.n.* (TCD); Naaupoort farm, 5 km from Boekenhout Siding, 25 Dec 1931, *Galpin E.E. 11668* (K, PRE). MPUMALANGA: **2430 (Pilgrim's Rest)**: 634 LJ, Wolkberg (-AB), 24 Apr, *Muller P.J. & Scheepers J.C. 190* (K, PRE); Lekgalameetse Nature Reserve (-AB), 5 Aug 1984, *Stahlmans M. 109* (PRE); Bo-op Mariepskop naby SAUK-Stasie (-DB), 2 Dec 1981, *Grobbelaar N. 2614* (PRE); Mariepskop, (-DB), 21 Sep 1978, *Zambatis N. 813* (PRE); 17 Jan 1959, *Werdermann E. & Oberdieck H.D. 1890* (B, K, PRE); 10 Apr 1958, *Van der Schijff H.P. 4379* (PRE), 27 May 1960, *4482* (K, PRE); 18 Jan 1969, *Hilliard O. M. & Burtt B.L. 5977* (K, PRE); 10 Jan 1964, *Bos J.J. 1027* (NBG); Aug 1923, *Keet J.D.M. 1113* (PRE); 17 Nov 1958, *Killick, D.J.B. & Strey R.G 2417* (K, PRE); Mahalaholo near Mariepskop (-DB), 13 Aug 1981, *Oliver E.G.H. 7670* (PRE);

God's Window (-DC), 6 Aug 1966, *Davidson R.L. & Mogg A.O. D. 32916* (PRE); Morgenzon Nature Reserve (-DC), 1 May 1980, *Reid C.13* (PRE); Ohrigstad Dam Nature Reserve (-DC), 29 Aug 1973, *Jacobsen N.R. 2962* (PRE); 7 Jul 1978, *Edwards D. 4059* (K, PRE); Mt. Sheba, Pilgrims Rest (-DC), 12 Jul 1972, *Boucher C. 1864* (NBG); Nov 1915, *Rogers F.A. 14911* (K); Sep 1915, *Rogers F.A. 14702* (PRE), Jul 1919, *Rogers F.A. 23110* (PRE), Sep 1919 (PRE); May 1971, *Jones 64* (PRE); Mt. Sheba Nature Reserve (-DD), 12 Dec 1972, *Coetzee 1456* (PRE); Aug 1976, *Kerfoot O. 791*5 (J, MO); Kowyn's Pass (-DD), 24 Mar 1960, *Codd L.E.W. 10055* (K, PRE); 10 Mar 1937, *Galpin E.E. s.n.* (BOL, PRE); 13 Oct 1937, *Galpin E.E. 14422* (BOL, K, PRE); *Liebenberg L.C. 2969* (PRE); Graskop (-DD), 18 Jul 1935, *Nel G.C. 133* (PRE). MPUMALANGA: **2529 (Witbank):** Middelberg (-CB), 11 Oct 1954, *Prosser L.N. 1894* (K, PRE); Tweefontein, Loskop Dam (-DC), 14 Oct 1967, *Theron J.J. 1574* (PRE). **2530 (Lydenburg)**: 16 km from Sabie to White River (-BB), 29 Oct 1981, *Schrire B.D. 666* (NH); 12.5 m SE of Lydenberg (-BB), 5 Jul 1966, *Morris J.W. 15* (K, PRE); Near Mac Falls (-BB), Nov 1874, *McLea J.M. 3017* (K, BOL); 16 km from Sabie to White river (-BB), 29 Oct 1981, *Schrire B. 666* (PRE); Witklip, White River (-BB), 18 May 1973, *Kluge J. 20* (PRE); Uitsoek Forestry Station (-BC), 21 Dec 1971, *Howe B.J. 81* (PRE); Mount Anderson, Lydenburg, Transvaal (-BD), 1 May 1973, *Drummond J.M. 44* (NBG); 7 May 1980, *Mohle 339* (PRE); Elandshoogte, Waterval Boven region, Emgwenya (-CA), 23 May 1978, *Cooper K.H. 219* (PRE); Waterval Boven (-BA), May 1907, *Gough 3263* (PRE); 21 km from Machadodorp (eNtokozweni) to Badplaas (-CD), 8 Mar 1989, *Schrire B. 2373* (K, PRE); Buffelskloof Nature Reserve (-DA), 8 Oct 2011, *Muasya A.M. & Stirton C.H.* 6113, 9 Oct 2011, *Muasya A.M. & Stirton C.H. 6138* (BOL); Elandshoek (-DA), 26 Sep 1906, *Rogers F.A. 2586* (PRE); Starvation Creek Nature Reserve (-DA), 10 Aug 1979, Kluge R. 1916 (PRE); Amajuba Mt. near Nelspruit (-DA), 28 Dec 1933, *Liebenberg L.C. 3098* (PRE); Kaapsche Hoop (-DB), 11 Jan 1924, *Pole Evans I.B. 991* (PRE); Aug 1915, *Wager H.A. s.n.*(PRE). **2531 (Komatipoort)**: Endahwin Forest, 16 miles (26 km) East of Barberton (-CC), *van Jaarsveld E.J. 3* (NBG); Barberton (-CC), Jul 1889, *Galpin E.E. 401* (PRE); Rimers Creek, Barberton (-CC), 3 Jul 1926, *Potts G. 3658* (PRE); Oct 1914, *Rogers F.A 1401*5 (K, PRE); Jan 1907, *Thorncroft G. 5118* (PRE); Endahwin Forest, 16 miles (26 km) E of Barberton (-CD), 6 Sep 1974, *Van Jaarsveld E. 3* (K, NBG, PRE). FREE STATE: **2827 (Senekal):** Hammonia (-DB), 7 Feb 1911, *Dodd B.H. 8021* (PRE). KWAZULU-NATAL: **2930 (Pietermaritzburg):** Port Natal (-CC), *Sanderson J. 55* (TCD). **3030 (Port Shepstone)**: Beacon Hill West (-CD), 15 Jul 1976, *Van Wyk A.E. 1*686 (PRE). **3130 (Port Edward):** Umtamvuna Forest Reserve (-AA), 20 Jul 1971, *Ward C.J. 7161* (PRE); Mtentu (-AA), 15 May 1969, *Strey R.G. 8637* (K, PRE); Buttress Vlei (-AA), *Abbott A. 3130* (K, PCE), Mkambathi Nature Reserve (-AC), *Hutchings A.D. & Plumstead E.P. 1371* (KEI). EASTERN CAPE: **3127 (Lady Frere):** Cala, Eastern Cape (-CA), 9 Oct 1961, *Esterhuysen E. 29187* (K, PRE); Hills near Encobo (-DC), 8 Oct 1961, *Esterhuysen E. 29154* (BOL, K). **3128 (Umtata):** Gulandoda Mountain (-AA), 10 Oct 1961, *Esterhuysen E. 29225* (K, BOL); Misty Mount, Libode (-AC), 31 Aug 1982, *Johnson 735* (KEI). **3129 (Port St. Johns):** Lupatana (-BD), 10 Nov 1981, *Cawe S. 16* (KEI); Mkambathi River (-BD), 3 May 1985, *Shackleton C.M. 278* (KEI); 2.3 km N of Mzintlava River Mouth (-DA), 26 Aug 1976, *Venter S. & Vorster P. 183* (K, PRE); 2.5 m W of Ndindini Store (-DD), 10 Sep 1956, *Codd L.E.W. 9731* (PRE); Ndindini Stream, between Lusikisiki and Lupatana (-BD), *Van Wyk A.E. 1594* (PRE, PRU). **3226 (Fort Beaufort)**: Katrivier (-DA), *Ecklon C.F. & Zeyher C.L. 1514* (SAM, NBG); Katberg (-BC), 19 Oct 1970, *Clarke V.R. 65* (PRE); Stockenstroom (-DB), Nov 1926, *Lotsy J.P. & Goddijn W.A. 91* (L); Hogsback (-DB), Feb 1942, *Archibald E. s.n.* (K); 25 Oct 1921, *Stayner F.A. 11* (PRE); 29 Sep 1976, *Stirton C.H. 6239* (K, PRE); *Rattray G. 34* (PRE); *Leeman A.C. 7* (BOL, GRA); 24 Dec 1926, *Grant 2781* (BOL, PRE); 18 Oct 1968, *Dahlstrand K.A. 1556* (GRA, PRE), 14 Jan 1971, *2645* (PRE); Oct 1950, *Leighton F.M. 3082* (PRE); Nov 1926, *Dyer R.A. 719* (PRE); Mitchell's Pass (-DB), 28 Oct 1946, *Esterhuysen E. 14551* (BOL); Below Wolfridge lookout (-DB), 9 Oct 1981, *Phillipson R.B. 340* (K). **3227 (Stutterheim)**: Evelyn Valley, Stutterheim (-CB), 13 Jan 1947, *Compton R.H. 19174* (NBG); Kologha Forests (-CB), 9 Sep 1942, *Acocks J.P.H. 9046* (PRE); Kubisi Forest (-CB), 19 Jul 1972, *Osborne 81* (GRA). **Without precise locality:** Macoutie, Jul 1917, *Breijer H.G. 18388* (PRE); Vanetzi Poort, 2 Mar 1946, *Gerstner 6064* (PRE); Eastern frontier, Nov 1880, *Hutton H. s.n.* (TCD); Rietfontein, *Leendertz R. 8381* (L. PRE); Budworth Farm, 9 Jul 1958, *Mogg A.O.D. 28254* (PRE); Kaatjieskraal, 3 Apr 1914, *Burchell W.R. 5266* (K); 14 Nov 1910, *Stewart M.M. s.n.* (K); Cyprus, Letaba, 22 Jan 1961, *Penny A.* 145 (K, PRE).

Swaziland. **2631 (Siteki)**: Mbuluzi River (-AA), 12 Sep 1972, *Prior J. 57* (PRE); 18 km NNW of Mbabane, E of Ngwenya Escarpment (-AA), 19 Sep 1977, *Prior J. s.n.* (K); Mbabane Dist. Hlotwani Mountains (-AC), 22 Jan 1957, *Compton R.H. 26506* (NBG); Stroma, Mbabane (-AC), 10 Jun 1955, *Compton R.H. 25139*, 26 Jul 1947, *19774* (NBG, PRE); Duiker Bush (-AC), 24 May 1956, *Compton R.H. 25950*; Mbabane (-AC), 19 Jun 1911, *Burtt Davy J. 10584* (PRE); Stegi, Forest way by stream (-BD), 23 Jul 1947, *Compton R.H. 19765* (BOL, NBG); 5 km SE Stegi, 28 Aug 1978, *Prior J. 161* (PRE).

Mozambique. **2531 (Komatipoort):** Monte M'ponduine, Namaacha (-AA), 25 Aug 1980, *Schäfer P.A. 7197* (BM, K, LMU, MO); M'ponduine Hill (-AA), 17 Aug 1925, *Pedro 717* (PRE). **2532 (Lourenco Marques):** Proximo da Cascata da Namaacha (-CC), 7 Dec 1942, *Medonça F.A. 1698* (MO). **2632 (Bela Vista):** Namaacha (-AA), 30 Jun 1961, *Balsinhas A, 500* (PRE). **Without precise locality:** Moçambique, 13 Jun 1978, *de Koning J. 7089* (NBG).

4.6.2.15. *Psoralea speciosa* Eckl. & Zeyh., Enum. 224 (1836); Walp., Repert. 1: 656 (1842); Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Western Cape, Oudtshoorn (3322): "Inter frutices (altit. II) ad Keureboomrivier prope Langkloof (Georg.)" [Among bushes (altit. 2) along the Keureboomrivier near Langkloof] –CD, *Ecklon & Zeyher* 1517 (S!, lecto. here designated; S!, isolecto.). Note: Only two herbarium sheets have been found of this species. Of the two sheets in S one is depauperate (S08-12035). We have chosen the more robust specimen (S14-13322) as the lectotype.

A small to large, robust, colonial, reseeding shrub up to 2.5 m tall, *Stems* erect, 1−3, branching at or near the base, stiff and arching, blackish brown, pubescent; branches short, stiffly erect, rigid. *Seasonal shoots* glabrous, dark green. *Leaves* 5-foliolate, imparipinnate, distributed along the branches but on older plants occur only at the tips, rounded, petiolate, papillose on the margin, glabrous, channelled on both surfaces; leaflets 10–22×0.3–0.4 mm, but longer (18–22 mm) at base of seasonal shoots, linear lanceolate, terminal leaflet shortest, basal pair longest, three uppermost digitately inserted, rarely pinnately; petioles 2–4 mm long, shorter than terminal leaflets; rachis 3−7 mm long; stipules 3×1.5 mm caducous, triangular, puberulent, glandular. *Inflorescences* axillary, borne vertically on short vernal shoots off horizontal branches along the length of seasonal shoots but without leafy extension, pseudo-capitate, lax, pedunculate, 4−5 flowers per axil, pedicellate; peduncles stout and rigid, 1–3 mm long, shorter than the subtending leaf; cupulum trilobed, 2−3 mm long, glabrous, one of the lobes scarcely developed, triangular, inner bracts absent; pedicels 1–2 mm long. *Flowers* 17−18 mm long, deep blue or indigo-blue, each flower subtended by a pair of small cucullate bracts. *Calyx* 10−12 mm long, tube 5–8×6 mm, glabrous but fringed with dark hairs, nigro-punctate, fleshy, yellowish, accrescent, persistent after seed shed, densely glandular; lobes unequally developed, shorter than the tube, broadly triangular, carinal lobe much broader, vexillar lobes fused 2 mm above the tube, incurving, acuminate, inner face covered with soft white hairs. *Standard petal* 14–15×14–16 mm, very broadly ovate, deep blue or indigo-blue with large white M-shaped nectar guide, central purple flash absent, veins darker than blade, appendages present; claw 2–4 mm long. *Wing petals* 12–13×4–5 mm, same colour as standard, completely covering the keel, sculpturing present, upper and lower basal comprised of 2−4 arching lamellae, pocket well developed, auricle swollen, folded along the mid-line; claw 4–5 mm long. *Keel* 11×4 mm, acute at the apex; claw 5–6 mm long. *Androecium* 12 mm long, tenth stamen lightly attached, fenestrate. *Pistil* 11 mm long, ovary 1.5−2 mm long, glandular, style straight 6 mm long, then curved, entasis well developed at mid-point, height of curvature 5 mm long, stigma capitate, penicillate. *Fruits* oblong, 5–6×2.5–3.0 mm. *Seeds* solitary, 3–5×2.5 mm, dark brown to black (Figure 23).

4.6.2.15.1. Diagnostic characters

Psoralea speciosa is easily identified in the *P. pinnata* complex by a combination of features: arching stems with short erect branches; short, tightly packed, fleshy, congested leaflets; large flowers with large white M-shaped nectar guide without a central purple flash; and the largest accrescent calyces in the group. It is similar to *P. kougaensis* in having larger calyces but differs in its arching large multi-branched, reseeding shrubby habit (versus erect, columnar, multi-stemmed, branchless or few-branched resprouting shrub), 5-foliolate, fleshy, rounder, glabrous leaves without raised glands (versus 5(7)-foliolate, with mostly curving distinctly warty leaves), minute subulate stipules (versus large conspicuous elongated stipules), standard deep blue or indigo-blue with large white M-shaped nectar guide, central purple flash absent (versus dark mauve fading towards margins with a broad triangular purple central nectar guide above the swollen appendages from which arises a very dark erect flash tapering to the apex); wing petals deep blue or indigo-blue (versus white to pale mauve), calyces fleshy, yellowish, accrescent, glabrous but fringed with dark hairs (versus pale green, not accrescent, glabrous or sparsely pubescent).

4.6.2.15.2. Distribution and habitat

Psoralea speciosa is a montane species that extends from the Langeberg Mountains in the west to the Kouga mountains and as far-east as Humansdorp (Figure 24). It occurs on hillsides and to a lesser extent along streams in mountain fynbos between 600–1200 m elevations. It has been found in the North Outeniqua Sandstone Fynbos, South Outeniqua Sandstone Fynbos, North Swartberg Sandstone Fynbos, Montagu Shale Renosterveld, and Willowmore Gwarrieveld vegetation types (FFs 18, FFs 19, FFs 23, FRs 7, SKv 12) (Rebelo *et al.* 2006; Rutherford *et al.* 2006).

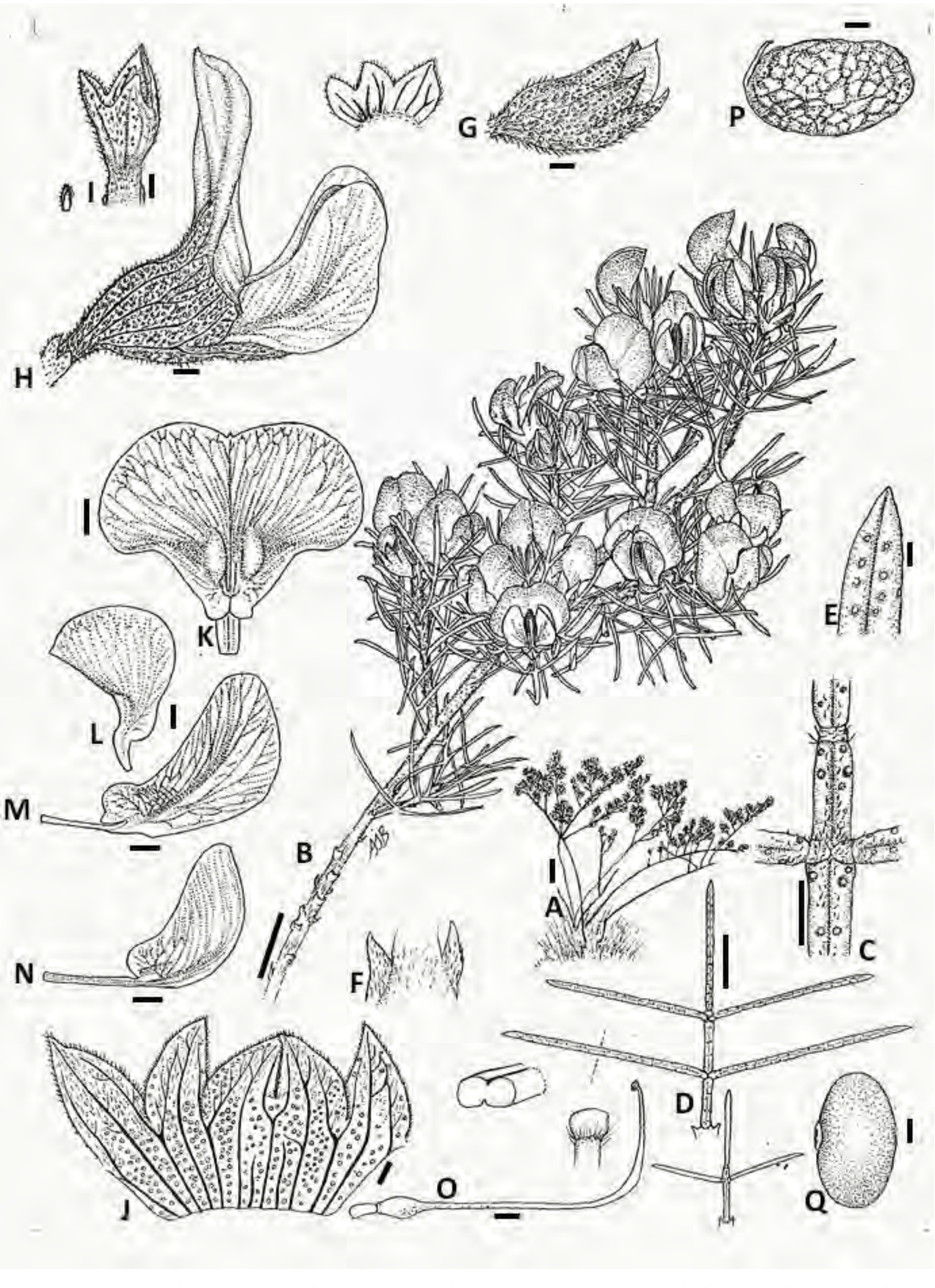

Figure 23. Vegetative and reproductive morphology of *P. speciosa* (A) habit; (B) flowering shoot; (C) leaf insertion; (D) 5-foliolate leaf; (E) leaflet; (F) stipule; (G) bud; (H) flower; (I) cupulum; (J) calyx opened out with vexillary lobes to the right; (K) front view of standard petal; (L) side view of standard petal; (M) wing petal; (N) keel; (O) pistil; (P) fruit; (Q) seed. Voucher *C.F. Ecklon & C. Zeyher s.n.* (PRE) Scale bars: A−F, 1 cm; G–Q, 1 mm. Artist: Angela Beaumont.

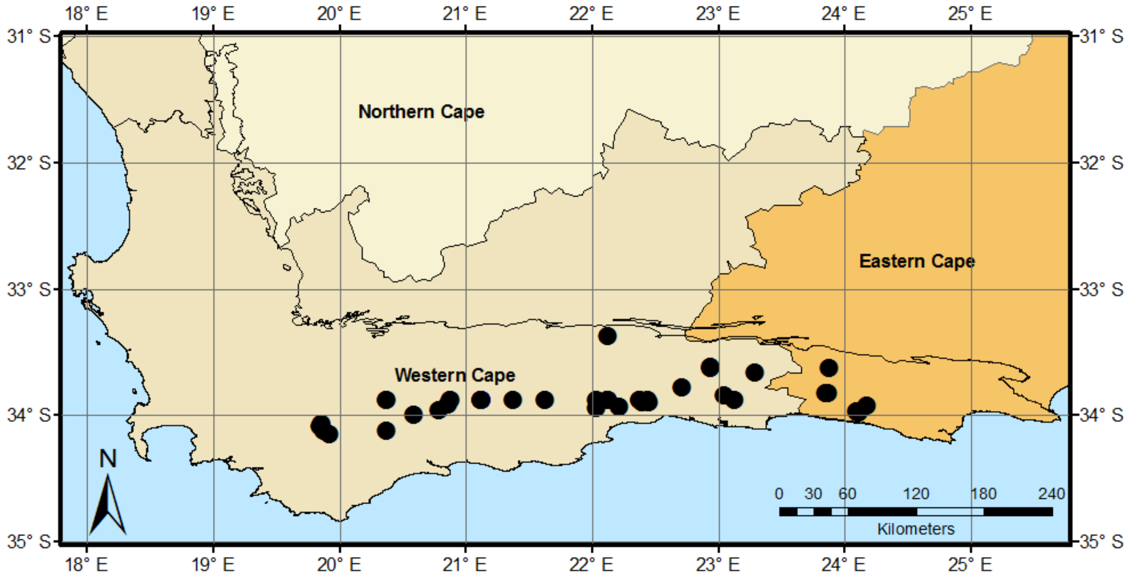

Figure 24. Known distribution of *P. speciosa*.

4.6.2.15.3. Etymology

The specific epithet *speciosa* is derived from the Latin adjective *speciosus -a –um* (beautiful, splendid, showy'); an apt name as the plants in full flower are very attractive.

4.6.2.15.4. Conservation status

Psoralea speciosa is widespread and abundant in its distribution, especially in protected areas, and does not qualify for any IUCN criteria of threat. It is therefore assigned a status of least concern (LC).

4.6.2.15.5. Specimens studied

South Africa. WESTERN CAPE: **3320 (Montagu)**: Slopes of the Dwarsberg, Boosmansbos Wilderness Area, Swellendam (-CB); Southern side, north of Grootvadersbosch, Boosmansbos Wilderness area, Langeberg (-CC), 18 Sep 1993, *Strid P.A.K. A+B 37981* (NBG), 13 Dec 2007, *Muasya A.M. & Stirton C.H. 3458* (BOL); State forest, Marloth Nature Reserve (-CD), 5 Aug 1972, *Haynes R.A. 662* (PRE, NBG); Swellendam, alongside trail to Boskloof (-DC), 15 Oct 1988, *Du Plessis J.L. 89* (NBG); Slope of Vaalrivierkloof (-DD), *Forsyth G. 82* (K, NBG); Slopes of the Dwarsberg, Boosmansbos Wilderness Area (-DD), 13 Dec 2007, *Muasya A.M. & Stirton C.H. 3458* (BOL); Lemoenshoek Peak, Langeberg (-DD), 7 Dec 1981, *Stirton C.H., Rourke J. & Esterhuysen E. 10201* (PRE). **3321 (Ladismith)**: Grootvadersbosch State Forest, Garcia's Pass (-CB), 23 Dec 1939, *Barker W.Z. 522* (NBG); Sep 1908, *Phillips E.P. 369* (K); Steep track to East of toll house, Garcia's Pass (-CC), 14 Sep 1981, *Van Wyk C.M. 689* (K, NBG); Halfway up Garcia's Pass (-CC), 5 Oct 1981, *Stirton C.H. 9709* (K, L, PRE); Top of Garcia's Pass (-CC), 5 Oct 1981, *Stirton C.H. 9713* (K, PRE); Oct 10 Nov 1986, *Stirton C.H. and Žantovská J. Z. 11542* (K, NBG). The Oaks, Gysmanshoek Pass, Riversdale (-CC); Langeberg (-CC), 20 Aug 1993, *Rourke J.P. 2009* (K, NBG, PRE), 16 Sep 1981, *Van Wyk C.M. 710* (NBG); Langeberg East, Compton 15 (Paardeberg) (-CD), 11 Aug 1977, *Haynes R.H. 1409* (NBG, PRE); Middle of Garcia's Pass, above road, Riversdale region (-CD), *Bremer K. 229* (PRE). **3322 (Oudtshoorn)**: Swartberg Pass (-AC), 30 Sep 1962, *Walters I.B. 725* (NBG); Swartberg Pass, Oudtshoorn (-AC), 30 Sep 1962, *Walters T.B. 128* (NBG); Ruyterskop, Mossel Bay (-CC), 26 Nov 1949, *Compton R.H. 21832* (NBG); 3 ml. (5 km) from 8 Bells – on Robinson Pass, Mossel Bay (-CC), 10 October 1953, *Heginbotham M. 221* (NBG); 3 km S of Robinson Pass (-CC), 22 Oct 1989, *Greuter W. 22132* (PRE); 22 Oct 1962, *Killick D.J.B. 3471* (K, PRE); Robinson Pass (-CC), *Hops B. 53* (BOL); 25 Oct 1932, *Barker W.F. 59* (K); Jonkersberg Plantation (-CC), Aug 1932, *Fourcade G.F. 4780* (NBG); Kogmanskloof (-CC), 1 Jan 2008, *Muasya A.M. & Stirton C.H. 3557* (BOL); Ruitersbosch, Mossel Bay Division (-CC), 13 Sep 1951, *van Niekerk G. 37* (BOL, PRE); 4 Ways Cross, Outeniqua Pass (-CD), 3 Jan 2008, *Muasya A.M. & Stirton C.H. 3610* (BOL); Outeniqua Pass (-CD), 2 Dec 2009, *Muasya A.M. & Stirton C.H. 5085* (BOL); start of pass on road to Oudtshoorn, Outeniqua mountains (-CD), 5 Dec 1986, *Germishuizen G. 4267* (PRE); Saasveld Forestry Station (-CD), 16 Sep 1969, *Dahlstrand K.A. 1281* (NBG); Jonkersberg, Outeniquas, near George (-CD), 4 Dec 1951, *Esterhuysen E. 19401* (NBG); Hopsvallei Farm, Tweewatersberg, Outeniqua Mountains, 2 Dec 2009, *Muasya A.M. & Stirton C.H. 5065* (BOL); Outeniqua Pass (-CD), 12 Nov 1986, *Stirton C.H. 11590* (PRE); Cradock's Berg (Post Berg), near George (-CD), 12 Sep 1914, *Burchell W. 5939* (K); Oct 1930, *Fourcade G.H. s.n.* (BOL); northern entrance to Montagu Pass (-CD), 4 Nov 1928, *Hutchinson J. 1181* (BOL, K); 4 Nov 1928, *Gillett J.B. 2047* (NBG); Mannetjiesberg, Kammanassie, Uniondale District (-DB), Nov 1941, *Esterhuysen E. 6450* (BOL); Lemoenshoek Peak (-DD), 7 Nov 1981, *Stirton C.H. 10210* (K, NU). **3323 (Willowmore)**: Hoopsberg (-CC), 6 Nov 1941, *Esterhuysen E. 6539* (PRE); Buffelsnek Forest Station (-CC), 26 Sep 1978, *Hugo L. 1306* (NBG, PRE). **3419 (Caledon)**: Ridges and slopes on south side Riviersonderend Peak (-BB), 2 Sep 1951, *Esterhuysen E. 18771* (NBG); Pilaarkop, mountains E of Lindeshof (-BB), 17 Nov 1965, *Esterhuysen E. 31401* (BOL); Riviersonderend mountains (-BB), 1940, *Stokoe T.P. 7390* (BOL); southern side of Peak, R.T.Z., Sep 1943, *Stokoe T.P. 8970* (BOL); 19 Oct 1945, *Stokoe T.P. 9281* (BOL); Oct 1950, *Stokoe T.P. s.n.* (PRE, NBG, SAM 55807). **3420 (Bredasdorp)**: Marloth Wildflower Reserve, below 10 o'clock Mountain (-AB), 26 Sep 1952, *Wurts T.M. 373* (BOL, NBG). **3423 (Knysna):** Reservaat, Knysna (-AA), *Geldenhuys G.A. 110* (PRE); 20 m (32 km) N of

Knysna (-CC), 25 Aug 1956, *Theron 2056* (GRA, K, PRE). EASTERN CAPE: **3324 (Steytlerville):** 8 km from Kareedouw to Joubertina (-CC), 5 Oct 1976, *Stirton C.H. 6531* (K, PRE); Joubertina (-CC), 24 Nov 1941, *Esterhuysen E. 6889* (PRE); Mountain slope, above Witte Els Bosch (−CC), 16 Nov 1941, *Esterhuysen E. 6796* (BOL); Witte Els Bosch (−CC), Sep 1920, *Fourcade G.F. 923* (BOL, GRA); Joubertina (-DD), 24 Nov 1941, *Esterhuysen E. 6889* (PRE).

4.6.2.16. *Psoralea ivumba* C.H. Stirt., A. Bello & Muasya *sp. nov.* Type**:** South Africa, Western Cape, Worcester (3319): R43 T-Junction of Tulbagh and Wolseley-Ceres roads (-AC), 9 Nov 2011, *Stirton C.H. 13468* (BOL!. holo.; NBG!, PRE! iso.).

A virgate slender shrub up to 2.5 m tall, reseeder. *Stems* erect, single, branching from lower portion, arching to erect, upper branches spreading, brownish green to tan with storied white lenticels when young, becoming rough and grey with age, branch scars prominent, branches and twigs angular, ridged. *Seasonal shoots* glabrous to glabrescent, bright green. *Leaves* 7−11-foliolate, imparipinnate, softly pilose, crowded at the end of bare branches on old shoots, or distributed along the branches on younger shoots, patent, petiolate; leaflets 18–50×0.2–0.3 mm, filiform and straight to narrowly linear and curved, somewhat flattened, adaxial surface more convex, abaxial surface grooved, apex acuminate, dark green, glandular, cavities slightly sunken in fresh state, terminal leaflet second longest, basal pair longer or equal to terminal, three uppermost digitately inserted; petioles 3–7 mm long, shorter than terminal leaflet; rachis 15−18 mm long; stipules 3–4×1–2 mm, persistent, shorter than petiole, narrowly triangular, fused for half their length on the stalked ridge overlapping the flanges, teeth caducous, erect, bases persistent, becoming woody with age, margins out curled, subulate, quickly senescent. *Inflorescences* borne in uppermost axils of seasonal shoots, pseudo-capitate, partly hidden within the subtended leaves, congested, 5–6 flowers per axil, pedunculate, pedicel absent; peduncles stout and rigid, 6–15 mm long, densely white long-haired; cupulum bilobed, lobes unequally developed, lower lobe partly cleft, carinal lobe longer, 3 mm long, tube 2 mm long, subulate, acuminate, sparsely white hairy, margins ciliate with few black hairs. *Flowers* 15−16 mm long, bright blue to purple, very strongly scented, subtended by a 2 mm cucullate ciliate bract, clasping the oldest flower. *Calyx* 8−9 mm long, tube 5–6×5 mm, half length of flower, papery, shaggy white-haired, pale green; lobes unequally developed, pale yellowish green, shorter than the tube, carinal longest, 4 mm long, ovate-acute, vexillar teeth fused for about one third their length; margins and inner face of lobe with few black-hairs, inner face of tube glabrous; densely glandular, glands constant in size, whitish. *Standard petal* 11–12×9–10 mm, very broadly ovate, purple in upper half becoming mauve towards margins with a white M-shaped nectar guide and a single 2 mm long purple flash situated between and above the free swollen appendages at the top rim of the claw, auricles present, swollen, veins coloured; claw 2–3 mm long, channelled, sharply recurved at anthesis. *Wing petals* 10–11×5−6 mm, longer than keel petals, pale mauve, blade folded and puckered along its mid-line; sculpturing present, basal, comprised of 2−4 arcuate rows of 9−11 transcostal lamellae; claw 3–4 mm long. *Keel* 8–9×2–4 mm, blade ridged for 3 mm from base, white apex broadly triangular; claw 3–4 mm long. *Androecium* 10 mm long, tenth stamen lightly attached. *Pistil* 10 mm long, stipitate, ovary 2 mm long, glabrous with club-shaped glands near apex only, style 6 mm long straight to the point of flexure then curved upwards, widest beyond point of flexure, height of curvature of 2.5 mm, stigma capitate. *Fruits* unknown. *Seeds* unknown (Figure 25).

4.6.2.16.1. Diagnostic characters

Psoralea ivumba is similar to *P. pinnata* sensu strict in growth form, but differs in being a virgate shrub up to 2.5 m tall (versus tall shrub to small tree up to 5 m tall), with filiform, softly pilose spreading leaves (versus villoso-pubescent), grooved, ridged, bright green seasonal shoots (versus round, smooth, greenish-brown seasonal shoots), inflorescences congested into pseudo-capitate heads terminating seasonal flowering shoots (versus inflorescences pseudo-capitate and pseudo-spicate in upper axils of seasonal shoots with leafy extensions), peduncles present, 6–7 mm long (versus peduncles absent), larger, 15–16 mm long, intensely scented, bright blue to purple, stalked flowers (versus 13–14 mm long, scentless or faintly scented, pale mauve to pale blue, sessile flowers), shaggy white-haired calyces with four teeth black ciliate (versus glabrescent with white to black haired or a mixture of both), bilobed cupulum (versus trilobed cupulum), and standard very broadly ovate, veins hyaline (versus obovate standard, veins coloured).

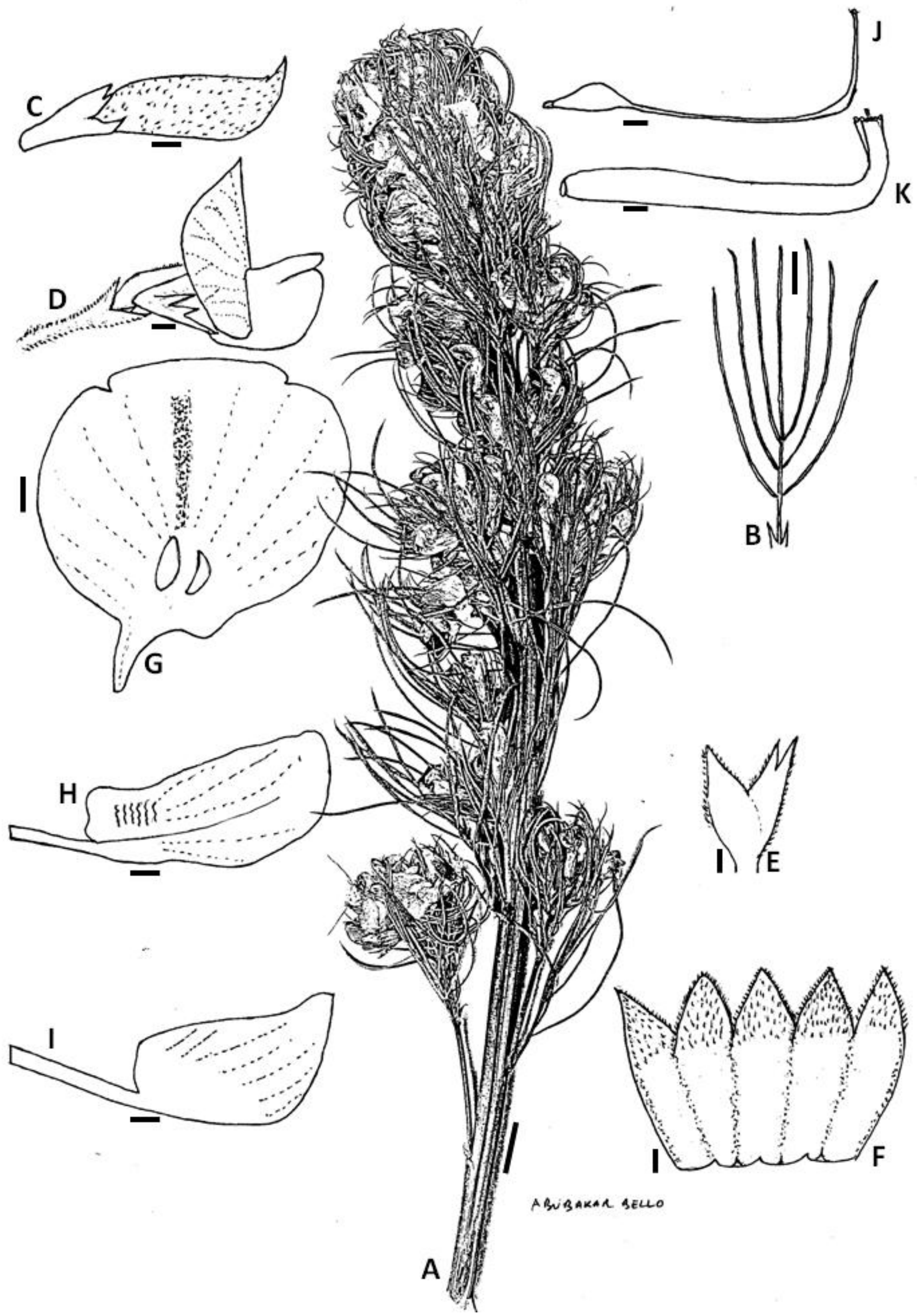

Figure 25. Vegetative and reproductive morphology of *P. ivumba* (A) flowering shoot; (B) 9-foliolate leaf (C) bud; (D) flower; (E) cupulum; (F) calyx opened out with vexillary lobes to the right; (G) front view of standard petal; (H) wing petal; (I) keel; (J) pistil; (K) androecium. Voucher *A. Bello 163* (BOL). Scale bars: A−B 1 cm; C–K, 1 mm. Artist: Abubakar Bello.

4.6.2.16.2. Distribution and habitat

Psoralea ivumba is endemic to the South Western Cape Province in South Africa and extends from Wolseley and Romansrivier southwards to Somerset West and Sir Lowry's Pass then westwards towards Hermanus and Caledon (Figure 26). It has been found in the Hangklip Sand Fynbos, Hawequas Sandstone Fynbos, Overberg Sandstone Fynbos and South Sonderend Sandstone Fynbos (FFd 6, FFs 10, FFs 12 and FFs 14) (Rebelo *et al.* 2006; Rutherford *et al.* 2006). Where conditions are ideal, the species forms large colonies in boulder strewn landscapes in fynbos on Table Mountain Sandstone but in some areas they occur in small patches. It does not particularly favour seepages and damp areas. It is pollinated primarily by Xylocopid bees. The population at Theewaterskloof Dam comprises thousands of individuals.

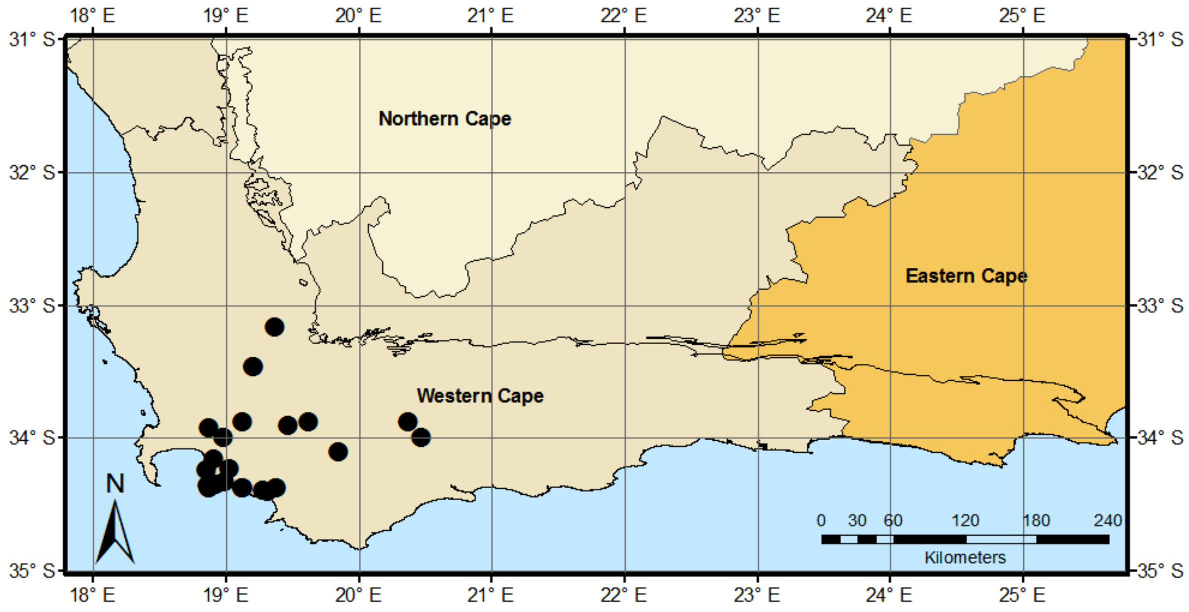

Figure 26. Known distribution of *P. ivumba*.

4.6.2.16.3. Etymology

The specific epithet *ivumba* is derived from the Xhosa phrase for aroma (*ivumba elimnandi*) and refers to the intensely sweet smelling flowers - a characteristic it shares with the dissimilar species *P. fleta* C.H. Stirt.

4.6.2.15.4. Conservation status

This species is of least concern (LC) as it has a wide distribution range and is abundant in its habitats. It does not fall under any category of threat among the five IUCN criteria (von Staden *et al.* 2009).

4.6.2.16.5. Specimens studied

South Africa. WESTERN CAPE: **3318 (Cape Town):** Stellenbosch (-DD), 12 Oct 1913, *Garside S. s.n.* (K); Jonkershoek, 1 Oct 1958, *Werdermann E. & Oberdieck H.D. 338* (K). **3319 (Worcester)**: Elandsfontein farm (-AC), 22 Oct 1986, *Stirton C.H. 11242* (K, NBG)**;** Purgatory Outspan, road side next to Du Toits river, Theewaterskloof area (-CC), 7 Dec 1979, *Hugo L. 2135* (PRE); Halfway down Franschhoek Pass (-CC), 3 Dec 1981, *Stirton C.H. 10119* (K, NBG); 22 Oct 1986, *Stirton C.H. 11222* (K, NBG); Valley below Jonas Kop (-CD), 29 Nov 2007, *Muasya A.M. & Stirton C.H. 3340* (BOL). **3320 (Montagu)**: Romansrivier (-AC), 22 Oct 1986, *Stirton C.H. 11240* (K); 26 Oct 2014, *Stirton C.H. & Bello A. 14148* (BOL); Farm North Station, 2 km from Heron to Montagu Pass (-CD), 2 Dec 2009, *Muasya A.M. & Stirton C.H. 5079* (BOL). **3418 (Simonstown)**: Near bridge Kogelbaai (-AD), 8 Sep 1969, *Boucher C. 505* (K, NBG); Sir Lowry's Pass (-BB), 28 Nov 1980, *Stirton C.H. 8366* (PRE); Bottom of Sir Lowry's Pass, 25 Nov 1981, *Stirton C.H. 9944* (K); Ernst se Bank (-BB), 6 Oct 1981, *Stirton C.H. 9754* (PRE); Vergelegen Estate, Somerset West (-BB), 15 Oct 2011, *Stirton C.H. 13371* (BOL); Steenbras Dam (-BB), Feb 1918, *Moss C.E. & Rogers F.A. 1553* (K); Plateau, behind Klein Hangklip (-BD), 19 Nov 1980, *Stirton C.H. 8208* (PRE); Harold Porter Botanic Garden, Betty's Bay (-BD), *Ebersohn W. s.n.* (NBG); Betty's Bay (-BD), 11 Dec 1972, *Rycroft H.B. 3154* (NBG). **3419 (Caledon)**: Grabouw, Lebanon (-AA), 9 Nov 1966, *Kruger F.J. 183A* (PRE); Lebanon, Grabouw (-AB), 19 Dec 1966, *Van der Zel D.W. 28* (K); Grabouw, along N2 (-AA), 16 Nov 2011, *Muasya A.M. & Stirton C.H. 6215* (BOL); River Valley, Grabouw (-AA), Dec 1951, *Stokoe T.P.* s.n. (NBG); Vogelgat Private Nature Reserve, Hermanus (-AC), 4 Dec 1980, *Stirton C.H. 8480* (PRE); Kleinmond (-AC), 10 Nov 1962, *Walters I.B. 1118* (NBG); Kleinmond (-AC) 23 Nov 1946, *de Vos M.P. 313* (NBG); Fernkloof Nature Reserve, adjacent to Botanical centre (-AD), 10 Oct 1976, *Woodvine F. 20* (K, MO, PRE); 11 Nov 1976, *Woodvine F. 40* (K, MO); Vogelgat, east of Base Camp (-AD), 2 Dec 1983, *Williams I.J.M. 3524* (PRE); Fernkloof Nature Reserve (-AD), 11 Nov 1976, *Woodvine F. 40* (HER, PRE); Vogelgat East of Base camp (-AD), 2 Dec 1983, *Williams I. 3524* (NBG); Vogelgat Nature Reserve (-AD), 9 Sep 1978, *Williams I. 2564* (PRE, NBG); Onrust, Hermanus Road side (-AD), Dec 1961, *Walters M. 28* (NBG); Top of Sir Lowry's Pass (-BB), 9 Dec 1981, *Stirton C.H. 10248* (K); 6 km from Sir Lowry's Pass to Grabouw (-BB), 9 Dec 1981, *Stirton C.H. 10249* (K); Elim-Stanford road, crest of hill, Stanford side (-DB), 27 Nov 2011, *Stirton C.H. 13582* (BOL); 6 miles E of Caledon (-DD), 5 Dec 1954, *Marais W. 597* (K, PRE); Caledon (-DD), Oct 1846, *Prior A. s.n.* (K); Caledon (-DD), *Pappe s.n.* (K); Kleinmond (-DD), 24 Oct 1958, *Werdermann E. & Oberdieck H.D. 720* (K, PRE).

4.6.2.17. *Psoralea affinis* Eckl. & Zeyh., Enum.: 224 (1836); Walp. in Linnaea 13: 512 (1839); Walp., Repert 1: 655 (1842); Forbes in Bothalia 3: 126 (1930); Bond & Goldblatt, Pl. Cape Flora: 296 (1984); Gibbs Russell *et al.* Mem. Bot. Surv. S. Afr. 56: 88 (1987); Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Eastern Cape, Port Elizabeth (3325): "inter frutices (altit. III) laterum montium Van Stadensrivierberge (Uitenhage)" [Among bushes (altit. 3) on the sides of the Van Stadensrivier mountains (Uitenhage)] –CD, *Ecklon & Zeyher 657* (S!, S08-12037, lecto., here designated; E!, K!, L!, M!, MO!, isolecto.). [Notes: i) The sheet in S is the only one which we have seen which bears an original label in their (Ecklon & Zeyher) hand writing so we choose it as the lectotype. ii) Burtt Davy's reference (Fl. Pl. Transv. 375, 1932) to this species is incorrect. His cited specimens refer to *P. latifolia* (Harv.) C.H. Stirt. iii) The specimen illustrated in Bot. Mag. 136: t.8331 (1910) is *P. arborea* Sims].

Psoralea krebsii Vogel nom. nud.

Psoralea pinnata L. β. glabriuscula Eckl. & Zeyh., Enum.: 224 (1836); *Psoralea pinnata* L. var. γ *subglabra* Harv. in Harv. & Sond., Fl. Cap. 2: 144 (1862). Type: "prope Swellendam" [near Swellendam], Ecklon & Zeyher 1515 (Not yet found).

Psoralea arborea Sims sensu Eckl. & Zeyh., Enum. 224 (1836), non Sims (1819). Correctly noted by Walp. in Linnaea 13: 512 (1839).

Small to large slender shrub or treelet up to 3 m tall, reseeder. *Stems* erect, single, branching from lower portion, grey or brown, branches spreading or arching, rigid, with scattered white storied lenticels. *Seasonal shoots* green, glabrous or glabrescent. *Leaves* (5)7–9(11)-foliolate, imparipinnate, patent, distributed along the branches, in older plants clustered at tips, petiolate; leaflets 20–35×0.2–0.4 mm, linear lanceolate, terminal leaflet shortest, basal pair longest, lateral about the same length of the terminal, three uppermost leaflet digitately inserted, sparsely hairy; petioles 4–5 mm long; rachis 15–20 mm long; stipules 2–3×1–2 mm, small, deltoid, spreading to recurved, twisted, callose, becoming woody. *Inflorescences* axillary, borne in upper axils of seasonal shoots but without leafy extensions, pseudo-spicate, lax, two, three or more flowers per axil, pedunculate, pedicellate; peduncles stout and rigid, 17–30 mm long; cupulum bilobed or trilobed, slightly overlapping base of the calyx or free of it; pedicels 1–3 mm long. *Flowers* 12–15 mm long, dark blue to dark purple, fading with age, maturing more or less simultaneously.

Calyx 7–8 mm long, tube 4.8–5.1×4 mm, nigro-hirsute; lobes equally developed, 2−3 mm long, shorter than the calyx tube; lateral and vexillar lobe lanceolate; carinal lobe acute, same width as other four lobes. *Standard petal* 10–12×8.5–10 mm, broadly ovate, dark blue to dark purple with large M-shaped white nectar guide with a small dark purple flash in crotch, veins darker than blade; claw 3–4 mm long. *Wing petals* 8–9×3–3.5 mm, blue to pale mauve, auricles well developed, sculpturing present, basal, comprised of 2−3 rows of 10−12 transcostal lamellae; claw 3–4 mm long; fused to and longer than keel. *Keel* 7–8×2.5–3.5 mm, white, suffused on inner faces of the apex with violet-purple; claw 3–4 mm long. *Androecium* 11 mm long, tenth stamen slightly attached, broadly fenestrate. *Pistil* 10 mm long, ovary 3 mm long, style filiform, 6 mm long to the point of flexure where it curves, swollen at point of flexure, height of curvature 3 mm long, glabrous but with many small stalked club-shaped glands on upper part, stigma penicillate. *Fruits* oblong, 4–5×2–3.0 mm. *Seeds* one, 3–4×2–2.8 mm, dark brown (Figure 27).

4.6.2.17.1. Diagnostic characters

Psoralea affinis is similar to *P. montana* in growth habit but differs in being an erect lowland shrub or low montane tree to 4 m tall, usually with greyish stems and storied lenticels (versus a tall montane shrub or tree to 5 m, with brown stems and (if present) stratified lenticels), with green, nigro-hirsute seasonal shoots (versus greyish green, sparsely pubescent seasonal shoots), leaflets narrower, 0.2−0.4 mm wide (versus 1.5−2.5 mm wide), terminal leaflet shortest (versus terminal leaflet second longest), petioles 4–5 mm long (versus 5–10 mm long), inflorescences 2−many flowers per axil but aggregated in terminal pseudo-spicate clusters in upper axils of seasonal shoots (versus1-flowered per axil but aggregated in terminal pseudo-capitate clusters in upper axils of seasonal shoots), and calyces nigro-hirsute (versus finely silvery pubescent). It is similar to *P. arborea* when in flower as both have deep blue to purple flowers and black-haired calyces but in *P. affinis* the plants start branching in the lower portion (versus in the upper portions); leaflets are smaller, 20–35×0.2–0.4 mm, glabrous when mature (versus 30–50×1–2 mm, glabrescent above, wispily hairy below), and inflorescences about as long as or just exerted from leaves (versus well exerted from leaves).

4.6.2.17.2. Distribution and habitat

Psoralea affinis has a wide distribution ranging from Caledon (Western Cape Province) eastwards to Port Elizabeth (Eastern Cape Province) (Figure 28). It occurs in lowland and montane fynbos in the rich soils of seepages, streamsides, open areas and forest margins in lower-lying areas but also in poorer stony soils on mountains at elevations between 100–1800

m. It has been found in Garden Route Granite Fynbos, Garden Route Shale Fynbos, South Outeniqua Sandstone Fynbos, Kouga Sandstone Fynbos, Kouga Grassy Sandstone Fynbos, Uniondale Shale Renosterveld, Langkloof Shale Renosterveld, and Tsitsikamma Sandstone Fynbos vegetation types (FFg 5, FFh 9, FFs 19, FFs 27, FFs 28, FRs 16, and FRs 17) (Rebelo *et al.* 2006).

4.6.2.17.3. Etymology

The specific epithet *affinis* is derived from the Latin words *ad* + *affinis* (neighbouring, connected with, similar by marriage) and refers to its affinity with *P. pinnata*.

4.6.2.17.4. Conservation status

Psoralea affinis is recorded by Foden & Potter (2009) as being of least concern (LC). We agree with this assessment.

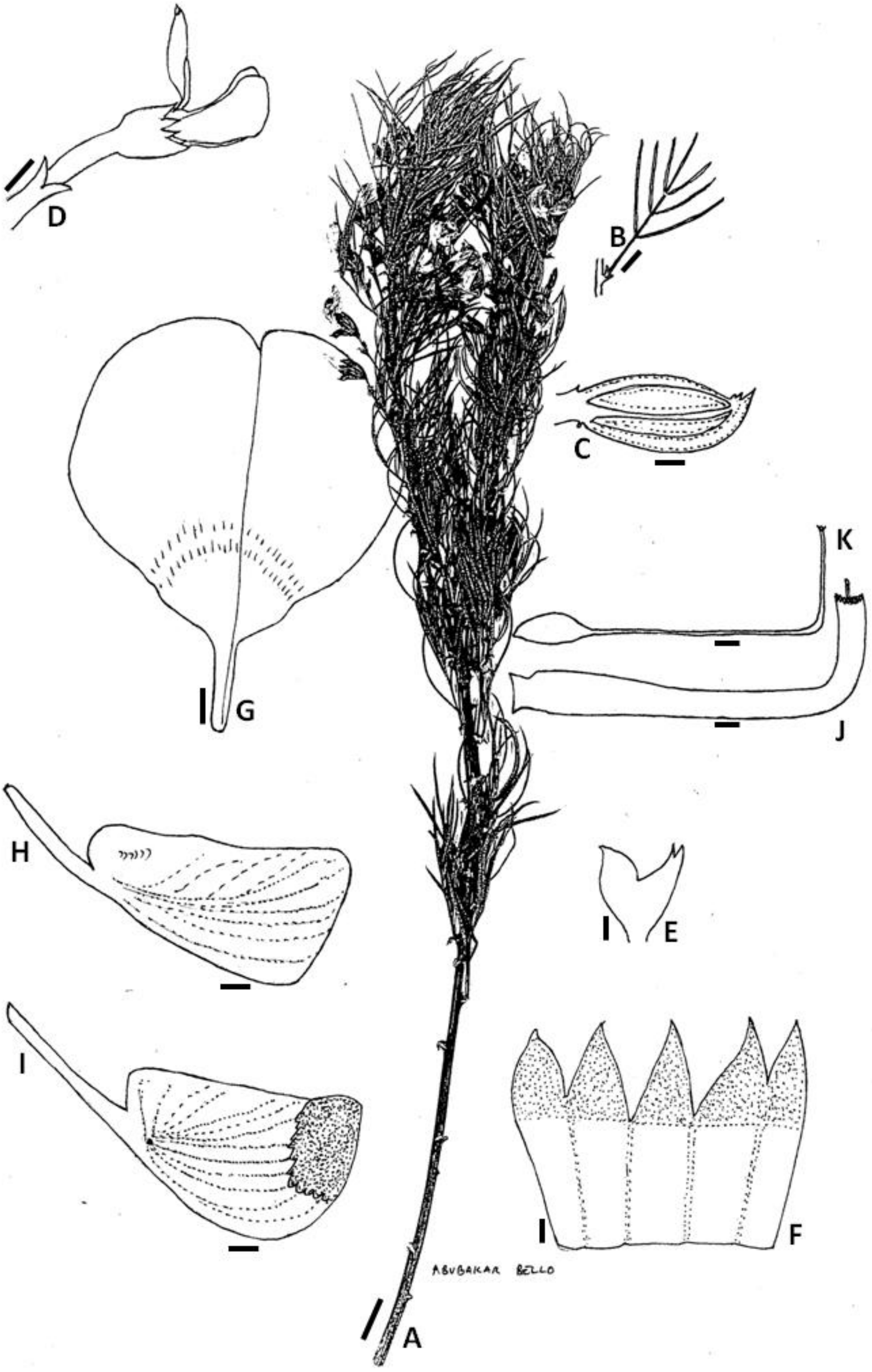

Figure 27. Vegetative and reproductive morphology of *P. affinis* (A) flowering shoot; (B) 7-foliolate leaf (C) bud; (D) flower; (E) cupulum; (F) calyx opened out with vexillary lobes to the right; (G) front view of standard petal; (H) wing petal; (I) keel; (J) androecium; (K) pistil. Voucher *A. Bello 77* (BOL). Scale bars: A, 1 cm; B–K, 1 mm. Artist: Abubakar Bello.

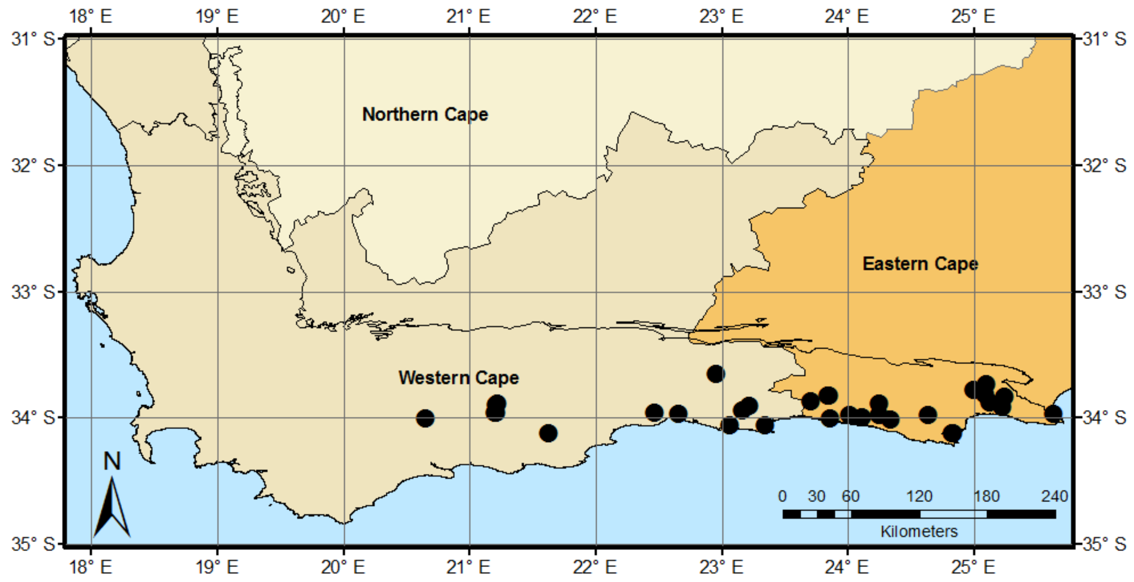

Figure 28. Known distribution of *P. affinis*.

4.6.2.17.5. Specimens studied

South Africa. WESTERN CAPE: **3320 (Montagu):** Road to Heidelberg, after Stonehaven (-CD), 5 Oct 1981, *Stirton C.H. 9719* (K, PRE); Bronze Grove (-DC), 5 Oct 1981, *Stirton C.H. 9718* (K, PRE); Tradouw Pass (-DC), 20 Oct 1980, *Stirton C.H. 11258* (K). **3321 (Ladismith):** Tollgate, halfway up Garcia's Pass (-CC), 10 Dec 1981, *Stirton C.H. & Žantovská J.Z.10285* (K); 5 Oct 1981, *Stirton C.H. 9708* (K, PRE); 20 km from Riversdale to Ladismith (-CC), 10 Dec 1981, *Stirton C.H. & Žantovská J.Z.10275* (K); 10 Nov 1986, *11543* (K, NBG). **3420 (Bredasdorp):** Buffeljagsrivier (-BA), 24 Sep 1976, *Stirton C.H. 6167* (K, PRE). **3421 (Riversdale)**: Wingerd se Berg, east of Albertinia (-BA), 20 Oct 1987, *McDonald D.J. 1372* (NBG). **3422 (Mossel Bay):** Goukamma (-BB), 4 Oct 2004, *Stirton C.H. 9676* (K, PRE); between Knysna River drift and Goukamma, *Burchell 5557* (K); Port Elizabeth West E. 50 (K). **3423 (Knysna):** Between Knysna and The Heads (-AA), 10 Nov 1913, *Peter A. 50460* (B, K); 2 km along road to Noetzie (-AA), 3 Oct 1981, *Stirton C.H. 10630* (K, PRE); Keurbooms valley (-AB), 16 Aug 1933, *Humbert H. 10076* (K, P); Plettenberg Bay (-AB), Sep 1921, *Rogers F.A. 15475* (K); Nov 1923, *Rogers F.A. 28563* (K); Stormsriver Bridge (-BB), 31 Aug 1947, *Story R. 2852* (K); 5 km from Stormsrivier mouth to Knysna road (-BB), 3 Oct 1981, *Stirton C.H. 9623* (K, PRE). EASTERN CAPE: **3323 (Willowmore)**: Formosa Peak, Tsitsikamma Mountains (-DC), 20 Jan 1941, *Esterhuysen E. 4649* (BOL); Greenside Inn, Tsitsikamma (-DC), 20 Dec 2009, *Muasya A.M. & Stirton C.H. 5288.1* (BOL); Green Side lodge, turn of Bluelilies river (-DC), 16 Feb 2014, *Bello A. & Stirton C.H. 57* (BOL). **3324 (Steytlerville)**: Blouberg Fire Lookout Tower (-CC), 13 Nov 1986, *Stirton C.H. & Žantovská J.Z.* 11614 (K); Bottom of Suuranys Pass (-CC), 1 Oct 1984, *Stirton C.H. & Žantovská J.Z.10890, 10891, 10927* (K); Kareedouw Pass (-CC), 1 Oct 1984, *Stirton C.H. 10931* (K, PRE); 12 mi. E of Witte Els Bosch (-CC), 12 Nov 1928, *Hutchinson J. 1418* (K); Between Billson and Two Streams (-CC), 15 Nov 1913, *Peter A. 50770* (B, K); Elandsrivierberg area, Otterford Forest Reserve (-DB), Sep 1973, *Thompson M.F. 1835* (NBG); Otterford Forest Reserve (-DB), 31 Aug 1947, *Rodin R.J. 1130* (BOL, PRE); Swartrivier Bridge on Jeffrey's Bay to Humansdorp road (-DD), 2 Oct 1981, *Stirton C.H. 9605* (K, PRE); Farm Rondebosch, Humansdorp (-DD), Sep 1918, *Schönland S. 3018* (K). **3325 (Port Elizabeth)**: Sand River Reservoir (-CA), 4 Oct 1931, *Holland T.H. 3368* (BOL); Between Weltevrede and Rocklands (-CA), 30 Sep 1984, *Stirton C.H. 10865* (K); Groot Plaat (-CA), 5 Sep 1974, *Scharf A.T. 1573* (K, PRE); Van Stadens (-CC), Nov 1910, *Patterson F.M. 734* (K); Mountains above Van Stadens River, Oct, *Ecklon C.F. 352* (K, TCD); 1 Dec 1930, *Long F.R. 132* (K). **3424 (Humansdorp):** Palmietvlei (-AB), 2 Oct 1981, *Stirton C.H. 9618* (K, PRE); Clarkson (-AB), Sep 1926, *Thode J. A. 803* (K). **Without precise locality:** Die Tol, 1822, *Thom G. s.n.* (K).

4.6.2.18. *Psoralea brilliantissima* C.H. Stirt., Muasya & A. Bello *sp. nov.* *Psoralea* sp. 22, Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Western Cape, Riversdale (3421): 7 km from Gouritz River mouth to Stilbaai (-CD) 6 Jan 2008, *Muasya A.M. & Stirton C.H. 3621*(BOL!, holo.: NBG!, PRE!, iso.).

Small to large robust shrub up to 3 m tall, reseeder. *Stems* erect, single, multi-branched, branching in upper parts, forming an umbrella shape, rigid, trunks short, grey, branches brown. *Seasonal shoots* pale green, glabrous or glabrescent, appressed, clustered, angular, very strong sweet odour. *Leaves* (5)7−9-foliolate, imparipinnate, distributed along the seasonal shoots, erect or patent, soft, petiolate; leaflets 30–35×0.3–0.6 mm, linear lanceolate, terminal leaflet shortest or equal to the basal pair which are longest, rounded abaxially, flattened and grooved adaxially, three uppermost digitately to occasionally pinnately inserted; petioles 3–4 mm long, grooved, shorter than terminal leaflet; rachis 10–15 mm long; stipules 1–1.7 mm long, rapidly senescent, persistent, shorter than petiole, subulate, glabrous, margins unrolled away from the axis, stiff, callose, leaving a raised scar when senescent. *Inflorescences* axillary, borne in upper axils of seasonal shoots without leafy extension, pseudo-capitate, congested, 3–6 flowers per axil, pedunculate, pedicellate; peduncles stout and rigid with black hairs, 25–35 mm long; cupulum overlapping the calyx, bilobed, with a 3 mm cleft in vexillary pair, sparsely hairy, lobes equally developed, broadly ovate, apex acuminate, greenish when young, senescent but persistent at anthesis; pedicels 2.5–2.7 mm long, with black hairs. *Flowers* 13–14 mm long, intense hyacinth blue, each flower shoot set subtended on the stem side by a pair of small free caducous bracts. *Calyx* 7−8 mm long, tube 4–5×4 mm, covered with a mixture of finely black and white hairs, glabrous inside; lobes equally developed, shorter than the tube, acuminate, carinal widest, 1.5–2 mm long, triangular, vexillar fused for half their length, densely black hairy inside, more sparsely outside, glandular. *Standard petal* 10–12×8.5–9.5 mm, broadly ovate, intense hyacinth blue, margin pale mauve, with a large white M-shaped nectar guide, visible from behind, with a small deep purple crotch; appendages well developed, free, veins paler; claw 2–4 mm long, elongated and narrow. *Wing petals* 10×3–3.5 mm, distal half mauve, the rest white, sculpturing present, upper base comprised of 8−10 transcostal lamellae, pocket well developed, auricle swollen; claw 3–4 mm long. *Keel* 9–10×2.5–3.5 mm; claw 3–4 mm long. *Androecium* 8−9 mm long, tenth stamen free, fenestrate. *Pistil* 8 mm long, ovary 1−1.5 mm long, stalked at upper end covered in club-shaped glands, style 6 mm long straight and then curved upwards, height of curvature of style 2 mm, thickest at point of flexure, stigma penicillate. *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* solitary, 2–4×2 mm, dark brown to black (Figure 29)**.**

4.6.2.18.1. Diagnostic characters

Psoralea brilliantissima is similar to *P. arborea* in growth form with which it was previously included. It differs in its shrubby umbrella habit usually up to 3 m tall (versus erect shrub or tree, 5–10 m tall), with pale green sparsely pubescent seasonal shoots (versus green villoso-pubescent), narrower leaflets, 0.3–0.6 mm (versus wider leaflets, 1–2 mm), petioles 3–4 mm long (versus longer petioles, 5–8 mm long), glabrous stipules (versus hairy), inflorescences borne in upper axils of seasonal shoots, pseudo-capitate, tightly congested, 3–6 flowers per axil, just longer than leaves (versus 1–3 flowers per axil in upper axils, well exerted from leaves), peduncles shorter, 25–35 mm long, glabrous (versus 45–60 mm long, hairy), flowers intense hyacinth blue with paler mauve margins (versus deep blue to purple), standard broadly ovate (versus standard very broadly ovate), calyces sparsely black- to white-haired (versus densely black-haired).

Figure 29. Vegetative and reproductive morphology of *P. brilliantissima* (A) flowering shoot; (B) 7-foliolate leaf; (C) bud; (D) peduncle; (E) cupulum; (F) calyx opened out with vexillary lobes to the right; (G) front view of standard petal; (H) wing petal; (I) keel; (J) androecium; (K) pistil. Voucher *A. Bello 142* (BOL). Scale bars: A, 1 cm; B–K, 1 mm. Artist: Abubakar Bello.

4.6.2.18.2. Distribution and habitat

Psoralea brilliantissima is a rarely collected species endemic to the Western Cape Province extending from Still Bay in the west, northwards to Albertinia and then as far east as Gouritz

(Figure 30). It occurs predominantly on limestone substrate between 95–220 m above sea level. *Psoralea brilliantissima* has been found in Garden Route Granite Fynbos, De Hoop Limestone Fynbos, South Outeniqua Sandstone Fynbos and Mossel Bay Shale Renosterveld vegetation types (FFg 5, FFl 2, FFs 19 and FRs 14) (Rebelo *et al.* 2006).

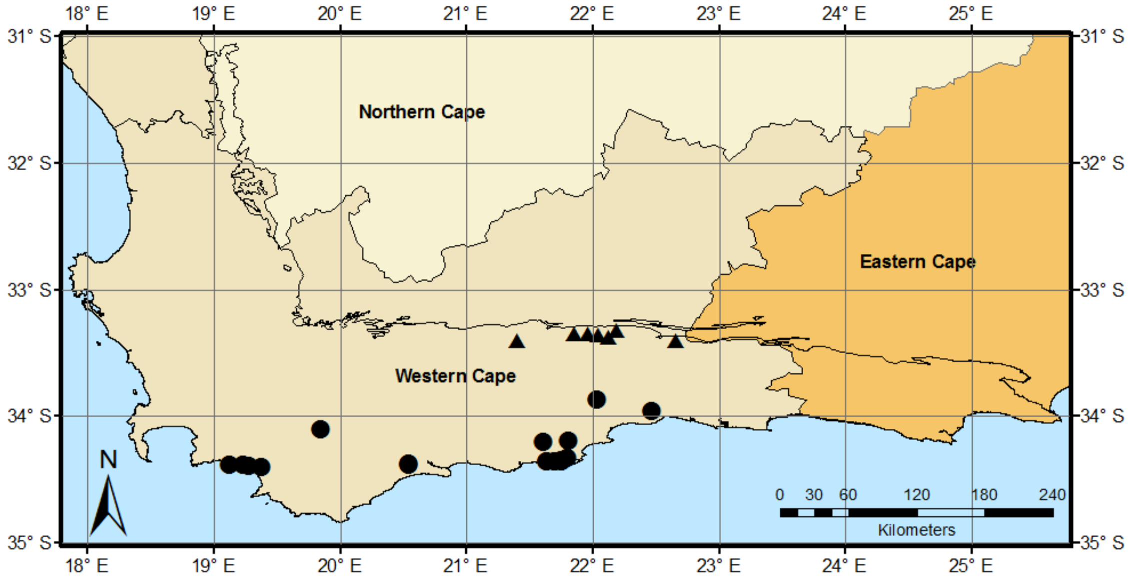

Figure 30. Known distribution of *P. brilliantissima* (circles) and *P. elegans* (triangles).

4.6.2.18.3. Etymology

The specific Latin epithet *brilliantissima* was chosen to represent the stunning display of its intense beautiful coloured flowers. The word is derived form the French word *brilliant* (shining) and is used in the sense of "having a high saturation and reflecting a considerable amount of light; vivid" (http://dictionary.reference.com/browse/brilliant).

4.6.2.18.4. Conservation status

This species is assessed as least concern (LC) due to its abundance in its distribution area and currently not under and/or close to any threat (von Staden *et al.* 2009). However, this needs to be monitored carefully as there is a spreading encroachment towards its habitat by invasive Australian Acacias which have accelerated their spread from frequent fires. Another encroaching threat is the eradication of bushland by chain dragging so as to stimulate the growth of thatching material (Restionaceae). The estern populations are at most risk.

4.6.2.18.5. Specimens studied

South Africa. WESTERN CAPE: **3322 (Oudtshoorn)**: Robinsonspas (-CC), 23 Sep 1994, *Meyer J.J. 340* (PRE, BOL); 12 Oct 1972, *Bremer K. 318* (PRE); George (-CD), 1 Aug 1912, *Rogers F.A. 4291* (BOL). **3419 (Caledon)**: Baviaansfontein, Grootbos (-CB), 1 Aug 1940, *Stokoe T.P. 7575* (BOL); Just before Strandskloof (-CB), 6 Oct 1981, *Stirton C.H. 9745* (PRE); Near Grootbos to Baviaansfontein on river (-CB), 6 Oct 1981, *Stirton C.H. 9746* (PRE); Between Franskraal and Strandkloof (-CB); Tierfontein Farm, Koudberge (-CD), 18 Nov 2007, *Muasya A.M. & Stirton C.H. 3248* (BOL), 24 Aug 1946, *Leighton F.M. 1918* (BOL); Before Goedvertrouw, after Bruinklip (-DA), 6 Oct 1981, *Stirton C.H. 9742* (PRE). **3420 (Bredasdorp)**: Kathoek (-AB), 10 Oct 1940 *Pillans N.S. 9386* (BOL). **3421 (Riversdale)**: Klein Jongensfontein farm, Riversdale (-AD), *Bohnen P. 7693* (K, PRE); Oudetuin, Albertinia (-BA), *Muir J. 1851* (BOL); Between Blikhuis and Driefontein farms on Stilbaai to Gouritz Road (-BC), 30 Nov 2009, *Muasya A.M. & Stirton C.H. 5033* (BOL); 12 km from Gouritz River mouth to Stilbaai (-BC), 6 Jan 2008, *Muasya A.M. & Stirton C.H. 3624* (BOL); 17 km from Gouritz River mouth to Stilbaai (-BC), 6 Jan 2008, *Muasya A.M. & Stirton C.H. 3626* (BOL); 7 km from Gouritz River mouth to Stilbaai (-BC), 6 Jan 2008, *Muasya A.M. & Stirton C.H. 3621* (BOL); Potberg (-BC), 21 Nov 1980, *Stirton C.H. 8251* (PRE); Western end of Potberg near Eagles Lair (-BC), 1 Nov 2008, *Dludlu M. & Stirton C.H. 74* (BOL); Potberg mountain, edge of escarpment (-BC), 1 Nov 2008, *Dludlu M. & Stirton C.H. 35* (BOL).

4.6.2.19. *Psoralea elegans* C.H. Stirt. *sp. nov.* Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); *Psoralea* sp. 23, Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Western Cape, Ladismith (3321): 2.5 km West of turnoff to Gamkaskloof from Prince Albert / Swartberg Pass road (-BD), 11 Nov 1986, *Stirton C.H. & Žantovská J. 11568* (K!, holo.; PRE!, iso.).

Small to large robust shrub up to 3 m tall, reseeder, colonial. *Stems* single, ascending to erect, blackish brown; branches stiffly erect. *Seasonal shoots* pale green to brownish purple, with rough scales, glabrous. *Leaves* 5−7-foliolate, imparipinnate, patent, petiolate, distributed along the branches; leaflets 30–35×0.3–0.4 mm, linear lanceolate, glabrous, terminal leaflet shortest, basal pair longest, three uppermost digitately inserted; petioles 4–4.5 mm long; rachis 10−15 mm long; stipules 1–2×0.5–1 mm, persistent. *Inflorescences axillary*, borne in uppermost axils of seasonal shoots, pseudo-spicate, lax, (2)3–6 flowers per axil, pedunculate, pedicellate; peduncles stout and rigid, 30–45 mm long; cupulum free from calyx, bilobed with one of the vexillar lobes variously bilabiate; pedicel 1–2 mm long. *Flowers* 12−14 mm long, mauve to purple, without scent. *Calyx* 5–6 mm long, tube 4–4.5×3.5 mm, glabrous; lobes sub-equally developed, shorter than the tube, lateral and vexillar lobes acute, carinal lobe cucullate (boat-shaped). *Standard petal* 10–12×8–9 mm, narrowly ovate, mauve; claw 3–4 mm long, nectar guide white with a deep purple flash. *Wing petals* 10–11×3.2–3.5 mm, strongly up-curving, sculpturing present, white; claw 3−4 mm long; *Keel* 10–10.5×2.5–3 mm; claw 3–4 mm long. *Androecium* 11 mm long, broadly fenestrate, upper part recurved. *Pistil* 10 mm long, ovary 3 mm long, glabrous but with many small stalked club-shaped glands on upper part; style filiform, 6 mm long straight to point of flexure, then upcurved, swollen at point of flexure, height of curvature 4 mm, stigma pedicellate, penicillate. *Fruits* narrowly oblong, 4–5×2.5–3.0 mm. *Seeds* solitary, 2–4×2 mm, dark brown to black (Figure 31).

4.6.2.19.1. Diagnostic characters

Psoralea elegans is a montane species similar to the lowland species *P. imminens,* both occurring in the same general region, and having the same number of leaflets, long peduncles (30–60 mm long), and small calyces (5–6 mm long, tube 4–4.5×3.5 mm). *Psoralea elegans* differs in its blackish brown stems (versus yellowish-green brown), with numerous greyish white storied lenticels, when young, becoming grey and deeply fissured with age), stiffly erect branches (versus virgate or pendent), rigid glabrous leaflets (versus soft, hairy leaflets), (2)3–6 unscented mauve to purple flowers per axil (versus 1−3 greenish white strongly scented flowers per axil), peduncles stout and rigid (versus filiform and flexuous).

4.6.2.19.2. Distribution and habitat

Psoralea elegans is a montane species distributed from Groot Swartberg Mountains in the Western Cape Province to the Kouga region of the Eastern Cape Province (Figure 30). It occurs in gullies, streams and seepages of mountain and subalpine fynbos between 1300–1700 m. It is associated with North Swartberg Sandstone Fynbos, South Swartberg Sandstone Fynbos and Kango Limestone Renosterveld (FFs 23, FFs 24 and FRl 1) (Rebelo *et al.* 2006).

4.6.2.19.3. Etymology

The specific epithet *elegans* is derived from the Latin adjective *elegans*, *-antis* (fine, tasteful or elegant) and alludes to the attractiveness of this plant when in flower.

4.6.2.19.4. Conservation status

This species has been evaluated as Rare (Stirton & Raimondo 2008) due to its range restriction on the Swartberg Mountains. Its EOO is not likely to be larger than 500 km^2. This status is affirmed in this study since the condition has not changed.

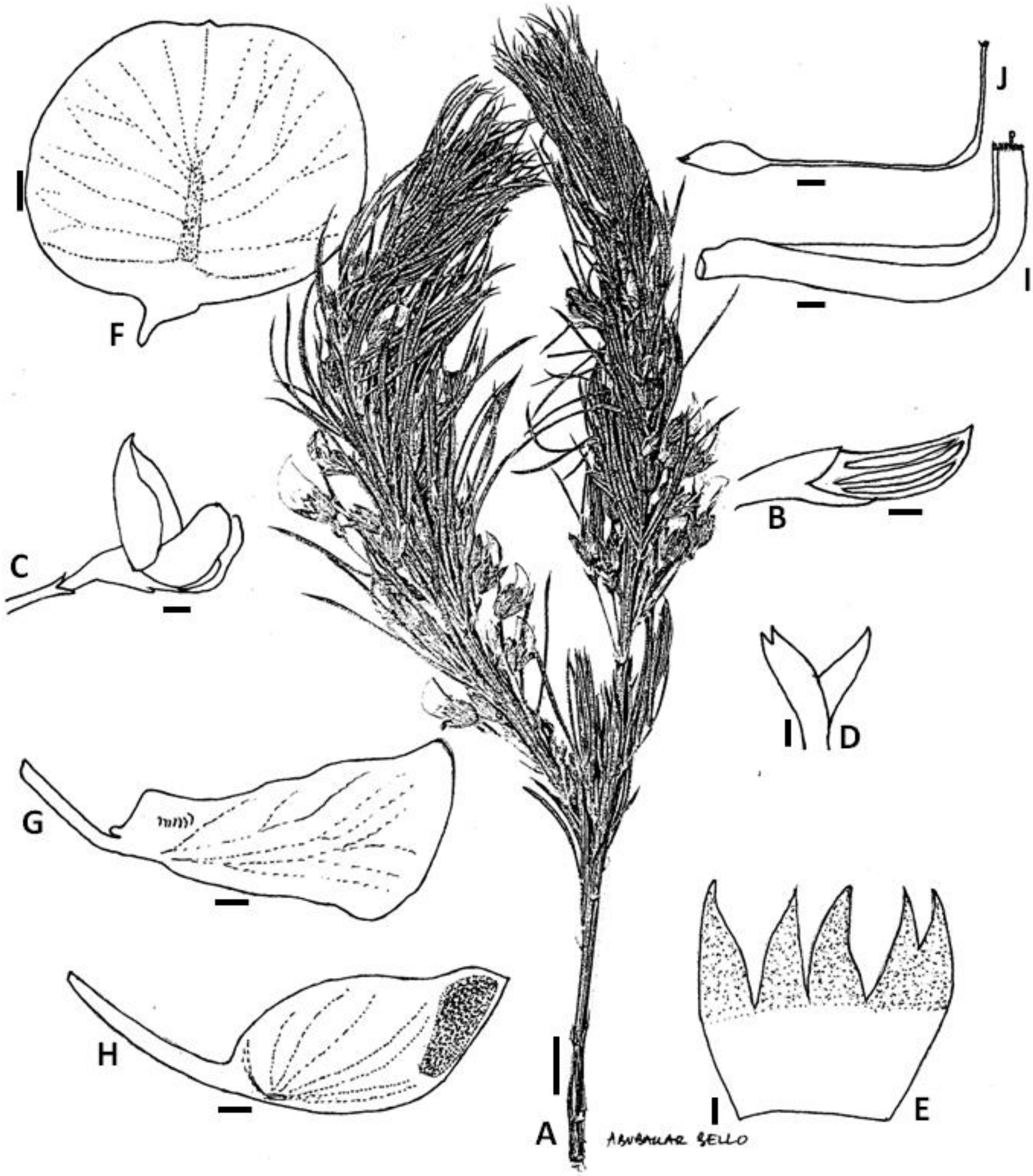

Figure 31. Vegetative and reproductive morphology of *P. elegans* (A) flowering shoot; (B) bud; (C) flower; (D) cupulum; (E) calyx opened out with vexillary lobes to the right; (F) front view of standard petal; (G) wing petal; (H) keel; (I) androecium; (J) pistil. Voucher *A. Bello 96* (BOL). Scale bars: A, 1 cm; B–J, 1 mm. Artist: Abubakar Bello.

4.6.2.19.5. Specimens studied

South Africa. WESTERN CAPE: **3221 (Ladismith)**: Poort beyond campsite, Swartberg Mts (-AD), 17 Oct 1966, *Taylor H.C. 6964* (K, PRE); 20 km from turn off to Die Hel, top of Swartberg Pass (-BD), 11 Nov 1983, *Jackson W.P.U. 9* (NBG). **3322 (Oudtshoorn)**: Bassonsrus, Upper Cango valley (-AC), 4 Nov 1974, *Moffett R.O. 427* (PRE, NBG); Swartberg mountains, mid northern slopes of Blesberg, at upper end Tierkloof (-BC), 15 Dec

1986, *Vlok J.H.J. 1781* (PRE); Swartberg Mountains (-AC), Nov 1945, *Stokoe s.n.* (SAM 55814). **3323 (Willowmore)**: Georgida, Uniondale District (-AD), 4 Nov 1941, *Esterhuysen E. 6385* (BOL); Avontuur Poort (-CA), Dec 1928, *Fourcade H.G. 4277* (BOL, K, NBG, PRE); Avontuur Mountain, Uniondale (-CA), 2 May 1941, *Compton R.H. 10775,* 2 Dec 1951, *23052,* 23 Nov 1949, *21789* (NBG); Kougaberge, river banks (-DB), 20 Oct 1986, *Oelofsen O.N. 77* (PRE); Klein Rivier, Joubertina, Kouga (-DB), 27 Nov 1941, *Esterhuysen E. 7030* (PRE); Robinson's Pass, Mossel Bay, Oudstroom (-DB), 12 Jan 1950, *Martin B. 170* (NBG).

4.6.2.20. *Psoralea floccosa* C.H. Stirt., Muasya & A. Bello *sp. nov.* Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); *Psoralea* sp. 24, Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Western Cape, Oudtshoorn (3322)**:** Outeniqua Mountains, upper southern slopes of Ruitersberg (-CC), 30 Nov 1986, *Vlok J.H.J. 1734* (NU!, NU0015574-0, holo.).

Weak sparsely branched shrub up to 2.5 m tall, reseeder. *Stems* erect, single, branching in upper portion, lax, arching to erect, greyish black. *Seasonal shoots* glaucous, rough, greenish white, densely covered in dense woolly patent white hairs. *Leaves* 9–11(13)-foliolate, imparipinnate, petiolate, pilose, two lowest pairs of leaflets emerge from the same node or very close, often bent downwards, separated from the rest by a wide gap on the rachis, the rest of the leaflets curved towards the apex; leaflets 13–17×0.3–0.4 mm, linear lanceolate, rounded, grooved, terminal leaflet shortest, basal pair longest (attached or separate at the node), three uppermost digitately inserted; petioles 2–3 mm long, shorter than terminal leaflets; rachis 7−10 mm long, hairy; stipules 2–3×0.5–1.5 mm, persistent, densely hairy, subulate, margins unrolled away from the axis, shorter or equal in length to the petiole, ovate. *Inflorescences* axillary, borne in upper axils of the old seasonal shoots (or along the length of young seasonal shoots) with terminal leafy extension, pseudo-spicate (on young seasonal shoots) or pseudo-capitate (on older shoots), lax, 1-3 flowers per axil; pedunculate, pedicel absent; peduncles 1−2 mm long, stout and rigid, densely pubescent; cupulum trilobed, overlapping the calyx, 3 mm long, densely pubescent with a mixture of black and white hairs, lobes unequally developed, carinal lobe longer, 2 mm long, vexillary lobe with a cleft, 1.5−2 mm long, all acuminate, upper surface of the lobes with white hairs. *Flowers* 10−11 mm long, white to mauve. *Calyx* 6−7 mm long, tube 2.5–3×4 mm; densely hairy, mainly white-haired with or without occasional black hairs; lobes sub-equal, shorter or equal to the tube, hairy, carinal lobe longer, 3−4 mm long, boat-shaped, lateral and vexillar lobe acuminate with dense black hairs on the margin. *Standard petal* 10–12×13–15 mm, very broadly ovate, white with a dark central purple flash in the crotch and purple veins; claw 3–4 mm long. *Wing petals* 9–11×3–4 mm, white to mauve; cultrate and upcurving; claw 2–2.5 mm long, much longer than keel, auriculate, sculpturing present, upper basal, comprised of 8–12 transcostal lamellae. *Keel* 9–10×3–4 mm, white, apically suffused with dark violet-purple on inner face of apex; claw 3–4 mm long; apex acuminate. *Androecium* 10 mm long, sheath split adaxially, tenth stamen lightly adherent to the right hand base of the sheath margin, fenestrate. *Pistil* 10 mm long, ovary 2 mm long, stipitate; style 6 mm long, straight to the point of flexure, swollen at point of curvature, height of curvature 3 mm long, stigma capitate, penicillate. *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* one, 2–4×2 mm, dark brown to black (Figure 32).

4.6.2.20.1. Diagnostic characters

Psoralea floccosa is similar to *P. pinnata* in growth habit with which it was previously included. It differs in being a lax weak sparsely branched shrub to 2.5 m tall (versus virgate multi-branched shrub to tree up to 5 m), covered in densely woolly patent white hairs at ends of seasonal shoots (versus pubescent), 9–11(13)-foliolate pilose leaves (versus (5)7–9-foliolate villoso-pubescent leaves), narrower 0.3–0.4 mm wide linear-lanceolate leaflets with raised glands (versus 0.5–1 mm wide linear leaflets with sunken glands), flowers 10–11 mm long (versus 13–14 mm long), standard very broadly ovate, white with a dark central purple flash and purple veins (versus standard obovate, white suffused with pale mauve in the central area and with a single purple 3–4 mm long flash situated between and above the free swollen callosities at the top rim of the claw, veins hyaline), and calyx concolorous, yellowish green, lobes shorter than tube (versus dicolorous, lobes dark green and tube yellowish green).

4.6.2.20.2. Distribution and habitat

Psoralea floccosa is a little known species endemic to the Langeberg, Outeniqua to Tsitsikamma Mountains (Figure 33). It occurs on dry sandy loams of northern slopes and also on gulleys and rocky outcrops in moist humid soils of southern slopes in mountain fynbos and eastern fynbos-renosterveld. *Psoralea floccosa* is known from the South Langeberg Sandstone Fynbos and Tsitsikamma Sandstone Fynbos vegetation types (FFs 16 and FFs 20) according to Rebelo *et al.* (2006).

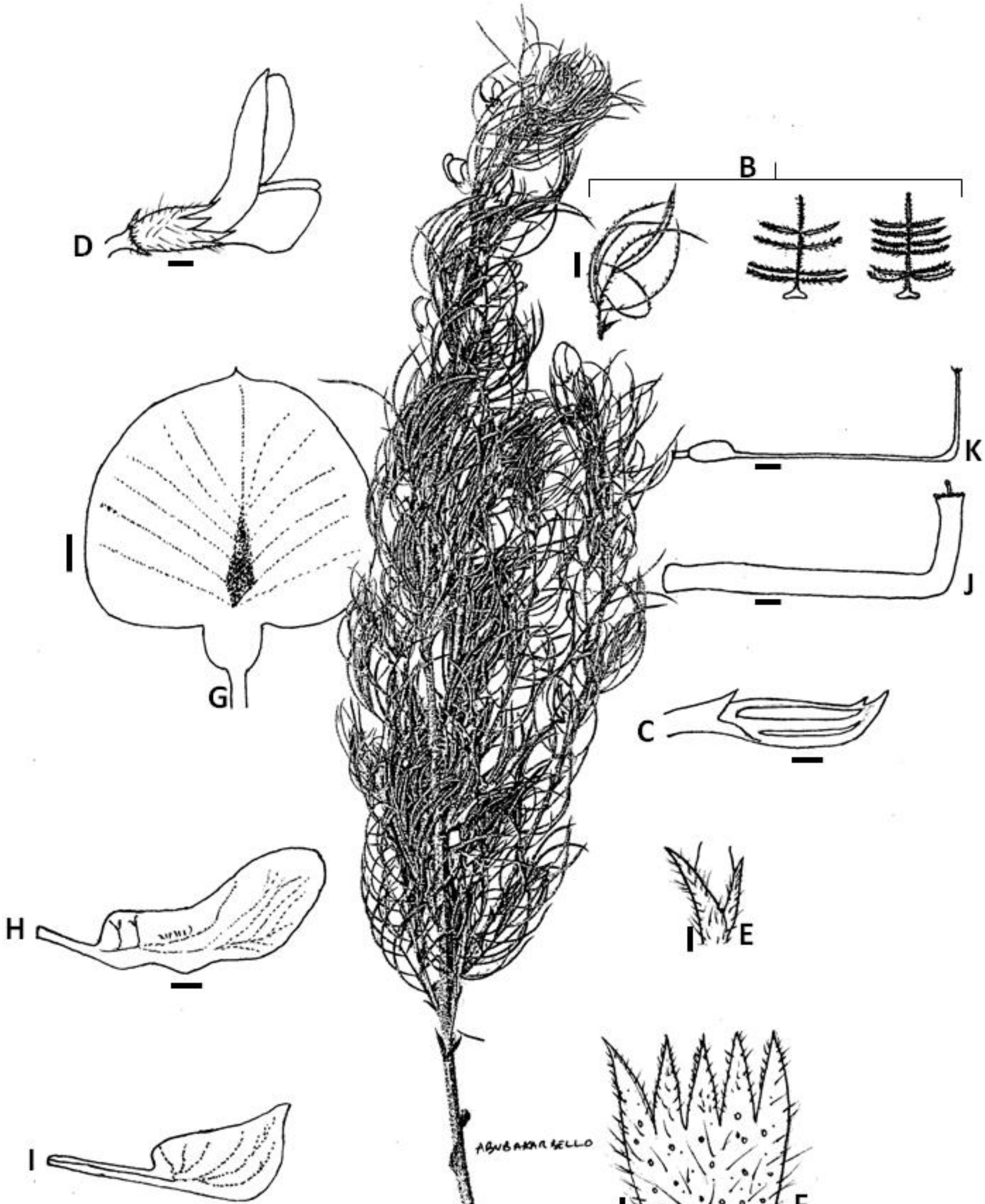

Figure 32. Vegetative and reproductive morphology of *P. floccosa* (A) flowering shoot; (B) 7, 9 and 13-foliolate leaf; (C) bud; (D) flower; (E) cupulum; (F) calyx opened out with vexillary lobes to the right; (G) front view of standard petal; (H) wing petal; (I) keel; (J) androecium; (K) pistil. Voucher *N. van Berkel 1117* (BOL). Scale bars: A, 1 cm; B–K, 1 mm. Artist: Abubakar Bello.

4.6.2.20.3. Etymology

The specific epithet *floccosa* is derived from the Latin word *floccus* –i, m. (flock of wool) and alludes to the long pilose hairs characteristic of this species.

4.6.2.20.4. Conservation status

Psoralea floccosa has been evaluated against all the five IUCN criteria of threat category (IUCN, 2012a, 2012b) and was found not to meet any of the categories of threat. It is therefore assigned a status of least concern (LC). The species is widespread and occurs mostly in protected mountain catchments.

4.6.2.20.5. Specimens studied

South Africa. WESTERN CAPE: **3321(Ladismith)**: West of Sleeping Beauty, on Corenteberg peak, near beacon 48, Mountain summit, Garcia Forest Reserve, Langeberg (-CC), 22 Nov 1991, *McDonald D.J. 2117* (PRE); Near beacon 48, Mount summit/peak, W of sleeping beauty on Corenteberg Peak, Garcia's Forest Reserve, Langeberg (-CC), 10 Dec 1982, *Taylor H.C. 10526* (NBG); Herbertsdale, top of Vreysberg peak, west of Cloete's Pass (-DC), 15 Nov 1987, *Vlok J.H.J. 1879* (NU, PRE); Attaquasberg, small peak E of Vreysberg (-DC), *McDonald D. 1765* (NU, NBG); Extreme summit of Langeberg, above Witte Els (-DC), Nov 1913, *Muir J. 1250* (BOL). **3322 (Oudtshoorn)**: Lower northern slopes of Engelseberg (-CC), 28 Sep 1984, *Vlok J.H.J. 846* (NU, PRE); between two tunnels, Montagu Pass, Railway (-CC), *van Berkel N. 1117* (BOL); Outeniqua mountains, upper southern slopes of Ruitersberg (-CC), 30 Nov 1986, *Vlok J.H.J. 1734* (PRE); Kaaimans River Pass, north of river (-DC), 4 Oct 1981, *Stirton C.H. 9681* (K, PRE). **3323 (Willowmore)**: Helpmekaar peak, Uniondale (-CA), 28 Jan 1941, *Esterhuysen E. 4609* (BOL). EASTERN CAPE: **3324 (Steytlerville)**: Mountain slope, above Witte Els Bosch (-CC), Sep 1920, *Fourcade H.G. 924* (BOL, GRA, K, NBG, PRE); Nov 1913, *Muir J. 125*0 (BOL, PRE). **Without precise locality:** Addo road, 28 Sept 1030, *Long F.R. 132* (K).

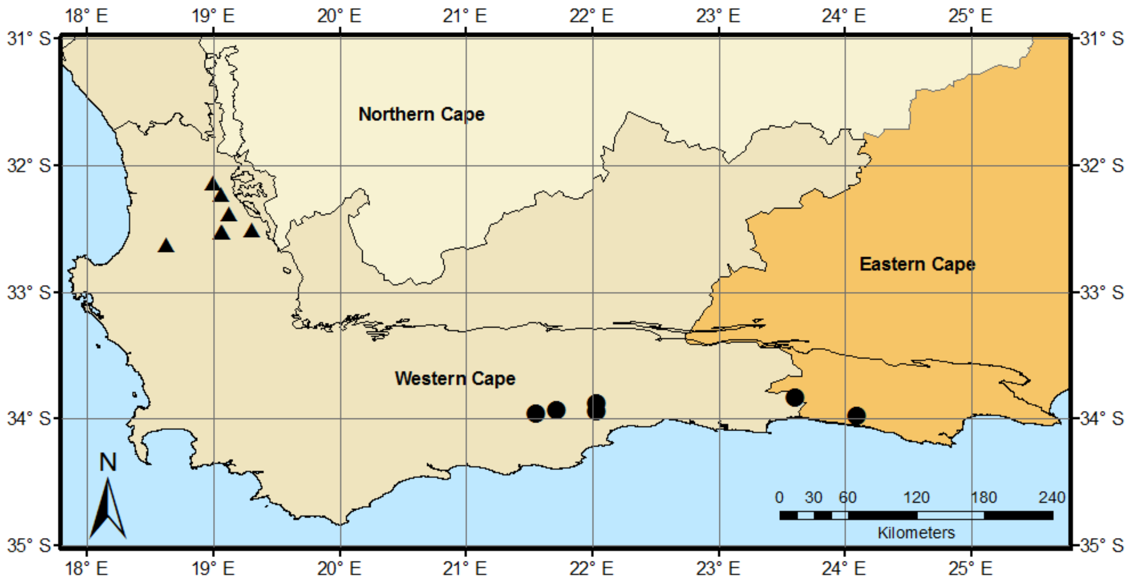

Figure 33. Known distribution of *P. flocc*osa (circles) and *P. intonsa* (triangles).

4.6.2.21. Psoralea intonsa **C.H. Stirt., Muasya & A. Bello *sp. nov.*,** Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); *Psoralea* sp. 26, Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Type: South Africa, Western Cape, Wuppertal (3219): Cederberg path, between Jurie se Berg and Middleberg (-AC), 10 Dec 1982, *Taylor H.C. 10526* (K!, holo.; NBG!, iso.).

Robust shrub up to 2 m tall, reseeder. *Stems* erect to spreading, stiff, single, branching from near base, rigid, greyish brown with white storied lenticels. *Seasonal shoots* densely pubescent with white hairs, greenish white, soon glabrescent, persisting as they age. *Leaves* (7)9−11-foliolate, imparipinnate, distributed along the branches, congested, petiolate; leaflets 10–11×0.3–0.4 mm, linear lanceolate, crowded, terminal leaflet second longest or equal to the basal pair, basal pair longest, three uppermost digitately inserted; petioles 1–2 mm long, shorter than terminal leaflet; rachis 5−7 mm long, grooved; stipules 1−2×0.7−1 mm, persistent becoming woody, hairy, triangular, acuminate. *Inflorescences* axillary, borne in upper axils of seasonal shoots with or without terminal leaf extension, pseudo-capitate, congested, two, three or more flowers per axil, pedunculate, pedicellate; peduncles stout and rigid, 5–7 mm long; cupulum bilobed, hairy, lobes equally developed, one of the vexillar lobes variously bilabiate, glandular, narrowly triangular; pedicel 1–1.5 mm long. *Flowers* 11−13 mm long, colour uncertain (recorded variously as dark blue, mauve to white to cream), scented. *Calyx* 7−8 mm long, tube 4–4.5×4 mm, sparsely pubescent; lobes unequally developed, longer than the tube, carinal lobe boat shaped, the rest acute. *Standard petal* 10–

12×8–8.5 mm, broadly elliptic, colour uncertain, swollen but not ridged at apex, auricles slightly swollen; claw 3–4 mm long. *Wing petals* 9–11×3–4 mm, colour uncertain, comprised of 2–3 rows of 10–14 transcostal lamellae; claw 3−4 mm long. *Keel* 9–10×3–4 mm, white, apically suffused with dark violet-purple; claw 3–4 mm long. *Androecium* 10–11 mm long, tenth stamen lightly attached, fenestrate. *Pistil* 10 mm long, ovary 2 mm long, sparsely glandular near apex, style 5 mm long straight, then curved upwards, swollen at point of flexure, height of curvature 3−4 mm, stigma stipitate, penicillate. *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* solitary, 2–4×2 mm, dark brown to black (Figure 34)**.**

4.6.2.21.1. Diagnostic characters

Psoralea intonsa is similar to *P. floccosa* in being hairy but differs in its greenish white seasonal shoots (versus glaucous); 7−11-foliolate leaves (versus 9−11(13)-foliolate); terminal leaflet second longest or equal to the basal pair (versus shortest); petioles 1–2 mm long (versus 2–3 mm long); peduncles 5–7 mm long (versus 1–2 mm long); cupulum bilobed (versus trilobed); flowers 11−13 mm long (versus 10–11 mm long); calyx lobes longer than the tube (versus shorter or equal to the tube); and standard broadly elliptic (versus very broadly ovate). The two species are geographically isolated.

Psoralea intonsa is known to hybridize with another widespread Cederberg species *P. oreopola* sp. nov. ined. (Stirton, pers. obs.) in the Heuningsvlei region. Specimens from this area are sometimes difficult to attribute to either species and unfortunately are more common in herbaria than either of the parental species.

4.6.2.20.2. Distribution and habitat

Psoralea intonsa is a montane species endemic to the Cederberg and Piketberg mountains (Figure 33). It occurs along or near seepages in mountain fynbos at elevations between 750–1160 m and is recorded from Olifants Sandstone Fynbos Cederberg Sandstone Fynbos, and Piketberg Sandstone Fynbos vegetation types (FFs 3, FFs 4, and FFs 6) as shown by Rebelo *et al.* (2006).

4.6.2.21.3. Etymology

The specific epithet *intonsa* is derived from the Latin adjective *intonsus –a –um* (bearded or unshaven) and makes reference to the overall hairiness of the species.

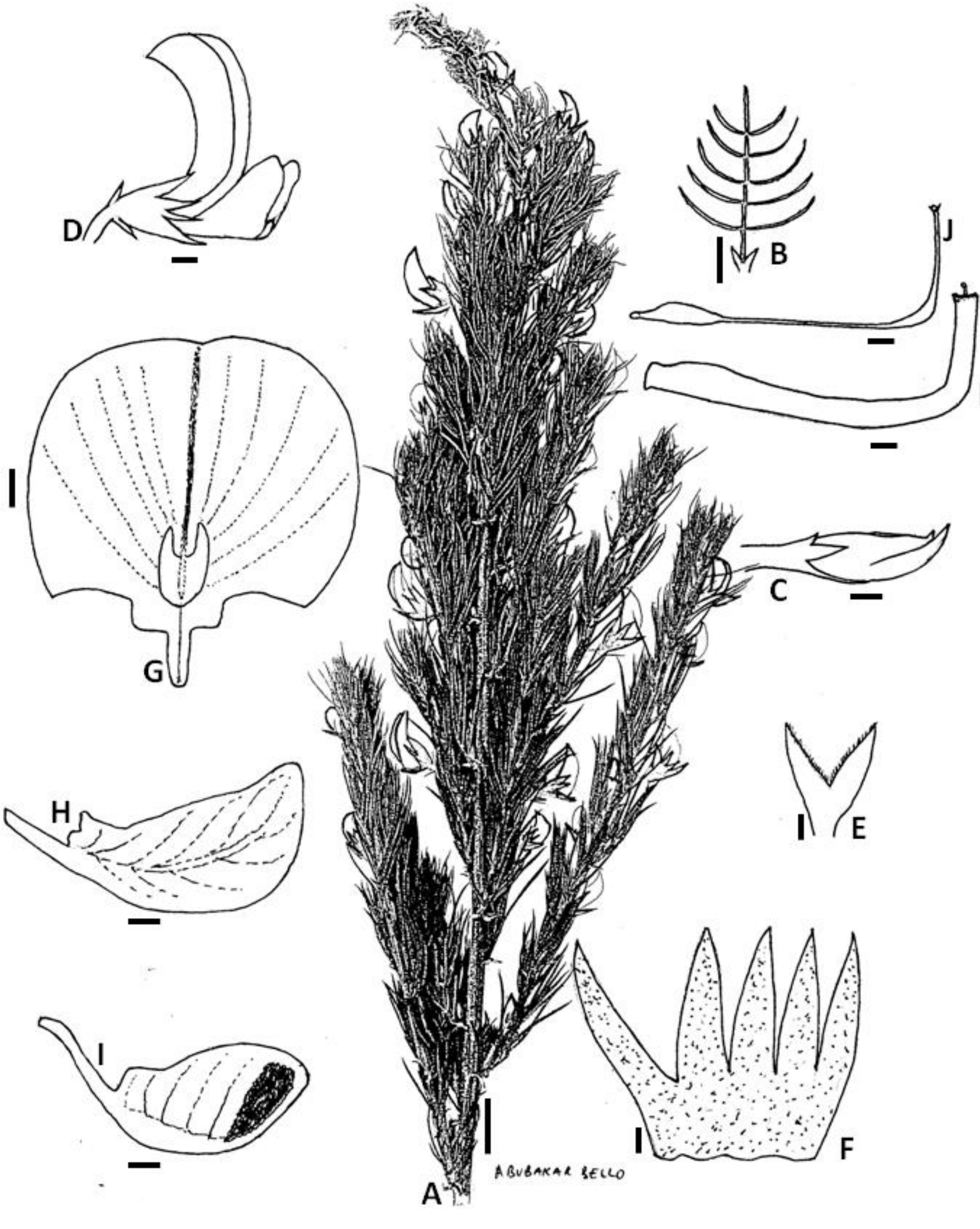

Figure 34. Vegetative and reproductive morphology of *P. intonsa* (A) flowering shoot; (B) 9-foliolate leaf; (C) bud; (D) flower; (E) cupulum; (F) calyx opened out with vexillary lobes to the right; (G) front view of standard petal; (H) wing petal; (I) keel; (J) androecium; (K) pistil. Voucher *A. Bello 112* (BOL). Scale bars: A−B 1 cm; C–K, 1 mm. Artist: Abubakar Bello.

4.6.2.21.4. Conservation status

Psoralea intonsa does not qualify for any category of threat according to the five IUCN criteria (von Staden *et al.* 2009) so is evaluated and assigned a least concern (LC) status. It is common wherever it occurs and is not currently under any threat, except through local hybridization wherever the habitat has been disturbed.

4.6.2.21.5. Specimens studied

South Africa. WESTERN CAPE: **3218 (Clanwilliam)**: Aurora, Engelsman Se Baken, Slopes southwest of Radar station (-DA), 28 Sep 1982, *Oliver E.G.H. 7916* (PRE, NBG): Gryskop, Piketberg (-DA), 2 Dec 1981, *Stirton C.H. 10057* (PRE); Between Grootplaat and Gys-se-kraal, above Aurora (-DA), 19 Oct 1986, *Rourke J.P. 1865* (PRE). **3219 (Wuppertal)**: Cederberg mountains, Krakadouwsberg (-AA), 30 Dec 1941, *Esterhuysen E. 7511* (BOL); Top of Pakhuis Pass (-AA), *Stirton C.H. 10191*(NU); Saron (-AA), Nov 1941, *Stokoe T.P s.n.* (PRE); Heuningvlei, Pakhuis Pass, Cedarberg Mountains (-AA), 4 Aug 2011, *Stirton C.H. & Muasya A.M. 13211* (BOL); Pakhuis Pass, 0.2 km south of Kliphuis River bridge, Clanwilliam (-BB), 25 October 2008, *Helme N.A. 5746* (NBG); Pakhuis Pass path between Jurie se berg and Middleberg Hut (-AC), 10 Dec 1986, *Taylor H.C. 10526* (PRE, NBG); Middleberg plateau (-AC), 14 Dec 1941, *Compton R.H. 12717* (NBG); Pienaarsvlak, above Algeria, Cedarberg (-AC), 7 Nov 1986, *Stirton C.H. & Žantovská J.Z. 11490* (K); Top of Duiwelskop near Citrusdal (-CA), Jan 1945, *Stokoe T.P. 9239* (BOL); Hoek se Kloof, next to road in the vicinity of Vaalkloof, Allendal, Citrusdal (-CA), 12 Nov 2001, *Hanekom W.J. 3372* (K, NBG).

4.6.2.22. *Psoralea odoratissima* Jacq., Hort. Schoenbr. 2: 54, t.229 (1797); Dietr., Lex. Gart. Bot. 7: 611 (1807); DC., Prodr. 2: 216 (1923); G. Don, Gen. hist. 2: 201 (1832); Walp. in Linnaea 13: 512 (1839); Reichb., Ic. Pl. Descrip.: 97, t.2 (1821); Harv. in Harv. & Sond., Fl. Cap. 2: 144 (1862); Forbes in Bothalia 3: 124 (1930); Bond & Goldblatt, Pl. Cape Flora: 296 (1984); Hall & Veldhuis, S. Afr. Red Data Book: 85 (1985). Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012). Lectotype: Cultivated at Hortus Schoenbrunn, *Anonymous s.n.* (BM000901311!, Lecto. here designated). [Note: No potential type material was found for this species at OXF, S, W or LINN, the only material that we have found is at BM (labelled “ex Hortus Schoenb.”) and PRC (labelled “ex Herb Zahlbruckner”). Typification of Jacquin species is difficult. It is known that N-J. Jacquin (Snr.) sold a substantial portion of his herbarium to Joseph Banks in London (D’Arcy, 1970), so it is possible that this specimen could have been part of the material which Jacquin used to describe his species. However, BM000901311 may not have been the actual material that was used for the coloured illustration as it is not a good match of the plate. Specimens were often gathered from the same plant over a number of years and distributed to friends and institutions and so it is uncertain whether this applied here. The PRC material is a closer match to the plate t.229 and, from its robustness, is certainly gathered from cultivation but one of the sheets is dated “11 Mai 1842, Herb. Monar.” The 2nd PRC sheet (1382.2) cites Reichenbach (l.c.) and which was possibly used for his illustration. The BM material is currently the best material available hence our decision for designating it as the lectotype].

Robust tall shrub or tree up to 4(6) m tall, reseeder. *Stems* erect, single, branching from about 1.5 m tall, rigid, yellowish green with numerous white lenticels when young, aging grey, striate. *Seasonal shoots* green, shaggy with lax white hairs. *Leaves* (13)15–19-foliolate, imparipinnate, distributed along the branches, incurved and clasping when young but then spreading, petiolate; leaflets 13−19×0.5−2 mm, linear lanceolate, deeply channelled abaxially, rounded adaxially, cuneate at the base, tapering thereafter, apex acuminate, tips arching, hairs shaggy, thicker on adaxial midvein; secondary veins sunken, visible; basal pair longest becoming smaller towards the apex, three uppermost pinnately inserted; with many raised glands; petioles 4–5 mm long; rachis 15−50 mm long; stipules 2–3×1.5–2 mm, 3–4-veined, shorter than but not fused to petiole, persistent, rapidly senescent, semi-erect and inrolled, lanceolate-ovate, acute, ciliate. *Inflorescences* axillary, borne mainly in the upper axils of seasonal shoots, no leafy extension, pseudo-spicate, lax, three flowers per axil, pedunculate, as long as the subtending leaf, pedicellate; peduncles stout and rigid, 3–6 mm long; cupulum bilobed, hairy, lobes equally developed, triangular; pedicels 2–3 mm long. *Flowers* 13−14 mm long, white, intensely sweet-scented. *Calyx* 5–7 mm long, tube 2–3×4 mm; lobes unequally developed, longer than the tube, pale green, sparsely covered in small white flat hairs and densely encrusted with mixed sized glands on outside; margins of teeth densely black ciliate, inside of teeth densely stubby black-haired. *Standard petal* 10–11×8–10 mm, broadly ovate, white to pale mauve with purple nectar guide at the crotch, back of standard darker; claw 3–4 mm long. *Wing petals* 10–11×3–3.5 mm, white with auricles well developed, sculpturing present, basal comprised of 2−3 rows of 10−12 transcostal lamellae; claw 3–4 mm long. *Keel* 9–10×2–3 mm, white, apically suffused with dark violet-purple on inner face; claw 4–5 mm long. *Androecium* 10 mm long, tenth stamen free, scarcely fenestrate. *Pistil* 8 mm long, stipitate; ovary 1 mm long, glabrous, sparsely covered in club-shaped glands; style 6 mm long straight, then upcurved, height of curvature 3 mm long, thickened at point of flexure. *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* solitary, 2–4×2 mm, dark brown to black (Figure 35).

4.6.2.22.1. Diagnostic characters

Psoralea odoratissima is similar to *P. suaveolens* in being the only species in the genus with more than 13-foliolate leaves. They differ in that *P. odoratissima* ranges from a single-stemmed, erect, large shrub to a 5–6 m tall tree with a stem diameter of about 20 cm (versus slender virgate shrub up to 2 m tall with stem usually < 10 cm in diameter), with up to 19-foliolate, broader up to 2 mm wide leaflets (versus 13–15(17)-foliolate leaves with narrower leaflets < 2 mm wide); and shorter peduncles and pedicels, less than 15 mm long (versus very long peduncles and pedicels, usually up to 45 mm long).

Psoralea odoratissima exhibits a cline in flower colour being pure white at the western end of its distribution becoming progressively more mauve to pale purple towards the east. The species is pollinated by Xylocopid bees (Stirton, pers. obs.). Flowering appears to be bimodal with peaks in October and January.

4.6.2.22.2. Distribution and habitat

Psoralea odoratissima extends from the Langeberg mountains, east of Robertson to Seweweekspoort (Worcester and Uniondale regions) in the Western Cape Province. It occurs in seasonal seepages, gulleys, valley bottoms and streamsides in mountain fynbos at elevations between 300–1330 m (Figure 36). *Psoralea odoratissima* is found in Central Coastal Shale Band Vegetation, Breede Shale Fynbos, North Langeberg Sandstone Fynbos and South Langeberg Sandstone Fynbos Vegetation types (FFb 4, FFh 4, FFs 15, and FFs 16) (Rebelo *et al.* 2006).

4.6.2.22.3. Etymology

The specific name *odoratissima* is derived from the Latin word *odoratissimus –a –um,* meaning 'strongly fragrant’, and alludes to the intense sweet aroma of its flowers.

4.6.2.22.4. Conservation status

This species is assessed as nearly threatened (NT D2). It occurs in less than 10 known locations many of which are already threatened by alien plant invasion (Raimondo 2009).

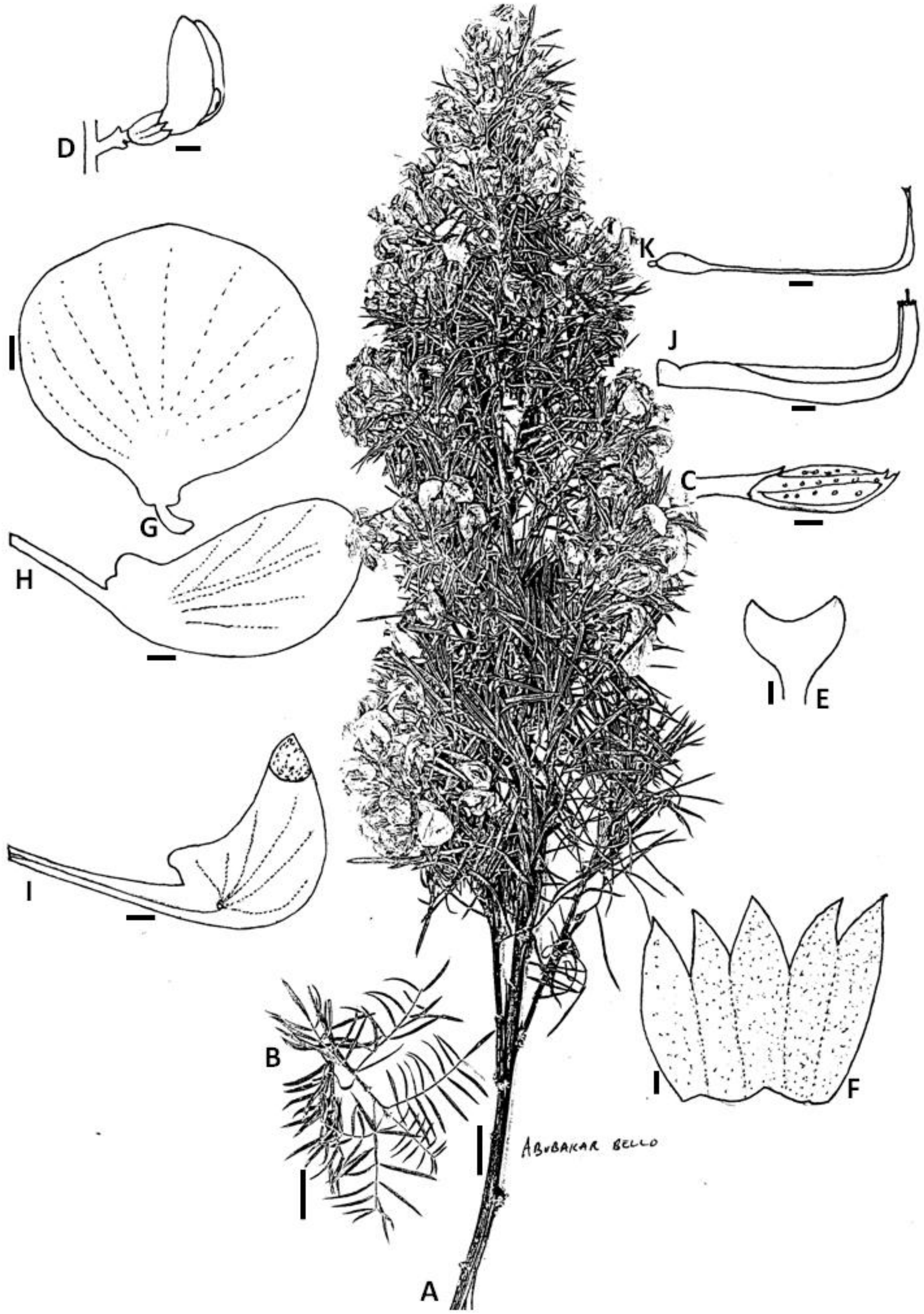

Figure 35. Vegetative and reproductive morphology of *P. odoratissima* (A) flowering shoot; (B) 5-foliolate claw like leaf (C) bud; (D) flower; (E) cupulum; (F) calyx opened out with vexillary lobes to the right; (G) front view of standard petal; (H) wing petal; (I) keel; (J); androecium (K) pistil. Voucher *A. Bello 164* (BOL). Scale bars: A−B 1 cm; C–K, 1 mm. Artist: Abubakar Bello.

4.6.2.22.5. Specimens studied

South Africa. WESTERN CAPE: **3319 (Worcester)**: Upper Dome kloof, Hex Rivier Mountains (-AD), 2 Mar 2003, *Helme N.A. 2644* (NBG); Bergendal farm, lower foothills of Langeberg at Klaasvoogds (-DB), 1 Nov 1983, *Rourke J.B. 1814* (NBG); Rondgangers kraal 29, Suurlemoen se Houtbos, Langeberg east of Robertson (-DD), 4 Oct 2011, *Helme N.A. 7228* (NBG); Bergendal farm, lower foothills of Langeberg at Klaasvoogds (-DD), 1 Nov 1983, *Rourke J.P. 1814* (PRE). **3320 (Montagu)**: Doornkloof, Montagu (-CC), 26 Sep 1946, *Compton R.H. 18457* (NBG); mountains east of Montagu (-CC), 28 Oct 1954, *Esterhuysen E.E. 23837* (BOL); Donkerkloof (-CD), 26 Sept 1946, *Levyns M.R. 8049* (BOL); N–Facing slopes of Langeberg along path between Goedgeloof Hut and Warmwater rivier (-CD), 11 Apr1987, *McDonald 1453* (NBG); 2 km from Barrydale to Montagu (-DC), 20 Oct 1972, *Loxton A.G. 223 (*PRE); Dwarriegarivier (-DC), 21 Nov 1980, *Stirton C.H. 8258* (NU, PRE); Cogmanskloof (-DC), 27 Dec 2007, *Muasya A.M. & Stirton C.H. 3532* (BOL); Oct 1922, *Michell M.R. 89* (PRE).

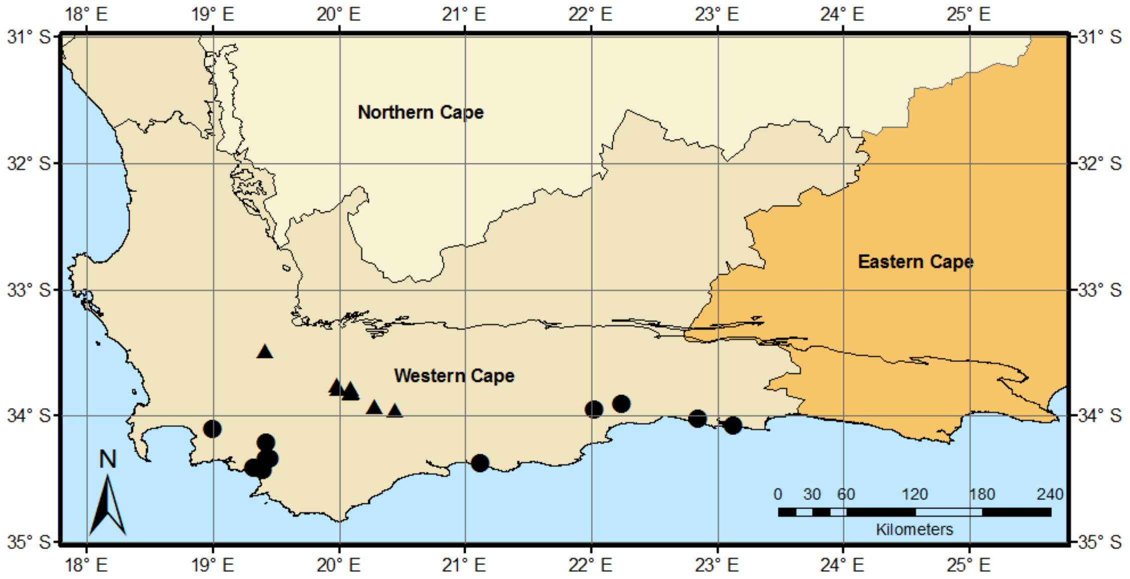

Figure 36. Known distribution of *P. odoratissima* (triangles) and *P. arborea* (circles).

4.6.2.23. *Psoralea arborea* Sims in Curtis's Bot. Mag. 46: t.2090 (1819); DC., Prodr. 2: 216 (1825); E. Mey. in Linnaea 7: 163 (1832); G. Don., Gen. Syst. 2: 201 (1832); E. Mey., Comm. 82 (1836); Walp., Repert. 1: 655 (1842); Lock, Leg. Afr. Check-list: 458 (1989), non Eckl. & Zeyh. (1836) nec Sesse & Mocino (1889); Moir, Fl. Vict.: 685 (1996); Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012); Stirton *et al.* in Muellaria 33: 97–107 (2015). Type: Bot. Mag. 46: t. 2090 (holo.).

Psoralea pinnata L. var. *quinquejuga* Eckl. & Zeyh., Enum.: 224 (1836). Type: "in humidis (altit. III) laterum montis Duyvelsberg supra Geele Klee (Cap.)" [in wetlands (altit. 3) on the sides of Duyvelsberg mountain above Geel Klee (Cape)], *Ecklon & Zeyher s.n.* (not found) [Note: Walpers (1839) synonymised this taxon in *Psoralea arborea* but the type has not yet been found].

Psoralea pinnata sensu Palmer & Pitman, *Trees of Southern Africa* 2: 918–919 (1976).

Psoralea affinis sensu Hutchinson, *Bot. Mag*. 136: t.8331 (1910) non Eckl. & Zeyh. (1836).

Verdcourt (2000) refers to this species in the Flora Zambesiaca but his species is *P. latifolia* (Harv.) C.H. Stirt.

Large shrub to small tree 3–5 (10) m tall, reseeder, colonial. *Stems* erect, 1(2), rigid, diameter up to 50 cm, green with vertical grey stripes and irregular lenticels when young becoming grey when old with scattered irregularly storied white lenticels, branching from near base when young, shedding branches when older. *Seasonal shoots* green, villoso-pubescent. *Leaves* 7−9(11)-foliolate, imparipinnate, spreading, glabrescent above, wispily hairy below, petiolate; leaflets 30–50×1–2 mm, terminal leaflet shortest, basal pair longest, decreasing in length towards the apex, uppermost three leaflets digitately inserted, linear or linear lanceolate, terete to flattish, acute, base rounded, glandular, adaxial surface grooved, abaxial surface convex, vein on adaxial surface prominent; green, cavities slightly sunken in fresh state, petioles 5–8 mm long, shorter on younger leaves; rachis 10–12 mm long; stipules 2−3×1−2 mm, persistent, shorter than petiole, fused for part of their length, margins inrolled, ovate, hairy, rapidly senescent, callous, recurved. *Inflorescences* axillary, well exerted from leaves, borne in upper axils of seasonal shoots without leafy extension, 1–3-flowered per axil, pseudo-capitate, congested or laxly clustered in axils, pedunculate, pedicellate; peduncles stout and rigid, 45–60 mm long, longer than the subtending leaf, hairy; cupulum bilobed, minutely hairy at the margins of equally developed lobes, covering base of calyx, broadly triangular, senescent becoming yellowish and papery, glands turn orange from green; pedicels 2–4 mm long, black-haired. *Flowers* (10)13–15(19) mm long, very broadly ovate, deep blue to purple. *Calyx* 6−8 mm long, tube 4–5×4.5 mm, densely hairy, mainly black-haired with or without occasional white hairs; lobes equally developed, shorter than the tube, blackish, lateral and vexillar large, reflexed, emarginate, acuminate, fused for about one third their length, carinal same width as other four, small, incurved. *Standard petal* 15–16×(13.5–

)14.5–15 mm, very broadly ovate, swollen but not ridged at apex, auricles slightly swollen, deep mauve to purple with large M-shaped white nectar guide with a dark purple flash in crotch, paler towards margins, veins darker than blade; claw 4–6 mm long, deeply channelled. *Wing petals* 14×5 mm, widest at two-thirds its length, mauve, blade folded and puckered along its mid-line, sculpturing present, basal, comprised of 2–3 rows of 10–14 transcostal lamellae; claw 4 mm long. *Keel* 10–11×3.5–4 mm, blade ridged for 3 mm from base, white; apex purple on inner face; claw 5 mm long. *Androecium* 12 mm long, tenth stamen lightly attached, fenestrate. *Pistil* 12 mm long, ovary 3 mm long, style 6 mm long, upper end covered in club-shaped glands, height of curvature 4 mm, widest at point of flexure, stigma penicillate. *Fruits* oblong. 4–5×3 mm. *Seeds* solitary, 4.5×2.8 mm (Figure 37).

4.6.2.23.1. Diagnostic characters

Unlike most other species in the complex, *P. arborea* can grow up to a height of 10 m wherever it is protected from fire for long periods. It is similar to *P. brilliantissima* in growth habit but differs in its erect, tall, large trunks sometimes up to 50 cm in diameter (versus large robust shrubs up to 2.5 m with the trunk less than 20 cm in diameter), longer and wider 30–50×1–2 mm leaflets (versus 30–35×0.3–0.6 mm), longer petioles, 5–8 mm long (versus 3–4 mm long), inflorescences 1–3-flowered per axil (versus 3 or more flowers per axil), longer peduncles, 45–60 mm long (versus 25–35 mm long), standard petal larger 15–16×(13.5)14.5–15 mm, very broadly ovate, and deep mauve to purple (versus 10–12×8.5–9.5 mm, broadly ovate, and intensely hyacinth blue with mauve margins), calyces covered densely by mainly black silky hairs (versus covered in a less dense mixture of black and white hairs).

4.6.2.23.2. Distribution and habitat

Psoralea arborea is primarily a lowland species of the Western Cape Province and ranges from Hermanus and Stanford in the east to Mossel Bay in the west but also occurs in fire-protected valleys of the Koudberg Mountains and Potberg. It is found mostly in marshy areas, stream sides, forest margins and forests in protected valleys between 30–330 m elevations (Figure 36). It is associated with Knysna Sand Fynbos, Canca Limestone Fynbos, Overberg Sandstone Fynbos, South Outeniqua Sandstone Fynbos, Southern Afrotemperate Forest and Overberg Dune Strandveld types (FFd 10, FFl 3, FFs 12, FFs 19, FOz 1 and Fs 7) (Rebelo *et al.* 2006).

4.6.2.23.3. Etymology

The specific epithet *arborea* is derived from the Latin adjective *arboreus –a –um* (relating to trees) and alludes to the tall tree-like nature of this species.

4.6.2.23.4. Conservation status

This species has been given an automated status of least concern (LC) (Raimondo *et al.* 2009, version 2014:1). Based on the current study it has been found to occur in small scattered subpopulations though only the forest margin or forest tree-like forms are threatened by frequent fires. It has started to spread sporadically along roadsides in the western part of its distribution; attributable to extensive road building. Based on the South African criteria for rarity (Von Staden *et al.* 2009 we rank it as Rare. The species has naturalised in Australia and New Zealand (Stirton *et al.* 2015).

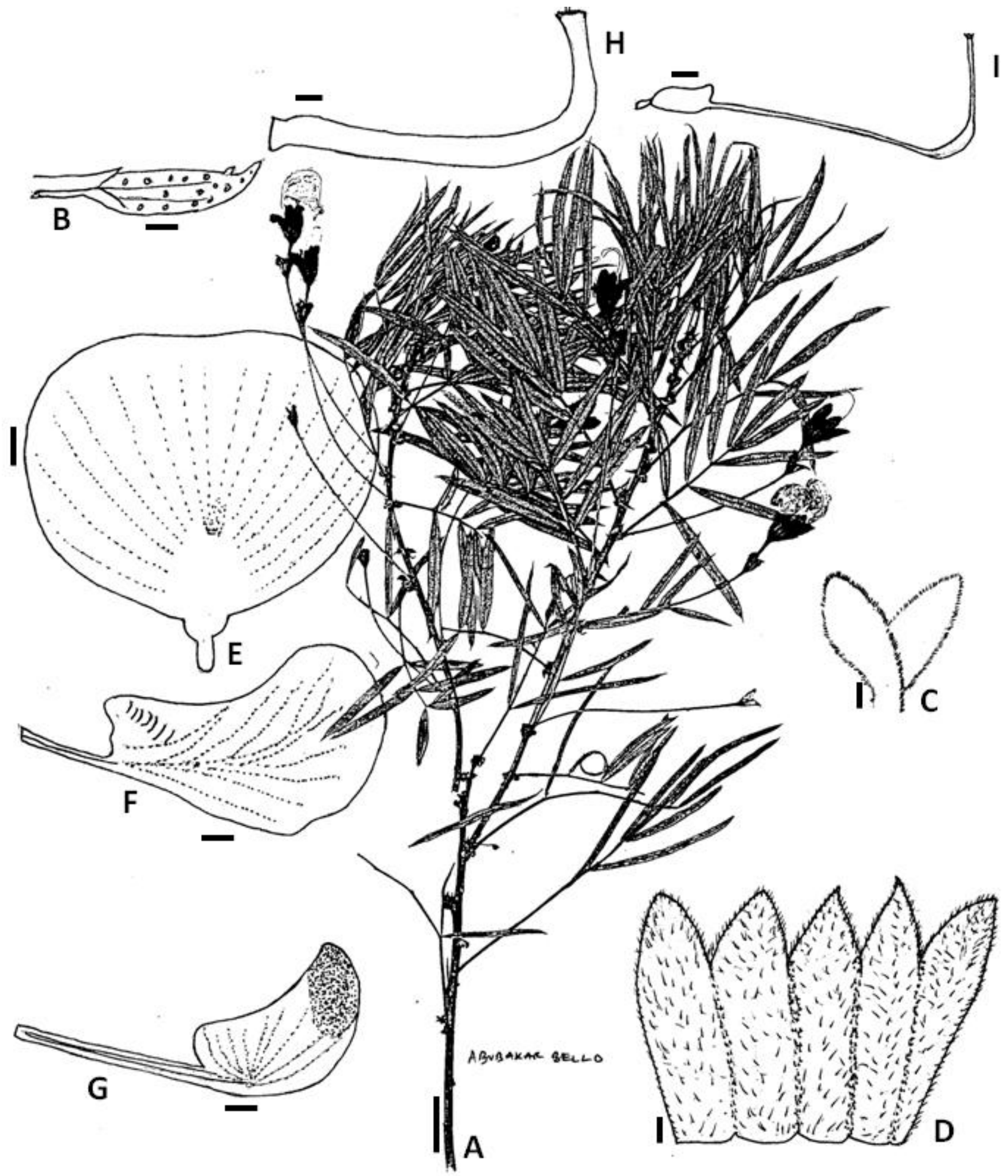

Figure 37. Vegetative and reproductive morphology of *P. arborea* (A) flowering shoot; (B) bud; (C) cupulum; (D) calyx opened out with vexillary lobes to the right; (E) front view of standard petal; (F) wing petal; (G) keel; (H) androecium; (I) pistil. Voucher *A. Bello 149* (BOL). Scale bars: A, 1 cm; B–I, 1 mm. Artist: Abubakar Bello.

4.6.2.23.5. Specimens studied

South Africa. WESTERN CAPE: **3322 (Oudtshoorn)**: Varingsrivier, between Grootbrakrivier and Kleinplaas (-CC), *Joffe H. 882* (PRE); Jonkersberg (-CC), 04 Dec 1951, *Esterhuysen E. 19401* (BOL). **3323 (Willowmore)**: Deepwalls, Knysna (-BB), Jan 1924, *Phillips J.F.V. 403* (PRE); Deepwalls Forest Reserve (-CC), 07 Jul 1955, *Bird W.R. 1007*

(PRE); 18 Jun 1961, Van Breda *P.A.B. 1158* (K). **3419 (Caledon):** Vogelgat Private Nature Reserve, Hermanus (-AC), 4 Dec 1980, *Stirton C.H. 8477* (PRE); Klippies Baai, Voelklip, Hermanus (-AD), 8 Aug 1980, *Williams S.L. 383* (K, MO); Fernkloof Nature Reserve (-AD), 30 Sep 1981, *Bean P.A. 700* (BOL); near turnoff to Elim from Gansbaai / Pearly Beach road (-CB), 31 Oct 1985, *Snijman D. 936* (K, NBG); roadside on the farm Strandkloof (-CB), 18 Aug 1995, *Peterson-Jones J.C. 503* (NBG); Kraaibos, roadside near a Marsh (-CB), 6 Oct 1981, *Stirton C.H. 9743* (PRE); Pearly Beach (-CB) *Verboom A.G. 1309* (BOL); Farm Heidehof (-CB), 17 Nov 2007, *Muasya A.M. & Stirton C.H. 3212* (BOL); near Bruinklip on road from Paviesvlei to Avoca (-DA), 6 Oct 1981, Stirton C.H. 9735 (K, PRE); before Avoca from Paviesvlei river valley (-DA), 6 Oct 1981, *Stirton C.H. 9740* (K, PRE). **3421 (Riversdale)**: Mosselbank Fontein, near Funtjie valley, inland of first major dune ridge (-AC), 12 Sep 1976, *Boucher C. 3199* (NBG). **Cultivated material:** Tresco Abbey, Scilly Isles 30 Apr 1909, *Anonymous s.n.* (K); Scilly Isles, 31 Mar 1909, *Smith D. s.n.* (K).

4.6.2.24. *Psoralea montana* A. Bello, C.H. Stirt., & Muasya *sp. nov.* Type: South Africa, Eastern Cape, Port Elizabeth (3325): Longmore Forest Station (-CC), 23 Jan 2009, *Muasya A.M. & Stirton C.H. 3823* (BOL!, holo.).

Tall shrub to small tree up to 5 m tall, reseeder, forming colonies. *Stems* erect, single, branching from halfway up or higher up the stem, rigid, branches striate, glabrous, woody, round, yellowish brown to brown with white stratified lenticels. *Seasonal shoots* sparsely pubescent, greyish green to purplish, striate. *Leaves* 7−9-foliolate, imparipinnate, spreading, distributed along the seasonal branches, erect, petiolate; leaflets 20–50×1.5–2.5 mm linear to linear-lanceolate, semi-flat with distinct mid-rib, apiculate at the apex, cuneate at the base, with sparse silvery pubescence, terminal leaflet second longest, basal pair longest, three uppermost pinnately inserted; petioles 5–10 mm long, shorter than terminal leaflet; rachis 15–20 mm long; stipules 2–2.5×2–1.5 mm persistent, shorter than petiole, fused, callused, twisted and recurved, ovate-lanceolate, acuminate, caducous. *Inflorescences* axillary, borne in upper axils of seasonal shoots without leafy extension, pseudo-capitate, fasciculate, 1−5 flowers per axil, pedunculate, pedicellate; peduncles stout and rigid, 25–45 mm long, longer than the subtending leaf; cupulum bilobed, bristly pubescent with silvery hairs, 2 mm long; lobes equally developed, triangular; pedicels 1–3 mm long. *Flowers* 13−14 mm long, mauve. *Calyx* 6–8 mm long, tube 3.5–4.5×4.5 mm with sparse glands on the surface, glands constant in size; lobes sub-equally developed, shorter than the calyx tube, oblong-ovate, sub-acute or rounded at the apex, lower lobe largest; finely silvery pubescent outside, black stubby hairs within, margins ciliate. *Standard petal* 10–12×8–9.5 mm, very broadly ovate, mauve but paler at the margins, with a large white M-shaped nectar guide and a small purple flash in crotch; claw 3–5 mm long. *Wing petals* 9–10×3–4 mm, mauve, up-curving, auricles and sculpturing present, basal comprised of 2–3 rows of 10–14 transcostal lamellae; claw 3–4 mm long. *Keel* 8–9×3.5–4 mm, blade ridged for 3 mm from base, white, inner face of apex purple; claw 4 mm long. *Androecium* 10 mm long, tenth stamen free, scarcely fenestrate. *Pistil* 8 mm long, stipitate, ovary 1 mm long, glabrous, sparsely covered in club-shaped glands, style 6 mm long straight then upcurved, thickened at point of flexure, height of curvature 3 mm long. *Fruits* oblong, 5.0×2.5–3.0 mm, finely reticulate, papery. *Seeds* one, 4.0×2.5 mm, ellipsoid, hilum sub-central, dark brown (Figure 38).

4.6.2.24.1. Diagnostic characters

Psoralea montana is most closely similar to *P. affinis* in having the same number of leaflets, stalked axillary inflorescences and pubescent calyces. It differs in being a tall montane shrub or tree up to 5 m tall, with rounded brown stems with, if present, stratified lenticels (versus an erect lowland shrub or treelet to 3 m tall, usually with greyish stems and storied lenticels), seasonal shoots greyish green, sparsely pubescent (versus green, nigro-hirsute seasonal shoots), leaflets broader 1.5−2.5 mm wide (versus 0.2−0.4 mm wide), terminal leaflet second longest (terminal leaflet shortest), petioles 5–10 mm long (versus 4–5 mm), inflorescences 1-flowered per axil but aggregated in terminal pseudo-capitate clusters in upper axils of seasonal shoots (versus 2−many flowers per axil but aggregated in terminal pseudo-spicate clusters in upper axils of seasonal shoots), and calyces finely silvery pubescent (versus sparsely to densely black-haired). *Psoralea montana* is predominantly montane whereas *P. affinis* occurs in lowlands (100–1800 m).

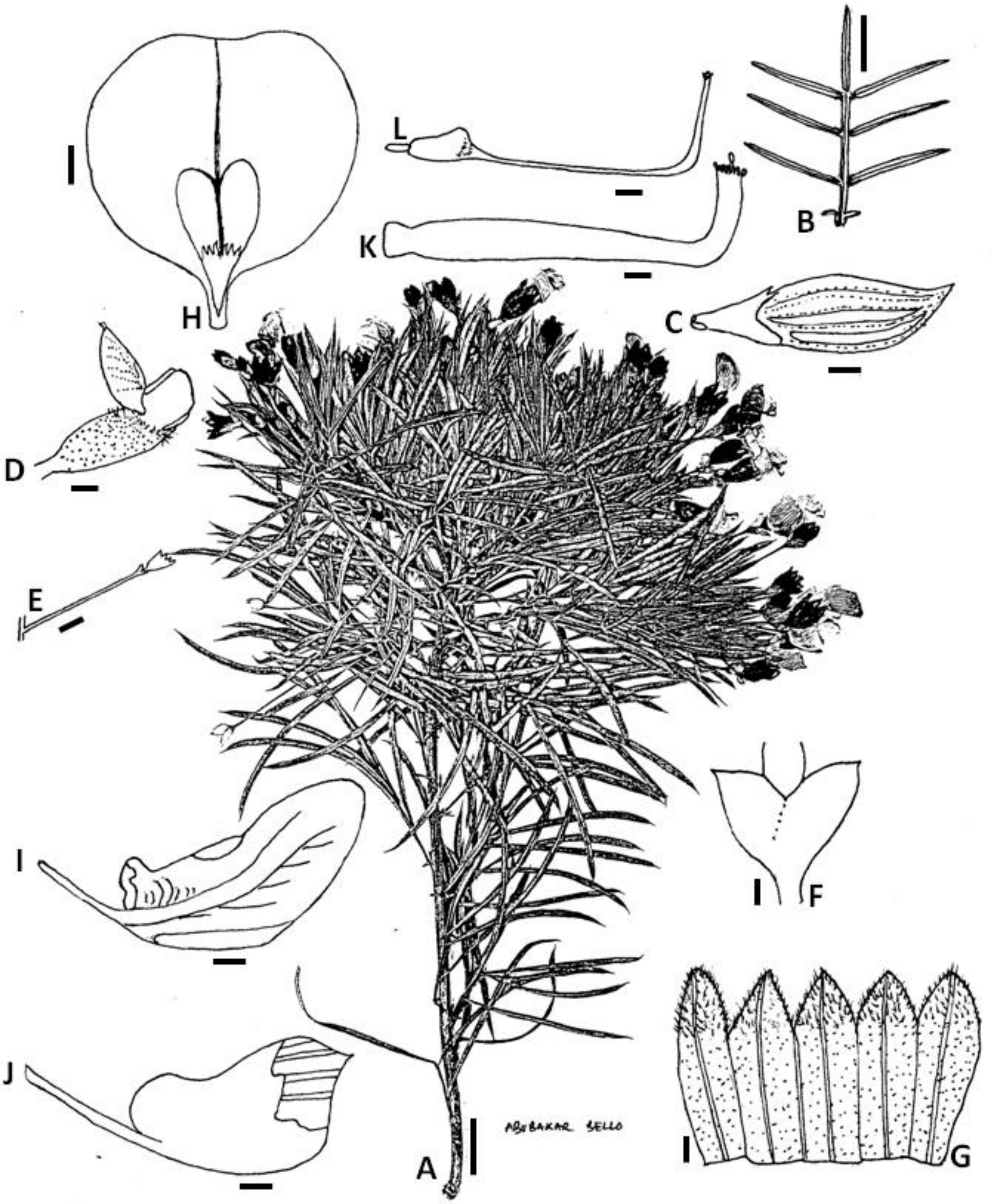

Figure 38. Vegetative and reproductive morphology of *P. montana* (A) flowering shoot; (B) 7-foliolate leaf; (C) bud; (D) flower; (E) peduncle; (F) cupulum; (G) calyx opened out with vexillary lobes to the right; (H) front view of standard petal; (I) wing petal; (J) keel; (K) androecium; (L) pistil. Voucher *A. Bello 125* (BOL). Scale bars: A−B, 1 cm; C–L, 1 mm. Artist: Abubakar Bello.

4.6.2.24.2. Distribution and habitat

Psoralea montana is a handsome montane species distributed along the mountains from George to Humansdorp (Figure 39). Unusually in the complex, it is not associated with seepages and streambeds and is found more generally in fynbos on rocky hillsides and more latterly along roadsides. It has been recorded in the North Outeniqua Sandstone Fynbos, South Outeniqua Sandstone Fynbos and Eastern Little Karoo vegetation types (FFs 18, FFs 19 and SKv 11) (Rebelo *et al.* 2006 and Rutherford *et al.* 2006).

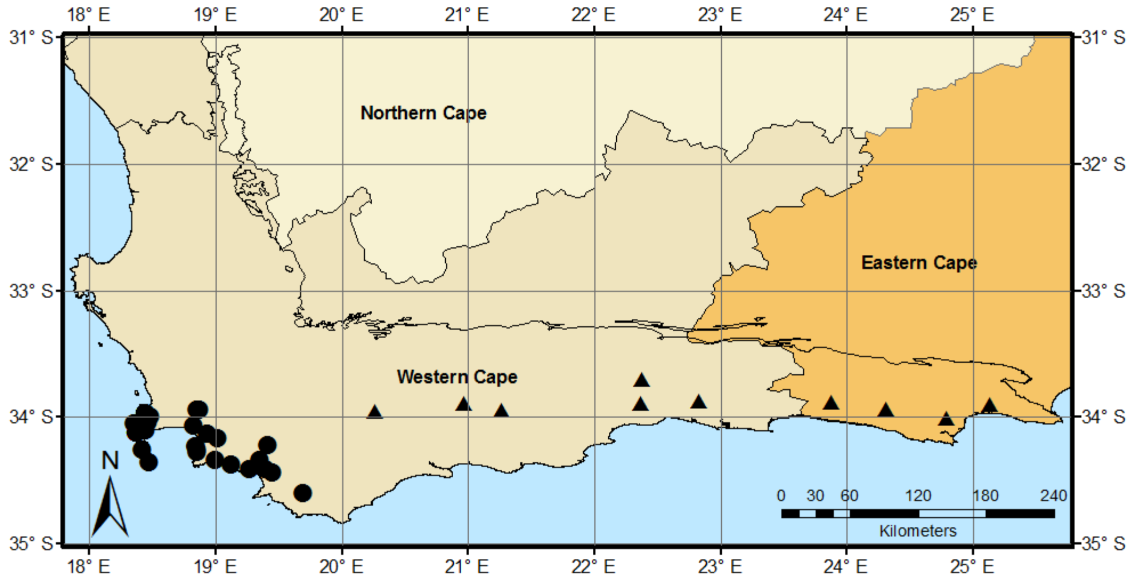

Figure 39. Known distribution of *P. montana* (triangles) and *P. pinnata* (circles).

4.6.2.24.3. Etymology

The specific epithet *montana* is derived from the Latin adjective *montanus –a –um* (dwelling on or found on mountains) and refers to the main altitudinal zone of occurrence of this species.

4.6.2.24.4. Conservation status

This species is assigned the status of least concern (LC) (IUCN 2012a, 2012b) as it is abundant and has a wide range of distribution. It is a widespread species in its area of distribution, often forming large colonies. In some localities it is threatened by too frequent fires and alien vegetation.

4.6.2.24.5. Specimens studied

South Africa. WESTERN CAPE: **3320 (Montagu)**: Kamdebooberg, Montagu Pass (-AC), 3 Jan 2008, *Muasya A.M. & Stirton C.H. 3609* (BOL); Tradouw Pass (-DC), 26 Oct 1986, *Stirton C.H. & Žantovska J.Z. 11258* (K, NBG, PRE); 26 Oct 1986, *Stirton C.H. & Žantovska J.Z. 11280* (K, NBG). **3321 (Ladismith)**: Garcia's Pass (-CC), 2 Dec 2009, *Muasya A.M. & Stirton C.H. 5090* (BOL). **3322 (Oudtshoorn)**: Near Kleinplaat Forestry Station, George District (-CB), 16 Sep 1970, *Wurts T.M. 2261* (NBG) Homtini Pass, George (-DD), 30 Sep

1962, *Walters M. 120*, *609* (NBG); The George, Knysna (-DD), 13 Nov 1949, *Morris G.D. 409* (NBG). **3323 (Willowmore)**: Top of Prince Alfred Pass (-CC), 20 Dec 2009, *Muasya A.M. 5281*, *6648* (BOL); Kom–se–pad, 3 km from Gouna to Deepwalls (-CC), 17 Feb 2007, *Muasya A.M. & Stirton C.H. 4412* (BOL); Bluelillies Bush Forest Station, East of Stormsriver, Willowmore (-DD), 24 Sep 1969, *Marsh J.A. 1342* (NBG); 31 km from N2 to Prince Alfred Pass (-DD), 24 Jan 2008, *Muasya A.M. & Stirton C.H. 3843* (BOL). **3422 (Mossel Bay)**: Groen Vlei, Wilderness George (-BA), 2 Oct 1949, *Martin B.E. 750* (NBG). **3423 (Knysna)**: Noetjie, Knysna (-AA), Sep 1948, *Middlemost G.J.* s.n. (NBG). EASTERN CAPE: **3324 (Steytlerville):** Assegaibosch (-CD), 1 Sep 1912, *Rogers F.A. 2034* (K); Winterhoek Mountains, near Port Elizabeth (-DB), 12 Sep 1930, *Fries C.E., Norlindh T. & Weimarck H. 870* (K); Elandsrivierberg, Otterford Nature Reserve (-DB), 9 Aug 1973, *Thompson M.F. 1835* (K, NBG); Otterford Forest Reserve (-DB), 31 Aug 1947, *Rodin R.J. 1130* (K). **3325 (Port Elizabeth):** Longmore Forest Reserve (-CC), 19 Feb 2009, *Muasya A.M. & Stirton C.H. 4420*; 23 Jan 2001, *3823* (BOL). **3424 (Humansdorp):** Humansdorp (-BB), 13 Sep 1974, *Bayliss R.D.A. 6822* (K).

***4.6.2.25. Psoralea pinnata* L.** Sp. Pl. 2: 1074 (1762); Berg., Descr. Pl. Cap. 218 (1767); Mant. Pl.: 225 (1767); Burm. f., Cap. 22 (1759); Willd., Sp. Pl. 3: 1342 (1764); Ait., Hort. Kew, edn. 1, 4: 374 (1789); Thunb., Prodr.: 136 (1800) & Fl. Cap.: 609 (1823); Link, Enum. 2: 257 (1822); Sweet, Hort. Brit.: 170 (1826); Poir. in Lam., Encycl. 5: 690 (1804); Dietr., Lex. Gart. Bot. 7: 612 (1807); DC., Prodr. 2: 216 (1825); E. Mey. in Linnaea 7: 163 (1832); Eckl. & Zeyh., Enum.: 224 (1836); E. Mey., Comm. 82 (1836); H. Richter, Codex: 739 (1840); Walp., Repert. 1: 655 (1842); Reichenb., Ic. Descr. Pl. Cult.: t.97, f.1 (1823); Harv. in Harv. & Sond., 2: 144 (1862); Bews, Introd. Fl. Natal Zulu. (1921); Forbes in Bothalia 3: 125 (1930); Salter in Adamson & Salter, Fl. Cape Penins.: 485 (1950); Kidd, Wild Flow. Cape Penins.: t.81.12 (1972); Moll, Trees Natal 485 (1981); Bond & Goldblatt, Pl. Cape Flora: 296 (1984); Gibbs Russell *et al.* Mem. Bot. Surv. S. Afr. 56: 88 (1987); Stirton & Schutte in Goldblatt & Manning, Strelitzia 9: 505 (2000); Stirton & Schutte in Manning & Goldblatt, Strelitzia 29: 575 (2012); Stirton *et al.* in Muellaria 33: 97–107 (2015). Type: Collector unknown (Hort. Cliff. 370.1!): *Dorycnium* 1A, (lecto!. BM BM000646705!) designated by Stirton *et al.* in Muellaria 33: 97 (2015).

Ruteria pinnata (L.) Moench., Meth. 115 (1794). *Lotodes pinnatum* (L.) Kuntze, Rev. Gen. 3, 2: 65 (1898).

Psoralea foliis pinnatis, linearibus floribus axillaribus. L., Hort. Ups. 225 (1745); Kniph., Bot. Orig. Centaur 12 (1764).

Psoralea foliis pinnatis, foliolis linearibus, acuminatis. Royen, Fl. Leyd. Prodr: 373 (1740).

Dorycnium caule fruticoso, foliis pinnatis, foliolis linearibus. L., Hort. Cliff. 370 (1737).
Spartium africanum. Riv., tetra. 11 (1691).

Genista affinis arbor africana, monospermos, flore caeruleo, foliis pinnatis. Herm., Hort. Lugd.-bat. Cat. 272, t.273 (1687).

Genista spartium caeruleum, africanum, foliis pinnatis. Breyne, Prodr. Rar. Pl. 1: 36 (1680).

Genista, chamaegenista, africana, frutescens, flore caeruleo. Tit., cat. Maurocen. 77 (1713).
Genista affinis africa, flore caeruleo, alatis et punctatis foliis, asperiusculus. Ten Rhyn., Fasc. 24.

Barba jovis africana, foliis viridibus pinnatis, flore coeruleo. Boerhaave, Ind. alter hort. Lugd.-Bat. 2: 40 (1720).

Psoralea decidua Sieb., nom. nud., sensu auctt., non Berg. (1767).

Ruteria africana Medik., Vorles. Churpfälz. Phys.-Okon. Ges. 2: 380 (1787), nom. illegit.

Psoralea conferta Bertol., Horti bonon. pl. nov. 2: 17, t.II.2 (1839). Type: Cultivated from seed received by Bertoloni from the Royal Botanical Garden at Wirtenburg. *Bosch* s.n. (BOLO†, holo.).

Psoralea altissima Bertol., Horti bonon. pl. nov. 2: 16, t.II.1 (1839). Type: Cultivated from seed received by Bertoloni from the Royal Botanical Garden at Wirtenburg. *Bosch* s.n. (BOLO†, holo.).

Robust, shrub or tree to 5 m tall; forming dense colonies, reseeder. *Stems* erect single to few, branching in lower portion, spreading or arching, rigid, branches and twigs angular, sparsely hairy when immature, yellowish tan with storied white lenticels when young, becoming grey with age. *Seasonal shoots* pubescent pale green. *Leaves* (5)7–9-foliolate, imparipinnate, number variable at different stages of growth, crowded at the ends of bare branches on older shoots, or distributed along younger shoots, erect or patent, petiolate; leaflets 15–35×0.5–1 mm, villoso-pubescent, cylindrical, linear lanceolate, apex acute, base rounded, terminal leaflet shortest, basal pair longest, three uppermost pinnately inserted; petioles 3–7 mm long; rachis 10−15 mm long; stipules 3–4×1–2 mm, persistent, shorter than petiole, narrowly triangular, or triangular, fused for most of their length, overlapping like short stacked planks, becoming woody with age, margins incurled, subulate, slightly pubescent, rapidly senescent. *Inflorescences* axillary, borne in upper axils of seasonal shoots but with leafy extension, pseudo-spicate, or pseudo-capitate, congested, 1-flower per axil, but fasciculate, 1−10 (20) on short shoot, with or without distinct peduncle and pedicel; peduncles where present, stout and rigid, 2–3 mm long; cupulum trilobed, large, 3 mm long, sparsely white haired, margins ciliate with few black hairs, overlapping the base of the calyx, lobes subequally developed, carinal longer than other two, acuminate; pedicels where present 1–(2) mm long. *Flowers* 13−14 mm long, subtended by a 3 mm cucullate bract, clasping the oldest flower, pale mauve to pale blue, always hidden within the subtending leaflet, weakly scented or odourless. *Calyx* 6−7 mm long, tube 4–5×5 mm, glabrescent, mainly white-haired but also with black hairs on margins, or a mixture of black and white hairs, yellowish green; lobes sub-equally developed, shorter than the calyx tube, dark green, carinal longest, ovate-acute to triangular, vexillar lobes acute, straight, triangular, fused for less than quarter their length, inner face covered with bristle white hairs; ribs present, distinctly thickened with dense glands than any other part of the calyx; glands present, constant in size. *Standard petal* 12–14×10–11 mm, obovate, white suffused with pale mauve in the central area and with a single purple 3–4 mm long flash situated between and above the free swollen callosities at the top rim of the claw, auricles present, swollen, veins hyaline; claw 4–5 mm long, channelled. *Wing petals* 12–13×4 mm, longer than keel petals, white to pale mauve, blade folded and puckered along its mid-lines, sculpturing present, basal, comprised of 2–3 rows of 10–14 transcostal lamellae; claw 5 mm long. *Keel* 11×3.5–4.0 mm, blade ridged for 3 mm from base, white to pale mauve; claw 7 mm long. *Androecium* 11–12 mm long, tenth stamen shortly united with staminal sheath adaxially. *Pistil* 12 mm long, stipe 1 mm long, ovary 2 mm long, covered in club-shaped glands, style 6 mm long straight, then up curved, widest at point of flexure, height of curvature of style 2−3 mm, stigma penicillate. *Fruits* oblong, 4–5×2.5–3.0 mm. *Seeds* solitary, 2–4 mm, dark brown to black (Figure 40).

4.6.2.25.1. Diagnostic characters

Psoralea pinnata is similar to *P. affinis* and *P. ivumba*; all reseeders and colonial. The three species can however be separated as follows: *Psoralea pinnata* is a large single stemmed, multi-branched shrub to small tree up to 5 m tall, with (5)7–9-foliolate, villoso-pubescent congested leaves with three uppermost leaflets pinnately inserted; subulate recurved persistent stipules that become woody when leaves are shed; sessile or sub-sessile pale mauve or pale blue weakly scented or odourless flowers borne in pseudo-spicate or pseudo-capitate inflorescences and hidden within the subtending leaves; yellowish green, white (mostly) and black-haired calyces; *Psoralea affinis* by contrast is an erect single or multi-stemmed less-branched shrub or tree up to 3 m tall, with (5)7−9 (11)-foliolate, sparsely hairy spreading leaves with three uppermost leaflets digitately inserted; deltoid, spreading to recurved, twisted, callose stipules; dark blue to purple scented, pseudo-spicate flowers with peduncles and pedicels nearly as long as the leaflets; calyces nigro-hirsute externally with short, broad and blunt lobes; stalked; *Psoralea ivumba* on the other hand is an erect, virgate pseudo-racemose shrub up to 2.5 m tall, with 7−11-foliolate, filiform, spreading leaves; persistent stipules; compact, stalked, clustered, intensely sweet-scented bright blue flowers borne at the apex of short flowering shoots in pseudo-capitate inflorescences with very much broader standard, wing and keel petals; 6−15 mm long peduncles terminating with a papery cupulum touching the shaggy white-haired calyces.

4.6.2.25.2. Distribution and habitat

Psoralea pinnata is distributed across the Cape Peninsula across to the Kogelberg Mountains in the Western Cape Province of South Africa (Figure 39). It occurs in mountain fynbos, forest margins, river beds seepages and open veldt at the elevation of 230–1060 m. It has been found mainly in the Cape Flats Sandstone Fynbos, Hangklip Sand Fynbos, Peninsula Granite Fynbos, Cape Winelands Shale Fynbos, Elgin Shale Fynbos, Bokkeveld Sandstone Fynbos, Peninsula Sand stone Fynbos, Swartland Shale Renosterveld, and Cape Flats Dune Strandveld vegetation types (FFd 5, FFd 6, FFg 3, FFh 5, FFh 6, FFs 1, FFs 9, FRs 9, and Fs 6) (Rebelo *et al.* 2006). There are records of this species occurring on St. Helena Island (Jackson, 1903) but it is uncertain whether it is an adventive or naturally occurring. The species has naturalised in Australia and New Zealand (Stirton *et al.* 2015).

Figure 40. Vegetative and reproductive morphology of *P. pinnata* (A) flowering shoot; (B) 7-foliolate leaf; (C) bud; (D) sessile flower; (E) stalked flower; (F) cupulum; (G) calyx opened out with vexillary lobes to the right; (H) front view of standard petal; (I) wing petal; (J) keel; (K) androecium; (L) pistil. Voucher *A. Bello 40* (BOL). Scale bars: A, 1 cm; B–L, 1 mm. Artist: Abubakar Bello.

4.6.2.25.3. Etymology

The specific epithet *pinnata* is derived from the Latin adjective *pinnatus –a –um* (feathered, winged) and refers to the pinnate leaves of the species.

4.6.2.25.4. Conservation status

Psoralea pinnata has an abundant and widespread occurrence across its range and is well-estabished in protected areas. It does not qualify for any of the categories of threat based on the IUCN criteria. It is therefore assessed as least concern (LC).

4.6.2.25.5. Specimens studied

South Africa. WESTERN CAPE: **3318 (Cape Town)**: Wynberg Ranges (-AB), 26 Oct 1895, *Wolley Dod A.H. 22* (K); Flora Bay (-AB), 27 Nov 1981, *Stirton C.H.* 9965 (K); Dieprivier Station 17 Jan 1894, *Kuntze O. s.n.* (K); Table Mountain (-AB), Jul, *Ecklon C.F. 657* (K); edge of forest in upper part of Skeleton Gorge, Table Mountain (-CD), 10 Feb 1929, *Gillet J.B. 56* (NBG); Table Mountain (-CD), Dec 1852, *MacGillivray J. 511* (K); Newlands Forest (-CD), 1 Mar 2009, *Muasya A.M. & Stirton C.H. 4460* (BOL); Rhodes Estate (-CD), 30 Nov 2011, *Muasya A.M. & Stirton C.H. 8424* (BOL); Rhodes Memorial (-CD), *Muasya A.M. & Stirton C.H. 5336* (BOL); Stellenbosch (-DD), 22 Oct 1846, *Alexander R.C. s.n.* (K). **3418 (Simonstown)**: Rondeheuwel, Chapman's Peak Drive 19 Oct 1986, *Stirton C.H. 11204* (K); St. James Mountain, Simonstown (-AB), 13 Oct 1980, *Davies D. 23* (NBG); Steenberg, Cape Peninsula (-AB), 9 Nov 1944, *Compton R.H. 16431, 16432* (NBG); Constantiaberg, Cape Peninsula (-AB), 22 Nov 1953, *Heginbotham M.C. 235* a&b, (NBG); Kirstenbosch Slopes (-AB), *Martinez-Azorin M. 41* (NBG); Reservoir above Silvermine (-AB), 10 Dec 2007, *Muasya A.M. & Stirton C.H. 3403* (BOL); Silvermine Nature Reserve, Steenberg Plateau (-AB), 10 Dec 2007, *Muasya A.M. & Stirton C.H. 3407* (BOL); Groot Silvermyn (-AB), 19 Oct 1986, *Stirton C.H. 11211* (K); Chapman's Peak, near Simonstown (-AB), 19 October 1986, *Stirton C.H. 11213* (NBG); Helderview, Somerset West off road to Stellenbosch (-BB), 8 Feb 2007, *Muasya A.M. & Stirton C.H. 3402* (BOL); top of Sir Lowry's Pass (-BB), 28 Nov 1980, *Stirton C.H. 8361* (PRE); 18 Sep 1981, *9091* (PRE); Sir Lowry's Pass (-BB), 6 Dec 1977, *Venter S. 1280* (PRE); 25 Nov 1981, *Stirton C.H. 9946* (K); Bottom of Sir Lowry's Pass (-BB), 25 Nov 1981, *Stirton C.H. 9944* (K); along Louwsrivier, Blauusteenberg side (-BB), 28 Nov 1981, *Stirton C.H. 9994* (K); Between Gordon's Bay and Betty's Bay(–BB), 13 Sep 2009, *Muasya A.M. & Stirton C.H. 4528* (BOL); Kogelberg Forest Reserve, Above Palmiet Louwrivier junction, Simonstown (-BD), 17 Jun 1970, *Boucher C. 1290* (NBG); Kogelberg Forest Reserve, Above Oudebos (-BD), 20 Dec 1968, *Boucher C. 206* (NBG); Kogelberg Research site (-BD), 11 Nov 1975, *Durand B.J. 103* (PRE, NBG); Kogelberg (-BD), 30 Oct 1986, *Stirton 11302* (K); ± 30 mi. (48 km) north of timber houses in vlei area, Kogelberg Forest Station, Southern Hottentots Holland Mountains (-BD), 24 Mar 1992, *Kruger I. 445* (PRE, NBG); Oostelike punt van Bettysbaai teen berg (-BD), 8 Jan 1983, *Grobbelaar N. 2769* (PRE); Rhodes estate (-CD), 30 Nov 1980, *Stirton C.H. 8424* (PRE); Lower slopes leading up to skeleton Gorge Kirstenbosch (-CD), 27 Sep 1981, *Stirton C.H. 9757* (PRE); Skeleton Gorge, Kirstenbosch Botanic Gardens (-CD), 1 Dec 1986, *Germishuizen G. 4085* (PRE); Kirstenbosch Botanic Gardens, on the way to Compton Herbarium (-CD) 27 Sep 2013, *Bello A. 40* (BOL); Kenilworth Race course (–CD), *Bennett N. & Cowell C. 1328* (K). **3419 (Caledon)**: Caledon (-AB), *Thom G. 967* (K); Kleinmond (-AB), 28 Nov 1981, *Stirton C.H. 9997* (K); Bot River Vlei (-AC), 16 Aug 1982, *O'Callaghan M. 287* (NBG); Bot Rivier vlei (-AC), 16 Aug 1982, *O'Callaghan M.G. 263* (PRE); Diepgat (-AD), 25 Nov 1981, *Stirton C.H. 9948* (K); Stanford (-AD), 27 Nov 1981, *Stirton C.H. 9957* (K); Vöelklip Beach, Hermanus (-AD), 13 Jan 1986, *Stirton C.H. 11112* (K); Buys se huis, Vogelgat 12 Jan 1987, *Stirton C.H. 11088* (K). **Without precise locality**: St. Helena, *Burchell W.J. s.n.* (K); Cape Peninsula, Dec 1929, *Maude A.H. s.n.* (K); Table Mountain, 1897, *MacOwan 1836* (K); C.B.S., *Mund s.n.* (TCD); Smitskraal Vlei, 31 Jan 1896, *Wolley Dod 775* (K); Cape, Aug 1885, *Forster J.R. s.n.* (K).

CHAPTER FIVE: SYNTHESIS AND CONCLUSION

The main aim of this study was to infer phylogenetic relationships within Psoraleeae and test previous hypotheses, particularly by Egan & Crandall (2008a & b) and Dludlu *et al.* (2013), on generic relationships. The new generic level relationships in Psoraleeae proposed in this study are largely congruent with Egan & Crandall (2008a) and Dludlu *et al.* (2013) with minor exceptions. The majority of the genera were found to be monophyletic with the exception of *Otholobium*. The South American members of *Otholobium* were resolved as sister to the Mediterranean *Bituminaria.* A new classification is proposed to accommodate all the South American *Otholobium* and will be published separately. Similarly, the genus *Psoralea* has been found to be nested within the southern African *Otholobium.* This finding is similar to Dludlu *et al*. (2013). The genus *Psoralea* consists of some 75 species diagnosed by having a cupulum (fusion and intercalary growth of three to four successive bracts; Tucker & Stirton 1990), the absence of recurved mucronate-obovate to oblanceolate leaflets, and inflorescences with each flower subtended by two minute free opposed bracts (Stirton 1981). Due to its strong support as a clade revealed by the molecular data, as well as its multiple distinct morphological characters separating it from *Otholobium* and the rest of Psoraleeae, the classification of Stirton (1981), which recognises *Otholobium* and *Psoralea* as distinct genera, is maintained in this study pending a more resolved phylogenetic relationship (work in progress).

The phylogenetic position of the South African, European and Asian *Cullen* studied here, revealed for the first time that they form a basal grade sister to the Australian *Cullen*. Unlike the polyphyletic *Otholobium*, the results of this study showed that *Cullen* is monophyletic despite being the most disjunctly distributed genus in the tribe Psoraleeae. The results confirmed its phylogenetic position as sister to the genus *Orbexilum* as shown by Dludlu *et al.* (2013) or *Hoita* as indicated by Egan & Crandall (2008a). However, its sister relationship to *Bituminaria*, or South African *Otholobium* as shown by Egan & Crandall (2008a) and Grimes (1990) is not supported.

In terms of the evolution of characters, our ancestral character state reconstructions showed that the ancestors of Psoraleeae had a herbaceous growth habit, compound leaves, and indehiscent fruits with single seeds. The presence of a cupulum and a secondary dehiscence by transverse rupture of the pod are synapomorphies unique to *Psoralea* and *Pediomelum* respectively. Psoraleeae diversified as woody in the fynbos biome and more recently into the succulent, as well as an occasional move into afro-temperate Islands in Eastern Africa. In North America, early occupation in the succulent biome was herbaceous, but multiple diversifications into temperate biomes shifted Psoraleeae into a woody life form in South America. The habit switch from herbaceousness back to the ancestral woody habit in angiosperms is supported in this study.

Another objective of this study was to search for the potential ancestral range (ancestral area and biome), the dispersal routes, and the factors responsible for the current distribution of Psoraleeae. This was achieved using divergence time estimations. Dispersal was favoured to explain the current disjunct distribution pattern in Psoraleeae due to its younger age, supporting the Schrire *et al.* (2005b) hypothesis. The results showed that Psoraleeae originated in the Miocene, ca. 8.2 mya in the Mediterranean south biome of southern Africa. It then dispersed to the succulent biome of America ca. 6.7 mya. This was followed by a series of both local and transoceanic dispersal and speciation events. In summary, there were a total of 27 global dispersals based on area. The highest speciation event within a geographical area occurred in Africa with ca. 124 speciation events. This was followed by North America with ca. 44, Australia with 18, and one each in South America and Mediterranean regions. These results showed ca. 126 global dispersal events across biomes. The highest speciation event of 111 occurred within the Mediterranean south biome followed by ca. 28 in succulent, 15 in grassland, 11 in temperate forests and 9 in Mediterranean north biome. The results demonstrate the importance of long-distance dispersals (across three major oceans) in shaping the globally disjunct distribution in some recently diverged plants, such as Psoraleeae, by various agents including ocean currents or birds.

The third chapter focused on the application of large-scale DNA barcoding, which has recently gained a wide audience as a valuable technique for rapid species identification and for studying relationships within species and populations and also in revealing fundamental processes at generic and species levels. The quest for determining the usefulness of DNA barcoding in detecting phylogenetic signals (e.g., fire response trait - reseeding and resprouting) at species and generic levels, especially for speciose, recently diverged genera, was the main aim of this chapter.

In answering the above question, Bello *et al.* (2015a) applied DNA barcoding to two recently diverged legume genera of the tribe Psoraleeae (*Otholobium* and *Psoralea*), which are mostly confined to the Cape Floristic Region (CFR) of southern Africa, a fire climax flora with high levels of speciation and endemicity. We showed that psoraleoid legumes of the CFR exhibit a barcoding gap, with a combination of *matK* and *rbcLa* (*matK* + *rbcLa*) dataset as a better barcode than single regions. We also found that 100% of individuals of the two genera were correctly identified to their respective genera but that identifying them correctly to species was difficult (<50%). Our results also revealed a less considerable match (54%) between genetic species and morphologically-delimited species. For the detection of phylogenetic signals in fire response trait (reseeding and resprouting), the molecular phylogeny suggests a convergent origin of the reseeding trait in the African Psoraleoid genera. The different lineages showed a weak but significant phylogenetic conservatism in their response to fire as reseeders or resprouters, with more clustering of resprouters than would be expected by chance. This study showed that DNA barcoding may be useful in identification of SAf Psoraleoid genera (*Otholobium* and *Psoralea*) and in inferring the impacts of recurrent fires on gene flow in resprouting and reseeding taxa in the Cape Floristic Region.

The aim of Chapter 4 was to revise the taxonomy of the *Psoralea pinnata* complex. The objective was to determine whether the diversity that exists within the *P. pinnata* complex is a depiction of several local forms of a single variable species, as proposed by Harvey (1862), or whether it comprises distinct taxa that need to be recognised as separate species. This was achieved by sampling representative specimens of the complex, capturing their morphological and geographic diversity, and subjecting the data to various kinds of multivariate techniques (PCoA, CA). The methods proved to be very useful in species delimitation. Our results support the recognition of 25 species out of the ca. 28 supposed species. The results also rejected Harvey's (1862) taxonomic interpretation that *P. pinnata* is "one very variable species". We adopted a narrower circumscription of *P. pinnata* with a formal recognition of 24 additional taxa at specific rank. All the 25 species were fully described with illustrations, distribution maps and notes on their conservation status. We also provided a key for easier identification.

Although the majority of the members of the *Psoralea pinnata* complex were seen to have been forming some sub clades within *Psoralea* (Chapter 2), the phylogenetic position, the timing of speciation events leading to the species in the complex, and the divergence of the species could not be established in this study due to poor resolution within the southern African Psoraleeae. However, the results here have shown a change in some morphological traits within *Psoralea* e.g. a secondary loss of leaves at the adult stage among leaf species with leafy seedlings on the branches leading to the members of the *Psoralea aphylla* complex

(Figure 5b). It is anticipated that members of these complexes might form clades in a well resolved phylogeny. In terms of the fire response trait, the majority of the members of the *Psoralea pinnata* complex were found to be reseeders with few resprouter species e.g. *P. glabra*, *P. kougaensis, P. laevigata, P. margaretiflora*, *P. rhizotoma*, *P. suaveolens* and *P. vanberkelae,* that are mostly confined to drier habitats. Regarding the performance of the DNA barcoding in the identification of species, more importantly the newly described species in *Psoralea* and the *P. pinnata* complex, the results showed that the standard DNA barcodes markers failed to identify the species within the SAf Psoraleeae but that a successful identification of genera is observed.

In conclusion, this thesis contributes to science by generating new knowledge on the taxonomy, evolution and biogeography of a charismatic but poorly understood plant taxon Psoraleeae. The description of new species in South Africa and the reporting of a previously unrecorded invasive species in Australia add to our understanding of biodiversity and provide valuable information for conservation managers and policy makers.

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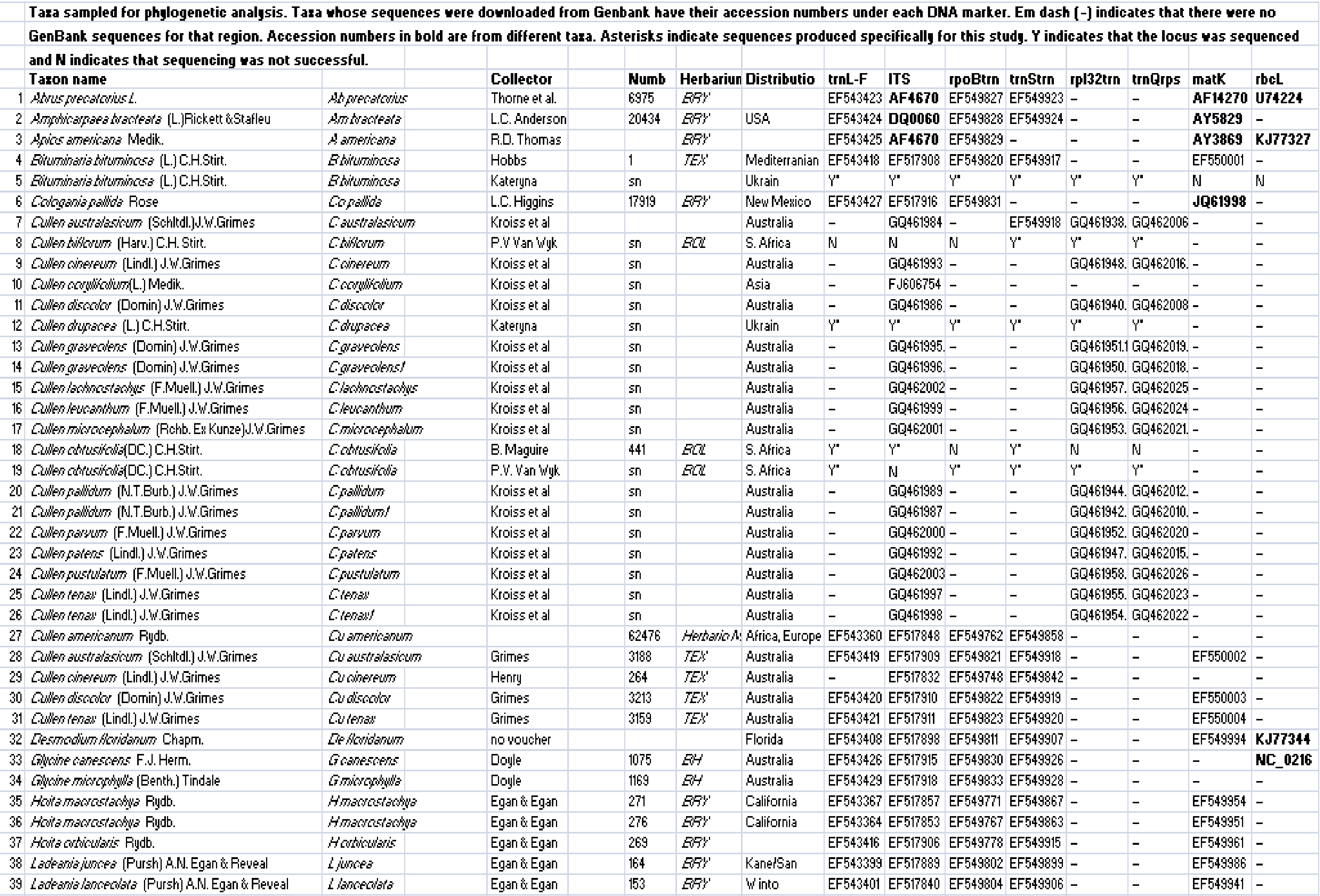
Appendix A



Taxa sampled for phylogenetic analysis. Taxa whose sequences were downloaded from Genbank have their accession numbers under each DNA marker. Em dash (-) indicates that there were no GenBank sequences for that region. Accession numbers in bold are from different taxa. Asterisks indicate sequences produced specifically for this study. Y indicates that the locus was sequenced and N indicates that sequencing was not successful.

	Taxon name		**Collector**	**Numb**	**Herbariun**	**Distributio**	**trnL-F**	**ITS**	**rpoBtrn**	**trnStrn**	**rpl32trn**	**trnQrps**	**matK**	**rbcL**
1	*Abrus precatorius L.*	*Ab precatorius*	Thorne et al.	6975	*BRY*		EF543423	**AF4670**	EF549827	EF549923	–	–	**AF14270**	**U74224**
2	*Amphicarpaea bracteata* (L.)Rickett &Stafleu	*Am bracteata*	L.C. Anderson	20434	*BRY*	USA	EF543424	**DQ0060**	EF549828	EF549924	–	–	**AY5829**	–
3	*Apios americana* Medik.	*A americana*	R.D. Thomas		*BRY*		EF543425	**AF4670**	EF549829	–	–	–	**AY3869**	**KJ77327**
4	*Bituminaria bituminosa* (L.) C.H.Stirt.	*B bituminosa*	Hobbs	1	*TEX*	Mediterranian	EF543418	EF517908	EF549820	EF549917	–	–	EF550001	–
5	*Bituminaria bituminosa* (L.) C.H.Stirt.	*B bituminosa*	Kateryna	sn		Ukrain	Y*	Y*	Y*	Y*	Y*	Y*	N	N
6	*Cologania pallida* Rose	*Co pallida*	L.C. Higgins	17919	*BRY*	New Mexico	EF543427	EF517916	EF549831	–	–	–	**JQ61998**	–
7	*Cullen australasicum* (Schltdl.)J.W.Grimes	*C australasicum*	Kroiss et al			Australia	–	GQ461984	–	EF549918	GQ461938.	GQ462006	–	–
8	*Cullen biflorum* (Harv.) C.H. Stirt.	*C biflorum*	P.V Van Wyk	sn	*BOL*	S. Africa	N	N	N	Y*	Y*	Y*	–	–
9	*Cullen cinereum* (Lindl.) J.W.Grimes	*C cinereum*	Kroiss et al	sn		Australia	–	GQ461993	–	–	GQ461948.	GQ462016.	–	–
10	*Cullen corylifolium*(L.) Medik.	*C corylifolium*	Kroiss et al	sn		Asia	–	FJ606754	–	–	–	–	–	–
11	*Cullen discolor* (Domin) J.W.Grimes	*C discolor*	Kroiss et al	sn		Australia	–	GQ461986	–	–	GQ461940.	GQ462008	–	–
12	*Cullen drupacea* (L.) C.H.Stirt.	*C drupacea*	Kateryna	sn		Ukrain	Y*	Y*	Y*	Y*	Y*	Y*	–	–
13	*Cullen graveolens* (Domin) J.W.Grimes	*C graveolens*	Kroiss et al	sn		Australia	–	GQ461995.	–	–	GQ461951.1	GQ462019.	–	–
14	*Cullen graveolens* (Domin) J.W.Grimes	*C graveolens1*	Kroiss et al	sn		Australia	–	GQ461996.	–	–	GQ461950.	GQ462018.	–	–
15	*Cullen lachnostachys* (F.Muell.) J.W.Grimes	*C lachnostachys*	Kroiss et al	sn		Australia	–	GQ462002	–	–	GQ461957.	GQ462025	–	–
16	*Cullen leucanthum* (F.Muell.) J.W.Grimes	*C leucanthum*	Kroiss et al	sn		Australia	–	GQ461999	–	–	GQ461956.	GQ462024	–	–
17	*Cullen microcephalum* (Rchb. Ex Kunze)J.W.Grimes	*C microcephalum*	Kroiss et al	sn		Australia	–	GQ462001	–	–	GQ461953.	GQ462021.	–	–
18	*Cullen obtusifolia*(DC.) C.H.Stirt.	*C obtusifolia*	B. Maguire	441	*BOL*	S. Africa	Y*	Y*	N	Y*	N	N	–	–
19	*Cullen obtusifolia*(DC.) C.H.Stirt.	*C obtusifolia*	P.V. Van Wyk	sn	*BOL*	S. Africa	Y*	N	Y*	Y*	Y*	Y*	–	–
20	*Cullen pallidum* (N.T.Burb.) J.W.Grimes	*C pallidum*	Kroiss et al	sn		Australia	–	GQ461989	–	–	GQ461944.	GQ462012.	–	–
21	*Cullen pallidum* (N.T.Burb.) J.W.Grimes	*C pallidum1*	Kroiss et al	sn		Australia	–	GQ461987	–	–	GQ461942.	GQ462010.	–	–
22	*Cullen parvum* (F.Muell.) J.W.Grimes	*C parvum*	Kroiss et al	sn		Australia	–	GQ462000	–	–	GQ461952.	GQ462020	–	–
23	*Cullen patens* (Lindl.) J.W.Grimes	*C patens*	Kroiss et al	sn		Australia	–	GQ461992	–	–	GQ461947.	GQ462015.	–	–
24	*Cullen pustulatum* (F.Muell.) J.W.Grimes	*C pustulatum*	Kroiss et al	sn		Australia	–	GQ462003	–	–	GQ461958.	GQ462026	–	–
25	*Cullen tenax* (Lindl.) J.W.Grimes	*C tenax*	Kroiss et al	sn		Australia	–	GQ461997	–	–	GQ461955.	GQ462023	–	–
26	*Cullen tenax* (Lindl.) J.W.Grimes	*C tenax1*	Kroiss et al	sn		Australia	–	GQ461998	–	–	GQ461954.	GQ462022	–	–
27	*Cullen americanum* Rydb.	*Cu americanum*		62476	*Herbario N*	Africa, Europe	EF543360	EF517848	EF549762	EF549858	–	–	–	–
28	*Cullen australasicum* (Schltdl.) J.W.Grimes	*Cu australasicum*	Grimes	3188	*TEX*	Australia	EF543419	EF517909	EF549821	EF549918	–	–	EF550002	–
29	*Cullen cinereum* (Lindl.) J.W.Grimes	*Cu cinereum*	Henry	264	*TEX*	Australia	–	EF517832	EF549748	EF549842	–	–	–	–
30	*Cullen discolor* (Domin) J.W.Grimes	*Cu discolor*	Grimes	3213	*TEX*	Australia	EF543420	EF517910	EF549822	EF549919	–	–	EF550003	–
31	*Cullen tenax* (Lindl.) J.W.Grimes	*Cu tenax*	Grimes	3159	*TEX*	Australia	EF543421	EF517911	EF549823	EF549920	–	–	EF550004	–
32	*Desmodium floridanum* Chapm.	*De floridanum*	no voucher			Florida	EF543408	EF517898	EF549811	EF549907	–	–	EF549994	**KJ77344**
33	*Glycine canescens* F.J. Herm.	*G canescens*	Doyle	1075	*BH*	Australia	EF543426	EF517915	EF549830	EF549926	–	–	–	**NC_0216**
34	*Glycine microphylla* (Benth.) Tindale	*G microphylla*	Doyle	1169	*BH*	Australia	EF543429	EF517918	EF549833	EF549928	–	–	–	–
35	*Hoita macrostachya* Rydb.	*H macrostachya*	Egan & Egan	271	*BRY*	California	EF543367	EF517857	EF549771	EF549867	–	–	EF549954	–
36	*Hoita macrostachya* Rydb.	*H macrostachya*	Egan & Egan	276	*BRY*	California	EF543364	EF517853	EF549767	EF549863	–	–	EF549951	–
37	*Hoita orbicularis* Rydb.	*H orbicularis*	Egan & Egan	269	*BRY*		EF543416	EF517906	EF549778	EF549915	–	–	EF549961	–
38	*Ladeania juncea* (Pursh) A.N. Egan & Reveal	*L juncea*	Egan & Egan	164	*BRY*	Kane/San	EF543399	EF517889	EF549802	EF549899	–	–	EF549986	–
39	*Ladeania lanceolata* (Pursh) A.N. Egan & Reveal	*L lanceolata*	Egan & Egan	153	*BRY*	W into	EF543401	EF517840	EF549804	EF549906	–	–	EF549941	–

Appendix A Cont.

40	*Orbexilum gracile* (Torr. & A.Gray) J.W.Grimes	*Or gracile*	Egan & Egan	240	*TEX*		EF543363	EF517852	EF549766	EF549862	–	–	EF549950	–
41	*Orbexilum lupinellum* (Michx.) Isely	*Or lupinellum*	Egan & Egan	257	*BRY*	SE USA	EF543365	EF517899	EF549812	EF549908	–	–	EF549952	–
42	*Orbexilum melanocarpum* (Benth.) Rydb.	*Or melanocarpum*	Grimes	2287	*TEX*	Mexico	EF543361	EF517849	EF549763	EF549859	–	–	EF549948	–
43	*Orbexilum onobrychis* (Nutt.) Rydb.	*Or onobrychis*	Raven & Raven	27603	*TEX*	N Midwest	EF543378	EF517850	EF549744	EF549878	–	–	EF549933	–
44	*Orbexilum pedunculatum* (Nutt.) Rydb.	*Or pedunculatum*	Egan	sn	*TEX*		EF543413	EF517903	EF549816	EF549912	–	–	EF549950	–
45	*Orbexilum simplex* (Nutt. ex Torr. & A.Gray) Rydb.	*Or simplex*	Thomas	65475	*TEX*	S Central USA	EF543379	EF517869	EF549782	EF549879	–	–	EF549966	–
46	*Orbexilum virgatum* (Nutt.) Rydb.	*Or virgatum*	Egan & Egan	251	*BRY*	NE Florida	–	EF517854	EF549768	–	–	–	–	–
47	*Orbexilum virgatum* (Nutt.) Rydb.	*Or virgatum*	Egan & Egan	253	*BRY*	NE Florida	EF543349	EF517834	EF549750	EF549844	–	–	EF549937	–
48	*Otholobium acuminatum* (Lam.) C.H.Stirt.	*O acuminatum*	Muasya, Stirton & Dludlu	3603	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	Y*	Y*
49	*Otholobium arborescens* C.H.Stirt.	*O arborescens*	Muasya, Stirton & Dludlu	3279	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176481*	KU176681*
50	*Otholobium argenteum* C.H.Stirt	*O argenteum*	Muasya & Stirton	4811	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
51	*Otholobium bolusii* (Forbes) C.H.Stirt.	*O bolusii*	Dludlu, Muasya & Stirton	3	*BOL*	S. Africa	Y	Y*	N	Y*	Y*	Y*	Y*	N
52	*Otholobium bowieanum* C.H.Stirt.	*O bowieanum*	Muasya & Stirton	5093	*BOL*	S. Africa	N	N	Y*	Y*	Y*	N	N	N
53	*Otholobium bracteolatum* (Eckl. & Zeyh.) C.H.Stirt.	*O bracteolatum*	Muasya, Stirton & Dludlu	3164	*BOL*	S. Africa	N	Y	Y	Y*	Y*	Y*	KU176503*	KU176703*
54	*Otholobium caffrum* C.H.Stirt	*O caffrum*	Muasya & Stirton	6140	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
55	*Otholobium caffrum* C.H.Stirt	*O caffrum*	Muasya & Stirton	6749	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
56	*Otholobium brachystachyum* J.W. Grimes	*O brachystachyum*	Grimes	2513	*TEX*	Ecuador	EF543422	EF517913	EF549825	EF549922.	–	–	–	–
57	*Otholobium candicans* (Eckl. & Zeyh.) C.H.Stirt.	*O candicans*	Muasya, Stirton & Dludlu	3369	*BOL*	S. Africa	N	Y	N	Y*	Y*	Y*	KU176583*	KU176784*
58	*Otholobium carneum* C.H.Stirt	*O carneum*	Esterhuysen	10817	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
59	*Otholobium curtisiae* C.H.Stirt. & Muasya	*O curtisiae*	Curtis	57	*BOL*	S. Africa	N	N	N	N	Y*	N	N	N
60	*Otholobium decumbens* (Aiton) C.H.Stirt.	*O decumbens*	Salter	7680	*BOL*	S. Africa	N	Y*	N	N	N	N	N	N
61	*Otholobium flexuosum* C.H.Stirt.	*O flexuosum*	Muasya, Stirton & Dludlu	3276	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	KU176617*	KU176818*
62	*Otholobium foliosum* (Oliv.) C.H.Stirt.	*O foliosum*	Gehrke & Muasya	AF086	*EA*	Kenya	Y	Y	Y	N	N	N	N	N
63	*Otholobium fruticans* (L.) C.H.Stirt.	*O fruticans*	Muasya, Stirton & Dludlu	3480	*BOL*	S. Africa	N	N	Y	N	N	N	KU176659*	KU176861*
64	*Otholobium hamatum* (Harv.) C.H.Stirt.	*O hamatum*	Muasya, Stirton & Dludlu	3310	*BOL*	S. Africa	N	Y	N	Y*	Y*	Y*	KU176571*	KU176772*
65	*Otholobium heterosepalum* (Fourc.) C.H.Stirt.	*O heterosepalum*	Stirton	13500	*BOL*	S. Africa	N	N	Y*	Y*	Y*	Y*	N	N
66	*Otholobium hirtum* (L.) C.H.Stirt.	*O hirtum*	Muasya, Stirton & Dludlu	3372	*BOL*	S. Africa	Y	Y	Y	N	Y*	Y*	KU176671*	KU176873*
67	*Otholobium incanum* C.H.Stirt.	*O incanum*	Stirton	10131	*BOL*	S. Africa	N	Y*	Y	Y	Y*	Y*	N	N
68	*Otholobium lanceolatum* C.H. Stirt.	*O lanceolatum*	Dludlu, Muasya & Stirton	13	*BOL*	S. Africa	N	Y	N	Y	Y*	Y*	N	N
69	*Otholobium macradenium* (Harv.) C.H.Stirt.	*O macradenium*	Muasya, Stirton & Dludlu	4452	*BOL*	S. Africa	Y	N	Y	N	N	N	N	N
70	*Otholobium mexicanum* (L.f.) J.W.Grimes	*O mexicanum*	Jorgenson	92018	*TEX*	NW S.	–	EF517914	EF549826	–	–	–	–	–
71	*Otholobium mundianum* (Eckl. & Zeyh.) C.H.Stirt.	*O mundianum*	Muasya, Stirton & Dludlu	3885	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176668*	KU176870*
72	*Otholobium obliquum* (E.Mey.) C.H.Stirt.	*O obliquum*	Muasya, Stirton & Dludlu	3198a	*BOL*	S. Africa	N	Y	Y	Y*	Y*	N	KU176673*	KU176875*
73	*Otholobium parviflorum* (E.Mey.) C.H.Stirt.	*O parviflorum*	Muasya, Stirton & Dludlu	3199	*BOL*	S. Africa	N	Y	N	Y*	Y*	Y*	KU176596*	KU176797*
74	*Otholobium pictum* C.H.Stirt.	*O pictum*	Esuston-Brown	1625	*BOL*	S. Africa	N	Y*	Y*	Y*	Y*	Y*	N	N
75	*Otholobium pilliferum* C.H.Stirt.	*O pilliferum*	Suin	1507	*BOL*	S. Africa	N	Y*	N	N	N	N	N	N
76	*Otholobium polystictum* (Benth. ex Harv.) C.H.Stirt.	*O polystictum*	Muasya, Stirton & Dludlu	3730	*BOL*	S. Africa	N	Y	N	N	N	N	N	N
77	*Otholobium pungens* C.H.Stirt.	*O pungens*	Stirton	13534	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
78	*Otholobium pustulatum* C.H.Stirt.	*O pustulatum*	Muasya, Stirton & Dludlu	3286	*BOL*	S. Africa	N	Y	Y	Y*	Y*	Y*	KU176651*	KU176851*
79	*Otholobium racemosum* (Thunb.) C.H.Stirt.	*O racemosum*	Pillans	3492	*BOL*	S. Africa	N	Y*	N	N	N	N	N	N
80	*Otholobium rotundifolium* (L.f.) C.H.Stirt.	*O rotundifolium*	Dludlu, Muasya & Stirton	4	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	N	KU176661*	KU176863*
81	*Otholobium rubicundum* C.H.Stirt.	*O rubicundum*	Muasya & Stirton	5982	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	KU176658*	KU176859*
82	*Otholobium sabulosum* C.H.Stirt.	*O sabulosum*	Bello & Stirton	141	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N

Appendix A Cont.

83	*Otholobium sericeum* (Poir.) C.H.Stirt.	*O sericeum*	Muasya, Stirton & Dludlu	3869	*BOL*	S. Africa	N	Y	Y	Y*	Y*	Y*	KU176598*	KU176799*
84	*Otholobium sp. nov. ined. "crewii"*	*O crewii*	Muasya & Stirton	3264	*BOL*	S. Africa	N	N	N	N	N	N	KU176618*	KU176819*
85	*Otholobium sp. nov. ined. "dreweae"*	*O dreweae*	Drewe	586	*BOL*	S. Africa	Y*	N	Y*	N	Y*	Y*	N	N
86	*Otholobium sp. nov. ined. "lucens"*	*O lucens*	Muasya, Stirton & Dludlu	3570	*BOL*	S. Africa	N	Y*	Y*	Y*	Y*	Y*	KU176568*	KU176769*
87	*Otholobium sp. nov. ined. "nitens"*	*O nitens*	Muasya, Stirton & Dludlu	3884	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	N	N
88	*Otholobium sp. nov. ined. "prodiens"*	*O prodiens*	Muasya, Stirton & Dludlu	3854	*BOL*	S. Africa	N	Y	N	Y*	Y*	Y*	KU176489*	KU176689*
89	*Otholobium sp. nov. ined. "saxosum"*	*O saxosum*	Muasya, Stirton & Dludlu	102	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	N	N
90	*Otholobium sp. nov. ined. "spissum"*	*O spissum*	Muasya, Stirton & Dludlu	4101	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	N	N
91	*Otholobium sp. nov. ined. "velutinum"*	*O velutinum*	Stirton	13106	*BOL*	S. Africa	N	N	N	N	N	N	KU176634*	KU176834*
92	*Otholobium spicatum* (L.) C.H.Stirt.	*O spicatum*	Muasya, Stirton & Dludlu	3568	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	N	KU176550*	KU176751*
93	*Otholobium stachydis* Thunb.	*O stachydis*	Muasya, Stirton & Dludlu	3264	*BOL*	S. Africa	N	Y	N	Y*	Y*	Y*	N	N
94	*Otholobium stachyerum* (Eckl. & Zeyh.) C.H.Stirt.	*O stachyerum*	Muasya, Stirton & Dludlu	3851	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	KU176515*	KU176715*
95	*Otholobium striatum* (Thunb.) C.H.Stirt.	*O striatum*	Muasya, Stirton & Dludlu	3339	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176620*	KU176821*
96	*Otholobium swartbergense* C.H.Stirt.	*O swartbergense*	Muasya, Stirton & Dludlu	3587	*BOL*	S. Africa	Y	Y*	Y	Y*	Y*	Y*	KU176631*	KU176831*
97	*Otholobium thomii* (Harv.) C.H.Stirt.	*O thomii*	Muasya, Stirton & Dludlu	3187	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176541*	KU176742*
98	*Otholobium trianthum* (E. Mey.) C.H. Stirt.	*O trianthum*	Esterhuysen	4029	*BOL*	S. Africa	N	Y	N	N	N	Y*	N	N
99	*Otholobium uncinatum* (Eckl. & Zeyh.) C.H.Stirt.	*O uncinatum*	Muasya, Stirton & Dludlu	3263	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176495*	KU176695
100	*Otholobium uncinatum* (Eckl. & Zeyh.) C.H.Stirt.	*O uncinatum*	Muasya, Stirton & Dludlu	3261	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176629*	KU176829*
101	*Otholobium uncinatum* (Harms) C.H.Stirt.	*O uncinatum*	Muasya, Stirton & Dludlu	3175	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176587*	KU176788*
102	*Otholobium venustum* (Eckl. & Zeyh.) C.H.Stirt.	*O venustum*	Muasya, Stirton & Dludlu	4327	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	N	Y*
103	*Otholobium virgatum* (Burm. F.) C.H. Stirt.	*O virgatum*	Muasya, Stirton & Dludlu	3163	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176488*	KU176688*
104	*Otholobium zeyheri* (Harv.) C.H.Stirt.	*O zeyheri*	Muasya, Stirton & Dludlu	3173	*BOL*	S. Africa	Y	N	N	Y*	Y*	Y*	**KU176661**	**KU176863**
105	*Pediomelum argophyllum* (Pursh) J.W.Grimes	*Pe argophyllum*	Mcneilus	972	*TEX*	Midwest USA	EF543402	EF517847	EF549805	EF549901.	–	–	EF549964	–
106	*Pediomelum aromaticum var. tuhyi* S.L.Welsh	*Pe arom. var. tuhyi*	Egan & Egan	157	*BRY*	Moab co.	EF543404	EF517894	EF549807	EF549903	–	–	EF549990	–
107	*Pediomelum aromaticum* (Payson) W.A.Weber var.	*Pe arom. var. barnebyi*	Egan & Egan	143	*BRY*	Kane co. Utah	EF543403	EF517893	EF549806	EF549902	–	–	EF549989	–
108	*Pediomelum aromaticum var. aromatucum* (Payson)	*Pe arom. var. aromaticum*	Egan & Egan	151	*BRY*	E Central Utah	–	EF517835	EF549788	EF549885	–	–	–	–
109	*Pediomelum californicum* (S.Watson) Rydb.	*Pe californicum*	Egan & Egan	119	*BRY*	California	EF543386	EF517876	EF549789	EF549886	–	–	EF549973	–
110	*Pediomelum canescens* (Michx.) Rydb.	*Pe canescens*	Egan & Egan	265	*BRY*	SE USA	EF543410	EF517900	EF549775	EF549909	–	–	EF549958	–
111	*Pediomelum castoreum* (S.Watson) Rydb.	*Pe castoreum*	Egan & Egan	125	*BRY*	Virgin river,	EF543387	EF517877	EF549790	EF549887	–	–	EF549974	–
112	*Pediomelum cuspidatum* (Pursh) Rydb.	*Pe cuspidatum*	Egan & Egan	193	*BRY*	Texas into	EF543384	EF517841	EF549756	EF549884	–	–	EF549942	–
113	*Pediomelum cyphocalyx* (A.Gray) Rydb.	*Pe cyphocalyx*	Egan & Egan	201	*BRY*	Central Texas	EF543381	EF517842	EF549784	EF549881.	–	–	EF549943	–
114	*Pediomelum digitatum* (Nutt. Ex Torr & A.Gray) J.W.	*Pe digitatum*	Egan & Egan	190	*BRY*	Texas into	EF543388	EF517878	EF549791	EF549888	–	–	EF549975	–
115	*Pediomelum esculentum* (Pursh) Rydb.	*Pe esculentum*	Egan & Egan	216	*BRY*	Midwest USA	EF543347	EF517831	EF549747	EF549889	–	–	EF549976	–
116	*Pediomelum humile* Rydb.	*Pe humile*	no voucher			Val Verde Co.,	EF543417	EF517907	EF549819	EF549916.	–	–	EF550000	–
117	*Pediomelum hypogaeum* (Nutt.) Rydb.	*Pe hypogaeum*	Egan & Egan	209	*BRY*	Texas into	EF543390	EF517880	EF549793	EF549890	–	–	EF549977	–
118	*Pediomelum hypogaeum* (Nutt.) Rydb. var.	*Pe hyp. var. scaposum*	Egan & Egan	185	*BRY*	Central Texas	EF543398	EF517838	EF549801	EF549898	–	–	EF549939	–
119	*Pediomelum hypogaeum* (Nutt.) Rydb. var.	*Pe hyp. var. subulatum*	Egan & Egan	190a	*BRY*	E Texas	EF543407	EF517897	EF549810	EF549905	–	–	EF549993	–
120	*Pediomelum latestipulatum* (Shinners) Mahler var.	*Pe late. var. apressum*	Egan & Egan	186	*BRY*	Edwards	EF543391	EF517881	EF549794	EF549891.	–	–	EF549978	–
121	*Pediomelum linearifolium* (Torr. & A. Gray) J.W.	*Pe linearifolium*	Egan & Egan	206	*BRY*	Texas into	EF543383	EF517873	EF549786	EF549883	–	–	EF549970	–
122	*Pediomelum megalanthum* var. *retrorsum* (Rydb.)	*Pe meg. var. retrorsum*	Egan & Egan	144	*BRY*	NW Arizona,	EF543380	EF517845	EF549783	EF549880	–	–	EF549946	–
123	*Pediomelum megalanthum* var. *megalanthum* (Wootc	*Pe meg. var. megalanthum*	Egan & Egan	158	*BRY*	Eastern Utah	EF543393	EF517883	EF549796	EF549893	–	–	EF549980	–
124	*Pediomelum mephiticum* (S.Watson) Rydb.	*Pe memphiticum*	Egan & Egan	126	*BRY*	Washington	EF543394	EF517846	EF549760	EF549894	–	–	EF549947	–
125	*Pediomelum palmeri (Ock.) Grimes*	*Pe palmeri*	Grimes	2636	*TEX*	Mexico	EF543346	EF517830	EF549746	EF549840	–	–	EF549934	–

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126	*Pediomelum pariense* (S.L.Welsh & N.D. Atwood) J.W.(	*Pe pariense*	Egan & Egan	148	*BRY*	Kane co. Utah	EF543405	EF517885	EF549808	EF549895	–	–	EF549991	–
127	*Pediomelum pentaphyllum* (L.) Rydb.	*Pe pentaphyllum*	Egan & Egan	172	*BRY*	Cochise co.,	EF543406	EF517886	EF549809	EF549896	–	–	EF549983	–
128	*Pediomelum piedmontanum* J.R.Allison, M.W.Morris &	*Pe piedmontanum*	Egan & Egan	263	*BRY*	Fall Line of	EF543411	EF517862	EF549814	EF549910.	–	–	EF549959	–
129	*Pediomelum reverchonii* (S.Watson) Rydb.	*Pe reverchonii*	Orzell	55552	*BRY*	N Central	EF543397	EF517829	EF549800	EF549897	–	–	–	–
130	*Pediomelum rhombifolium* (Torr. & A.Gray) Rydb.	*Pe rhombifolium*	Egan & Egan	179	*BRY*	Texas,	EF543382	EF517836	EF549752	EF549846	–	–	EF549938	–
131	*Pediomelum subacaule* (Torr. & A.Gray) Rydb.	*Pe subacaule*	Egan & Egan	218	*BRY*	Cedar Glades	EF543412	EF517859	EF549773	EF549911.	–	–	EF549957	–
132	*Pediomelum tenuiflorum* (Pursh) A.N. Egan	*Pe tenuiflorum*	Egan & Egan	194	*BRY*	Midwest into	EF543400	EF517839	EF549803	EF549900	–	–	EF549987	–
133	*Phaseolus vulgaris* L.	*Ph vulgaris*	Baird	2461	*BRY*	Worldwide	EF543430	EF543518	EF549834	EF549929	–	–	**AY5829**	–
134	*Psoralea abbottii* C.H. Stirt.	*P abbottii*	Van Wyk A.E	7482	*BOL*	S. Africa	N	N	N	N	Y*	N	Y*	Y*
135	*Psoralea aculeata* L.	*P aculeata*	Muasya, Stirton & Dludlu	3170	*BOL*	S. Africa	Y	N	N	Y*	Y*	Y*	KU176552*	KU176753*
136	*Psoralea affinis* Eckl. & Zeyh.	*P affinis*	Muasya, Stirton & Dludlu	3903	*BOL*	S. Africa	Y	N	Y	N	N	N	KM896929	KM894258*
137	*Psoralea angustifolia* Jacq.	*P angustifolia*	Muasya, Stirton & Dludlu	3278	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	N	N
138	*Psoralea alata* (Thunb.) T.M.Salter	*P alata*	Muasya, Stirton & Dludlu	3880	*BOL*	S. Africa	N	N	N	Y*	Y*	Y*	KU176566*	KU176768*
139	*Psoralea aphylla* L.	*P aphylla*	Muasya, Stirton & Dludlu	3492	*BOL*	S. Africa	Y*	Y*	Y*	Y*	N	Y*	Y*	KU176625*
140	*Psoralea arborea* Sims.	*P arborea*	Dludlu, Muasya & Stirton	64	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	**KU17654**	**KU176746**
141	*Psoralia asarina* (P.J.Bergius) T.M.Salter	*P asarina*	Bello	29	*BOL*	S. Africa	N	N	Y*	Y*	Y*	Y*	**KU17664**	**KU176842**
142	*Psoralea axillaris* Thunb.	*P axillaris*	Muasya & Stirton	5874	*BOL*	S. Africa	N	N	N	N	N	N	Y*	Y*
143	*Psoralea congesta* C.H.Stirt.	*P congesta*	Muasya	5462	*BOL*	S. Africa	Y*	N	N	Y*	Y*	Y*	KU176574*	KU176775*
144	*Psoralea diturnerae* A.Bello, C.H.Stirt & Muasya	*P diturnerae*	Bello, Sirton & Muasya	41	*BOL*	S. Africa	Y*	N	Y*	Y*	Y*	Y*	N	N
145	*Psoralea ensifolia* (Houtt.) Merr.	*P ensifolia*	Bello, Sirton & Muasya	42	*BOL*	S. Africa	N	Y*	N	Y*	N	N	N	N
146	*Psoralea ensifolia* (Houtt.) Merr.	*P ensifolia*	Muasya	7155	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
147	*Psoralea filifolia* Eckl. & Zeyh.	*P filifolia*	Muasya, Stirton & Dludlu	4321	*BOL*	S. Africa	Y	N	Y	N	N	N	Y*	Y*
148	*Psoralea fleta* C.H.Stirt.	*P fleta*	Muasya, Stirton & Dludlu	3385	*BOL*	S. Africa	Y	Y	N	N	N	N	KU176564*	KU176766*
149	*Psoralea gigantea* Dludlu, C.H.Stirt. & Muasya	*P gigantea*	Muasya, Stirton & Dludlu	3201b	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176549*	KU176750*
150	*Psoralea glabra* E.Mey.	*P glabra*	Muasya, Stirton & Dludlu	3646	*BOL*	S. Africa	Y	Y	Y	N	N	N	Y*	Y*
151	*Psoralea glaucescens* Eckl. & Zeyh.	*P glaucescens*	Muasya, Stirton & Dludlu	3289	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176509*	KU176709*
152	*Psoralea imbricata* (L.f) T.M. Salter	*P imbricata*	Muasya, Stirton & Dludlu	4030	*BOL*	S. Africa	Y	Y*	Y	Y*	Y*	Y*	KU176533*	KU176733*
153	*Psoralea keetii* Schonl. ex H. M. Forbes	*P keetii*	Muasya & Stirton	3599	*BOL*	S. Africa	N	N	N	N	Y*	Y*	KU176532*	KU176732*
154	*Psoralea karooensis* C.H. Stirt. & Muasya	*P karooensis*	Bello, Sirton & Muasya	161	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
155	*Psoralea latifolia* (Harv.) C.H. Stirton ined.	*P latifolia*	Muasya & Stirton in Abbc	8841.5	*BOL*	S. Africa	Y	Y	N	Y*	Y*	Y*	N	N
156	*Psoralea laevigata* Eckl. & Zeyh.	*P laevigata*	Muasya & Stirton	3457	*BOL*	S. Africa	N	N	N	N	Y*	N	KU176652*	KU176852*
157	*Psoralea laxa* Salter	*P laxa*	Muasya, Stirton & Dludlu	3611	*BOL*	S. Africa	N	Y	N	Y*	Y*	Y*	KU176577*	KU176778*
158	*Psoralea margaretiflora* C.H. Stirt. & V.R. Clarke	*P margaretiflora*	Clark V.R	91	*BOL*	S. Africa	Y*	N	N	Y*	Y*	Y*	Y*	Y*
159	*Psoralea monophylla* (L) C.H.Stirt.	*P monophylla*	Muasya, Stirton & Dludlu	3476	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176483*	KU176683*
160	*Psoralea odoratissima* Jacq.	*P odoratissima*	Muasya, Stirton & Dludlu	3557	*BOL*	S. Africa	N	Y*	Y*	Y*	N	Y*	KU176501*	KU176701*
161	*Psoralea oligophylla* Eckl. & Zeyh.	*P oligophylla*	Muasya, Stirton & Dludlu	3798	*BOL*	S. Africa	N	Y	N	N	N	N	KU176522*	KU176722*
162	*Psoralea oreophila* Schltr.	*P oreophila*	Muasya, Stirton & Dludlu	3464	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	Y*	KU176529*	KU176729*
163	*Psoralea peratica* C.H. Stirt.	*P peratica*	Dludlu, Muasya & Stirton	80	*BOL*	S. Africa	N	Y	Y	N	N	N	N	N
164	*Psoralea pinnata* L.	*P pinnata*	Muasya, Stirton & Dludlu	3165	*BOL*	S. Africa	Y*	Y*	N	Y*	Y*	Y*	KU176580*	KU176781*
165	*Psoralea plauta* C.H.Stirt.	*P plauta*	Muasya, Stirton & Dludlu	4506	*BOL*	S. Africa	N	N	N	N	N	N	Y*	KU176728*
166	*Psoralea ramulosa* C.H.Stirt.	*P ramulosa*	Muasya & Stirton	4362	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
167	*Psoralea repens* P.J.Bergius	*P repens!*	Muasya, Stirton & Dludlu	3168	*BOL*	S. Africa	N	Y	Y	Y*	Y*	Y*	KU176484*	KU176684*
168	*Psoralea restioides* Eckl. & Zeyh.	*P restioides*	Muasya, Stirton & Dludlu	3216	*BOL*	S. Africa	N	Y	Y	N	N	N	KU176643*	KU176843*

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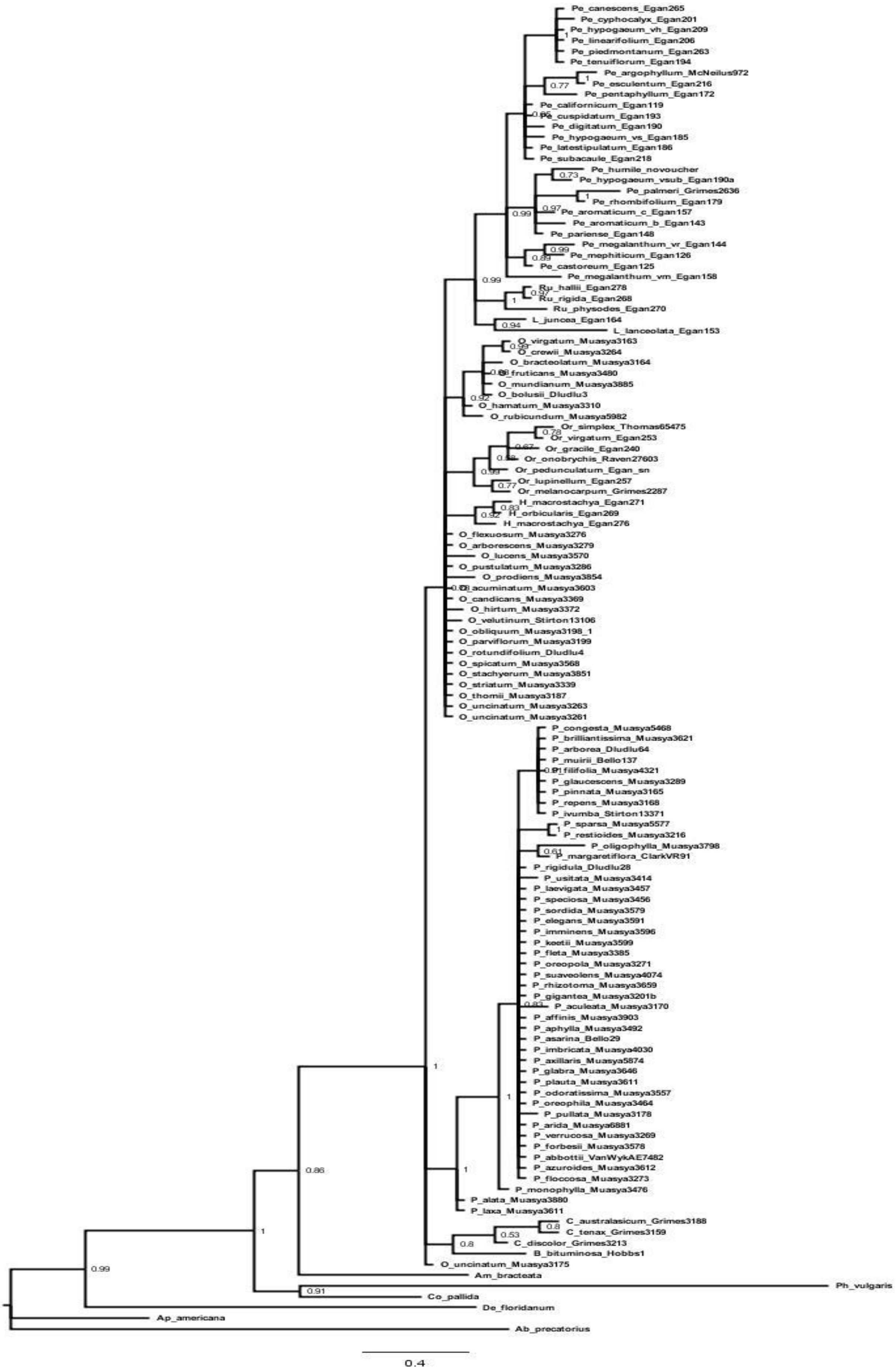
169	*Psoralea vanberkelae* C.H.Stirt., A. Bello & Muasya	*P vanberkelae*	NUB	1120	*BOL*	S. Africa	Y*	N	N	Y*	Y*	Y*	N	N
170	*Psoralea sp. nov. ined.* "*arida*"	*P arida*	Muasya	6881	*BOL*	S. Africa	Y*	N	N	Y*	Y*	Y*	N	N
171	*Psoralea sp. nov. ined.* "*azuroides*"	*P azuroides*	Muasya, Stirton & Dludlu	3612	*BOL*	S. Africa	N	Y	N	N	N	N	N	N
172	*Psoralea sp. nov. ined.* "*brilliantissima*"	*P brilliantissima*	Muasya, Stirton & Dludlu	3621	*BOL*	S. Africa	Y*	Y	Y*	Y*	Y*	Y*	KU176619*	KU176820*
173	*Psoralea sp. nov. ined.* "*elegans*"	*P elegans*	Muasya, Stirton	3591	*BOL*	S. Africa	Y	Y*	N	Y*	Y*	Y*	KU176536*	KU176736*
174	*Psoralea sp. nov. ined.* "*floccosa*"	*P floccosa*	Muasya, Stirton & Dludlu	3273	*BOL*	S. Africa	N	Y	N	N	N	N	KU176591*	KU176792*
175	*Psoralea sp. nov. ined.* "*forbesii*"	*P forbesii*	Muasya, Stirton	5965.2	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
176	*Psoralea sp. nov. ined.* "*forbesii*"	*P forbesii*	Muasya, Stirton	3578	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	KU176606*	KU176807*
177	*Psoralea sp. nov. ined.* "*fulva*"	*P fulva*	Bello, Sirton & Muasya	168	*BOL*	S. Africa	Y*	N	Y*	Y*	Y*	Y*	N	N
178	*Psoralea sp. nov. ined.* "*glausina*"	*P glausina*	Muasya	7192	*BOL*	S. Africa	Y*	N	Y*	Y*	Y*	Y*	N	N
179	*Psoralea sp. nov. ined.* "*imminens*"	*P imminens*	Muasya, Stirton & Dludlu	3596	*BOL*	S. Africa	Y*	Y*	Y	Y*	Y*	Y*	KU176667*	KU176869*
180	*Psoralea sp. nov. ined.* "*intonsa*"	*P intonsa*	Bello	112	*BOL*	S. Africa	Y*	Y*	N	Y*	Y*	N	N	Y*
181	*Psoralea sp. nov. ined.* "*ivumba*"	*P ivumba*	Stirton	13371	*BOL*	S. Africa	Y*	N	N	Y*	Y*	Y*	Y*	Y*
182	*Psoralea sp. nov. ined.* "*montana*"	*P montana*	Muasya	6648	*BOL*	S. Africa	Y*	N	N	Y*	Y*	Y*	N	Y*
183	*Psoralea sp. nov. ined.* "*muirii*"	*P muirii*	Bello & Stirton	137	*BOL*	S. Africa	Y*	Y*	Y	Y*	Y*	Y*	**KU17663**	**KU176838**
184	*Psoralea sp. nov. ined.* "*nubicola*"	*P nubicola*	Muasya & Stirton	3474	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	N	N
185	*Psoralea sp. nov. ined.* "*oreopola*"	*P oreopola*	Muasya & Stirton	3271	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	KU176507*	KU176707*
186	*Psoralea sp. nov. ined.* "*pullata*"	*P pullata*	Muasya, Stirton & Dludlu	3178	*BOL*	S. Africa	Y	Y	Y	Y*	Y*	N	KU176657*	KU176858*
187	*Psoralea sp. nov. ined.* "*rhizotoma*"	*P rhizotoma*	Muasya, Stirton & Dludlu	3659	*BOL*	S. Africa	Y*	Y*	Y	Y*	Y*	Y*	KU176664*	KU176866*
188	*Psoralea sp. nov. ined.* "*rigidula*"	*P rigidula*	Dludlu, Muasya & Stirton	28	*BOL*	S. Africa	Y	Y	Y	N	N	N	KU176648*	KU176848*
189	*Psoralea sp. nov. ined.* "*semota*"	*P semota*	Esterhuysen E.	21045	*BOL*	S. Africa	N	N	N	N	Y*	N	N	Y*
190	*Psoralea sp. nov. ined.* "*sordida*" x *Psoralea sp. nov.*	*P sordida*	Muasya, Stirton & Dludlu	3579	*BOL*	S. Africa	Y	Y	N	N	Y*	Y*	KU176666*	KU176868*
191	*Psoralea sp. nov. ined.* "*sparsa*"	*P sparsa*	Muasya & Stirton	5577	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	**KU176836**
192	*Psoralea sp. nov. ined.* "*tenuifolia*"	*P tenuifolia*	Muasya & Stirton	5710	*BOL*	S. Africa	Y*	Y*	Y*	Y*	Y*	Y*	N	N
193	*Psoralea sp. nov. ined.* "*triflora*"	*P triflora*	Bello, Sirton & Muasya	58	*BOL*	S. Africa	Y*	N	Y*	Y*	Y*	Y*	N	N
194	*Psoralea sp. nov. ined.* "*trullata*"	*P trullata*	Muasya & Stirton	3608	*BOL*	S. Africa	N	Y*	N	N	Y*	N	N	N
195	*Psoralea sp. nov. ined.* "*usitata*"	*P usitata*	Muasya, Stirton & Dludlu	3414	*BOL*	S. Africa	Y	Y	Y	N	N	N	KU176516*	KU176716*
196	*Psoralea sp. nov. ined.* "*vigilans*"	*P vigilans!*	Muasya, Stirton & Dludlu	4347	*BOL*	S. Africa	Y	N	N	Y*	Y*	Y*	N	N
197	*Psoralea speciosa Eckl. & Zeyh.*	*P speciosa*	Muasya, Stirton & Dludlu	3456	*BOL*	S. Africa	N	Y	N	N	N	Y*	KU176639*	KU176839*
198	*Psoralea suaveolens Jacq.*	*P suaveolens*	Muasya, Stirton & Dludlu	4074	*BOL*	S. Africa	N	Y	Y	N	N	N	KU176655*	KU176856*
199	*Psoralea verrucosa* Willd. ex Spreng.	*P verrucosa*	Muasya, Stirton & Dludlu	3269	*BOL*	S. Africa	Y	N	Y	Y*	Y*	Y*	KU176608*	KU176809*
200	*Rupertia hallii* (Rydb.) J.W.Grimes	*Ru hallii*	Egan & Egan	278	*BRY*	N California	EF543366	EF517833	EF549770	EF549843	–	–	EF549936	–
201	*Rupertia physodes* (Douglas ex Hook.) J.W.Grimes	*Ru physodes*	Egan & Egan	270	*BRY*	West Coast	EF543414	EF517858	EF549772	EF549913.	–	–	EF549960	–
202	*Rupertia rigida* (Parish) J.W.Grimes	*Ru rigida*	Egan & Egan	268	*BRY*	Baja	EF543415	EF517905	EF549818	EF549914.1	–	–	EF549999	–

Appendix B1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Pe_subacaule_Egan218
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0.99
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Pe_megalanthum_vr_Egan144
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Pe_castoreum_Egan125
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0.67
Or_gracile_Egan240
Or_onobrychis_Raven27603
Or_pedunculatum_Egan_sn
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Or_melanocarpum_Grimes2287
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Ab_precatorius
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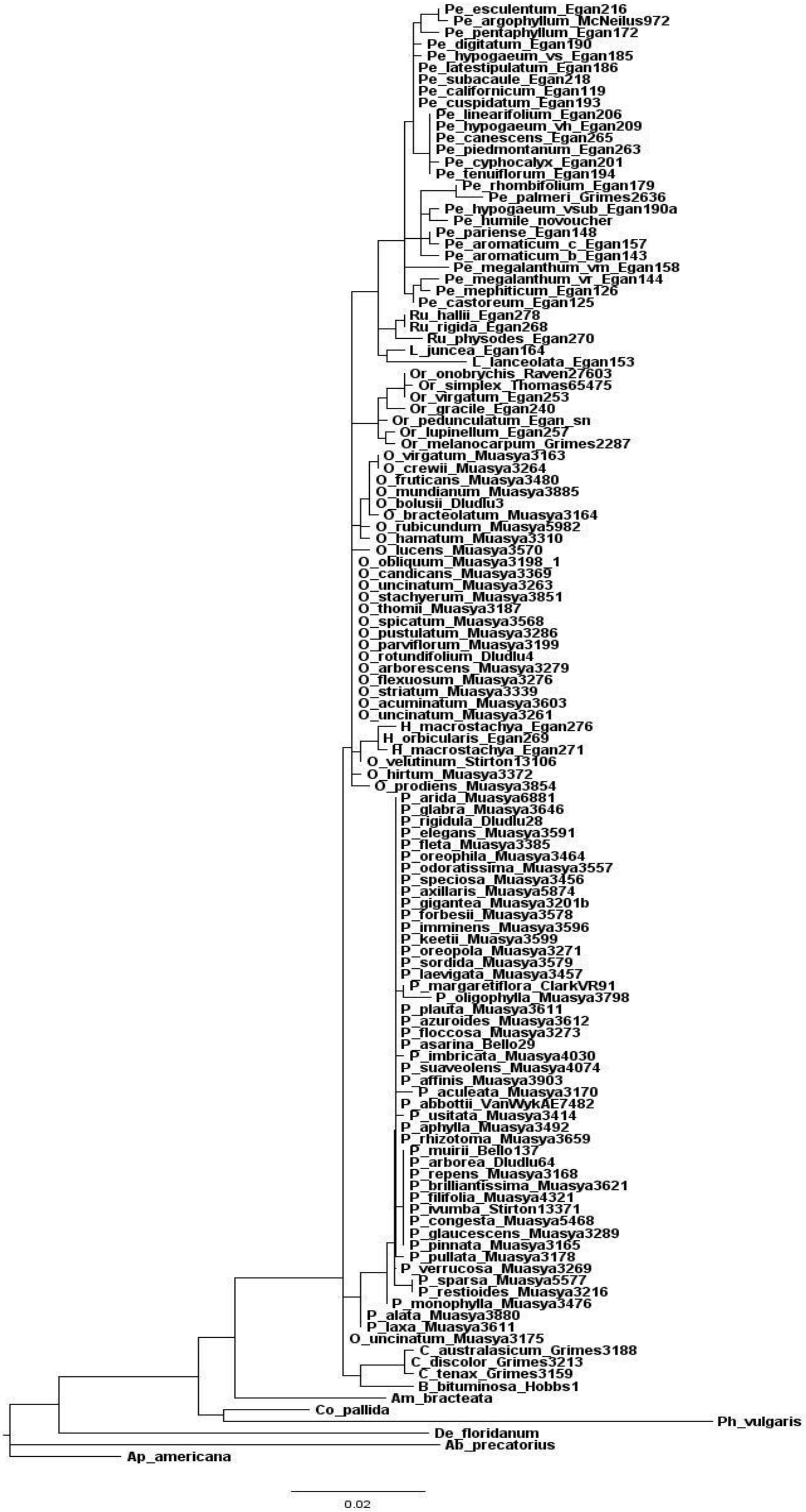
Appendix B2



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Ru_physodes_Egan270
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Or_simplex_Thomas65475
Or_virgatum_Egan253
Or_gracile_Egan240
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Or_lupinellum_Egan257
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C_discolor_Grimes3213
C_tenax_Grimes3159
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De_floridanum
Ab_precatorius
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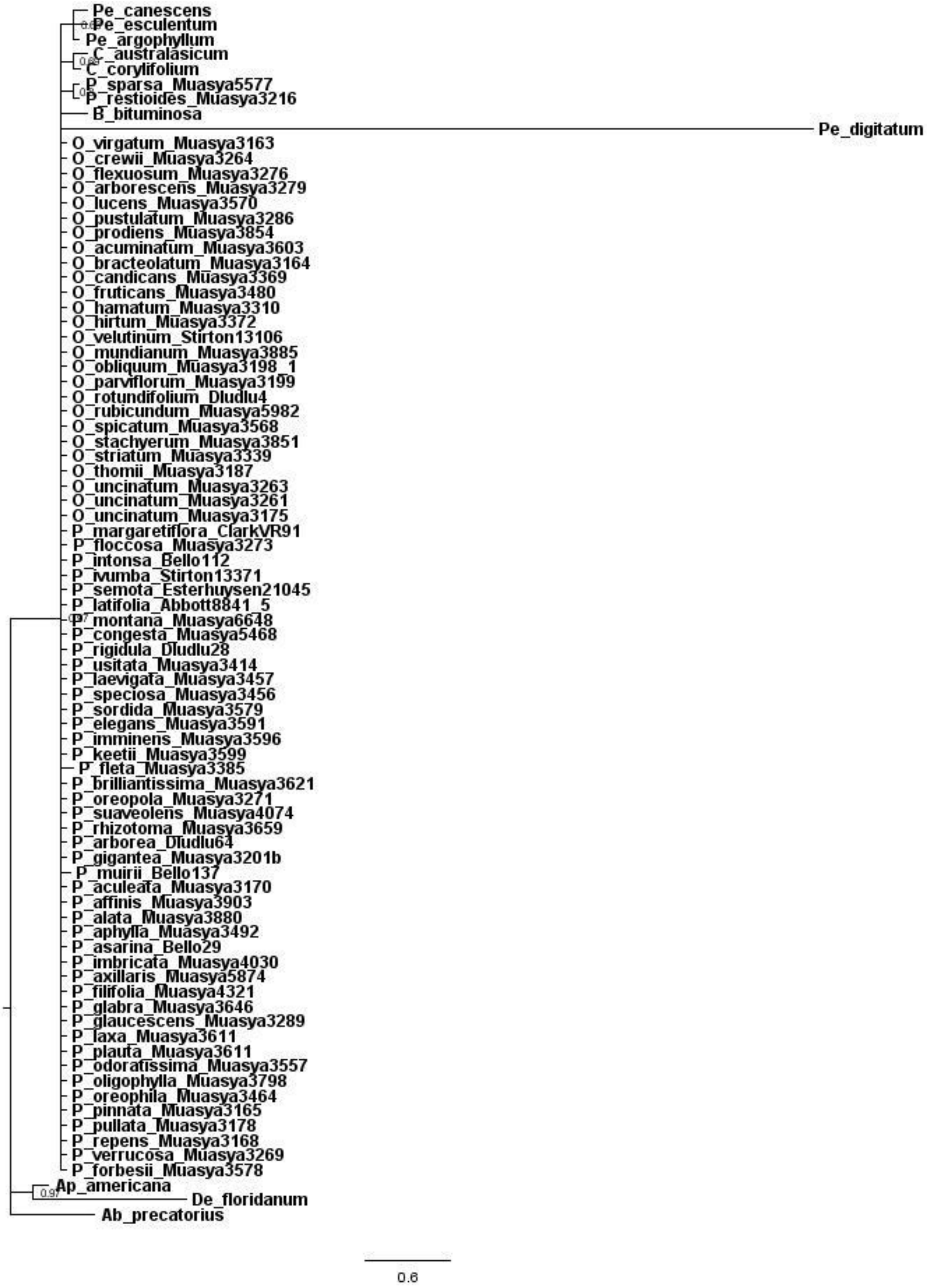
Appendix B3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Ab_precatorius
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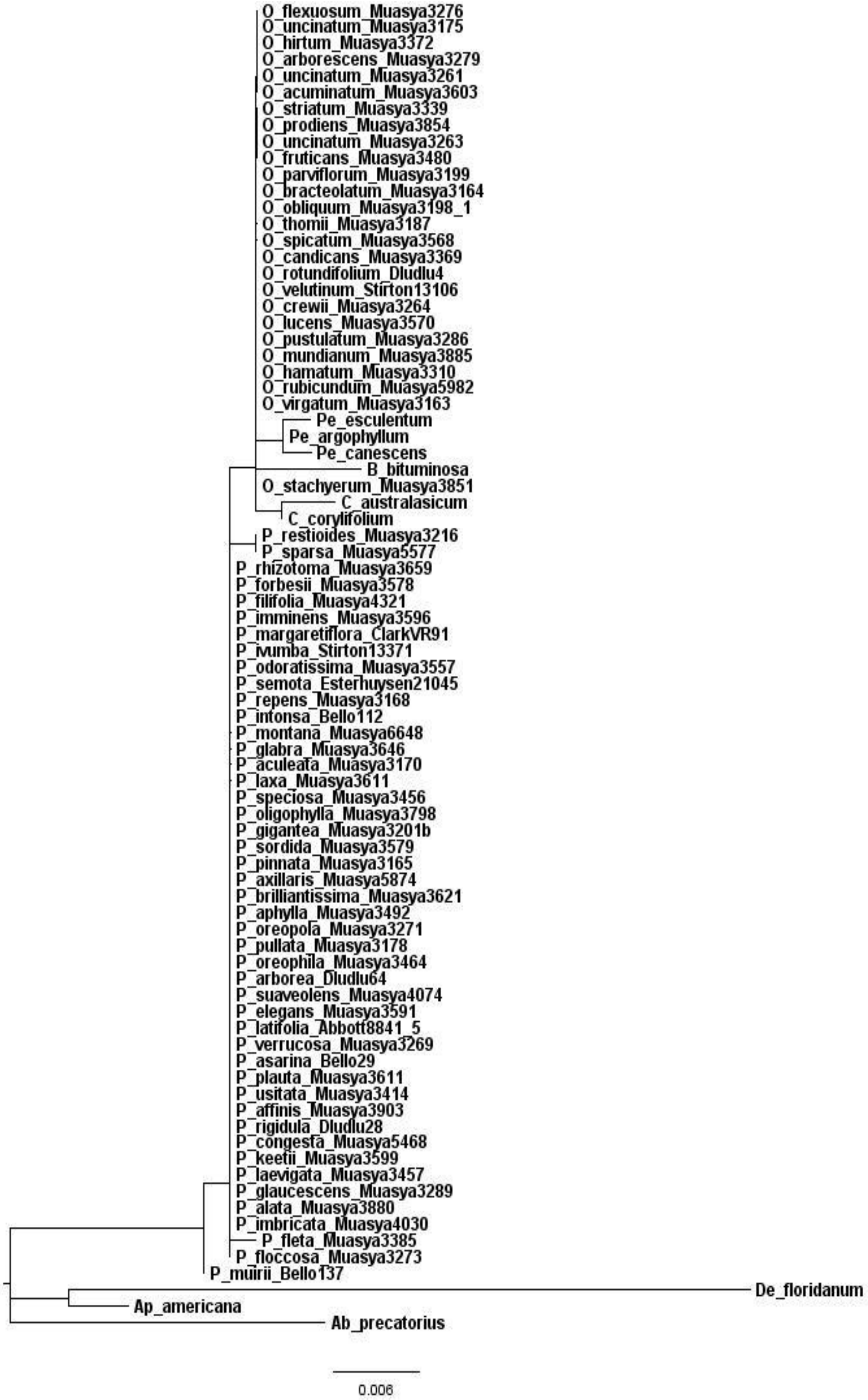
Appendix B4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Ab_precatorius
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Appendix B5


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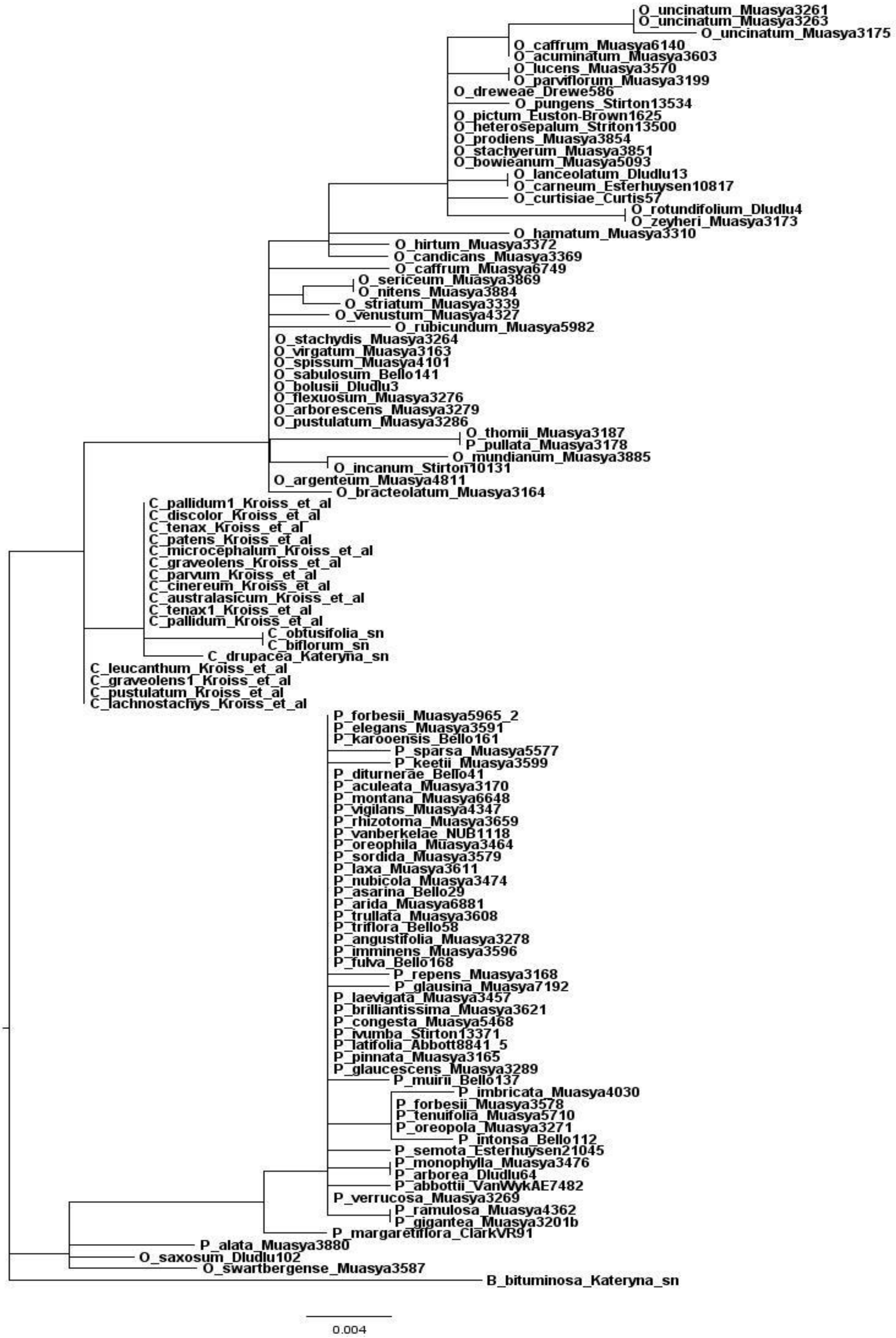
Appendix B6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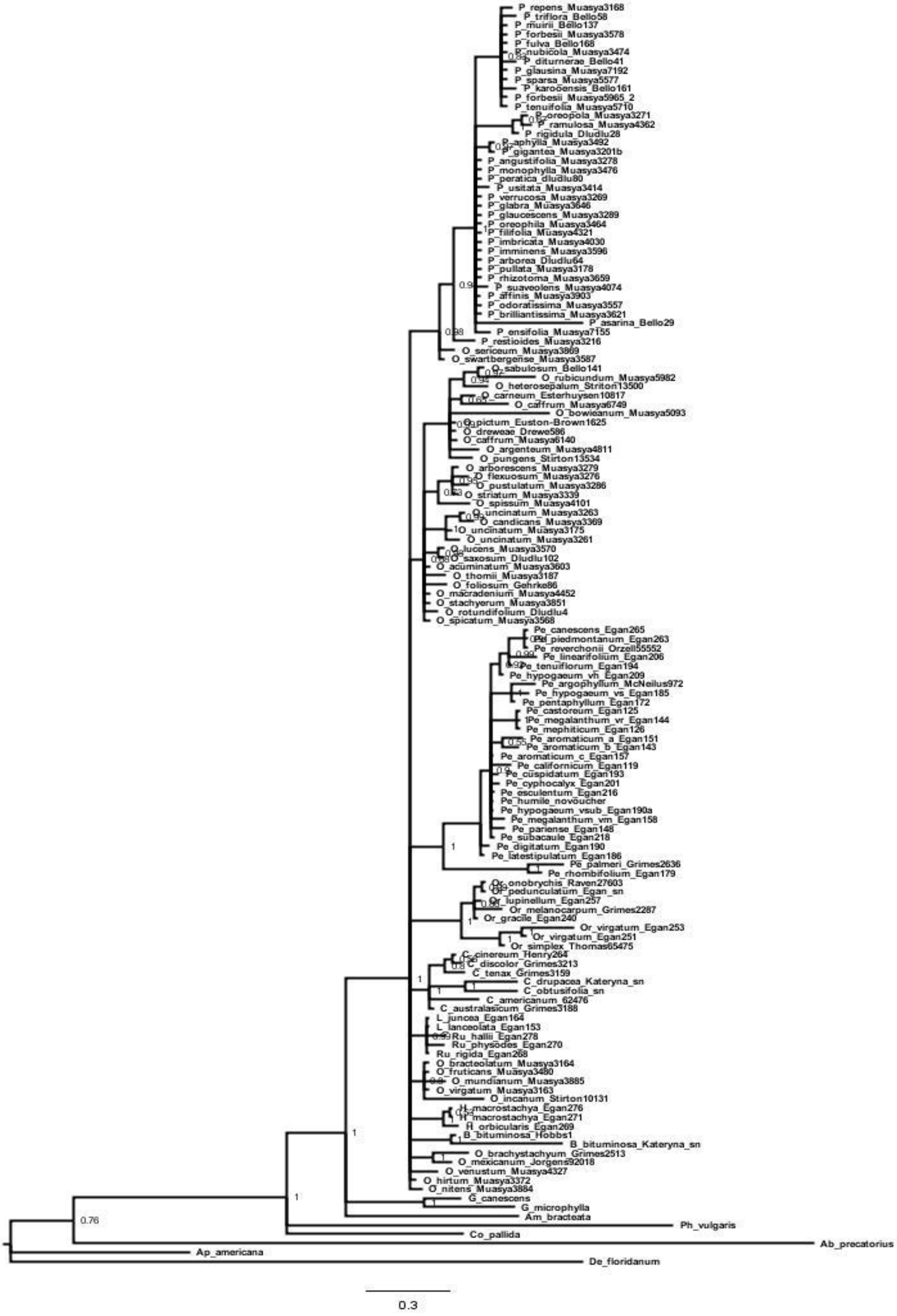
Appendix B7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Ru_physodes_Egan270
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O_hirtum_Muasya3372
O_nitens_Muasya3884
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G_microphylla
Am_bracteata
Ph_vulgaris
Co_pallida
Ab_precatorius
Ap_americana
De_floridanum
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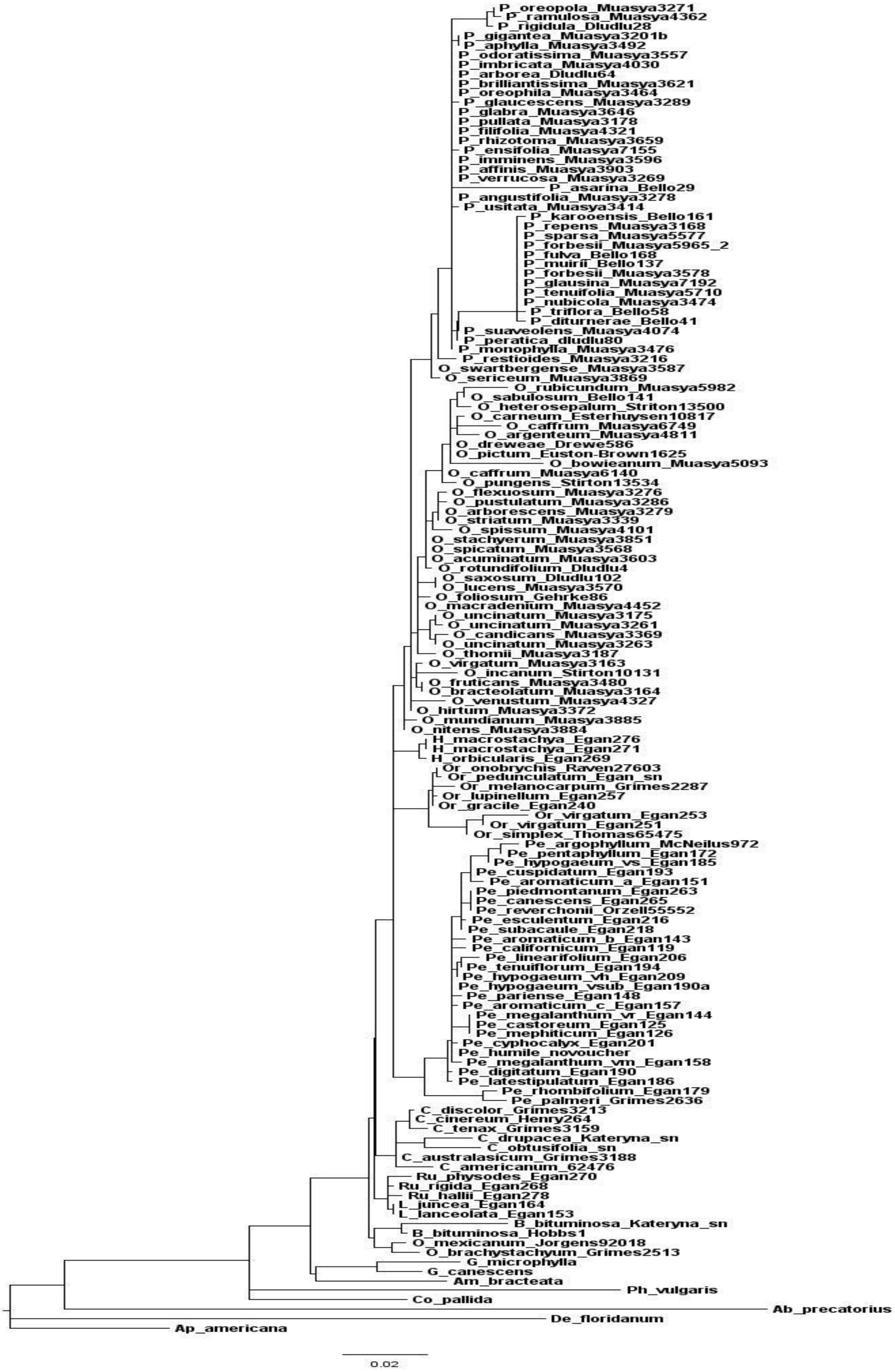
Appendix B8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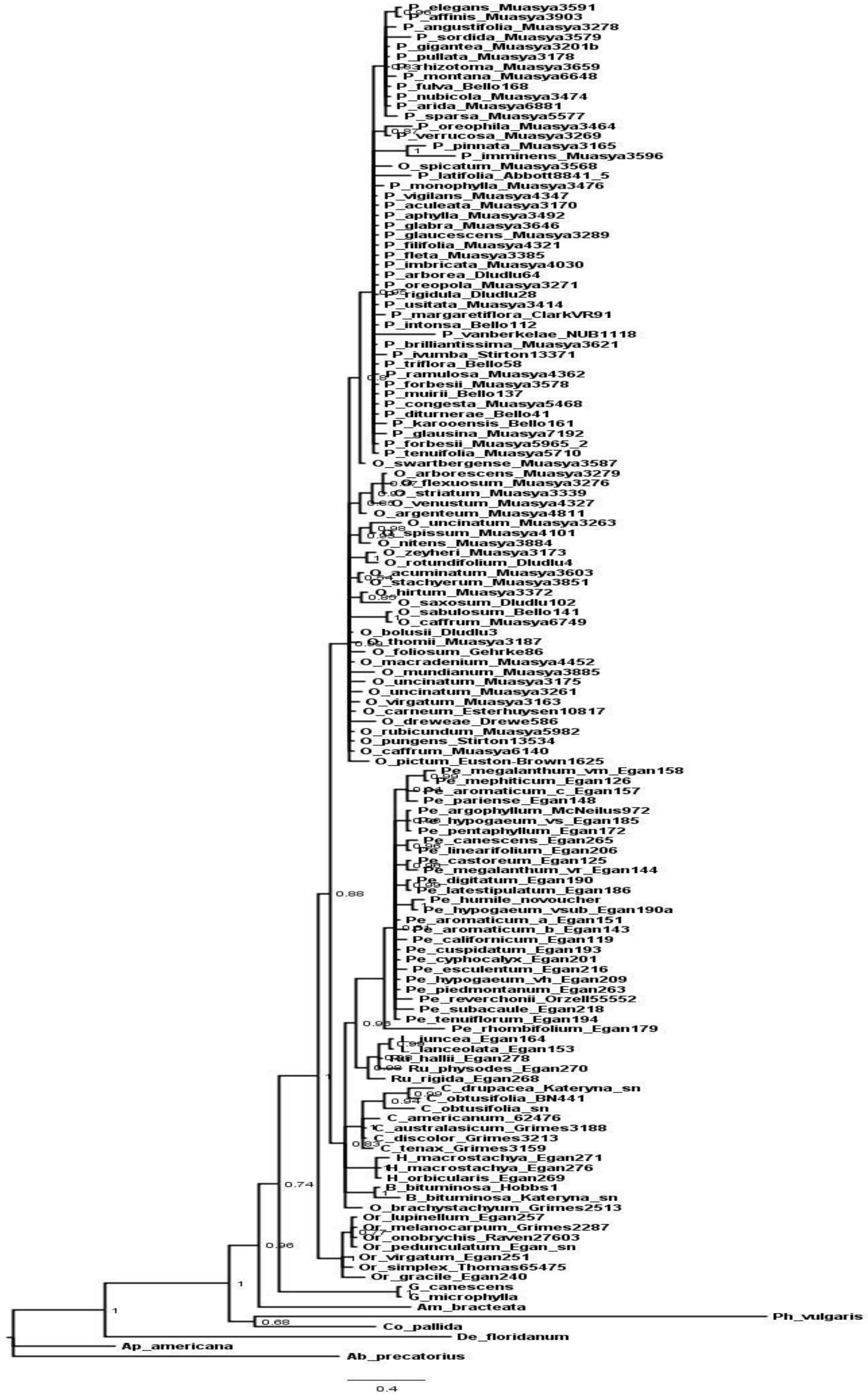
Appendix B9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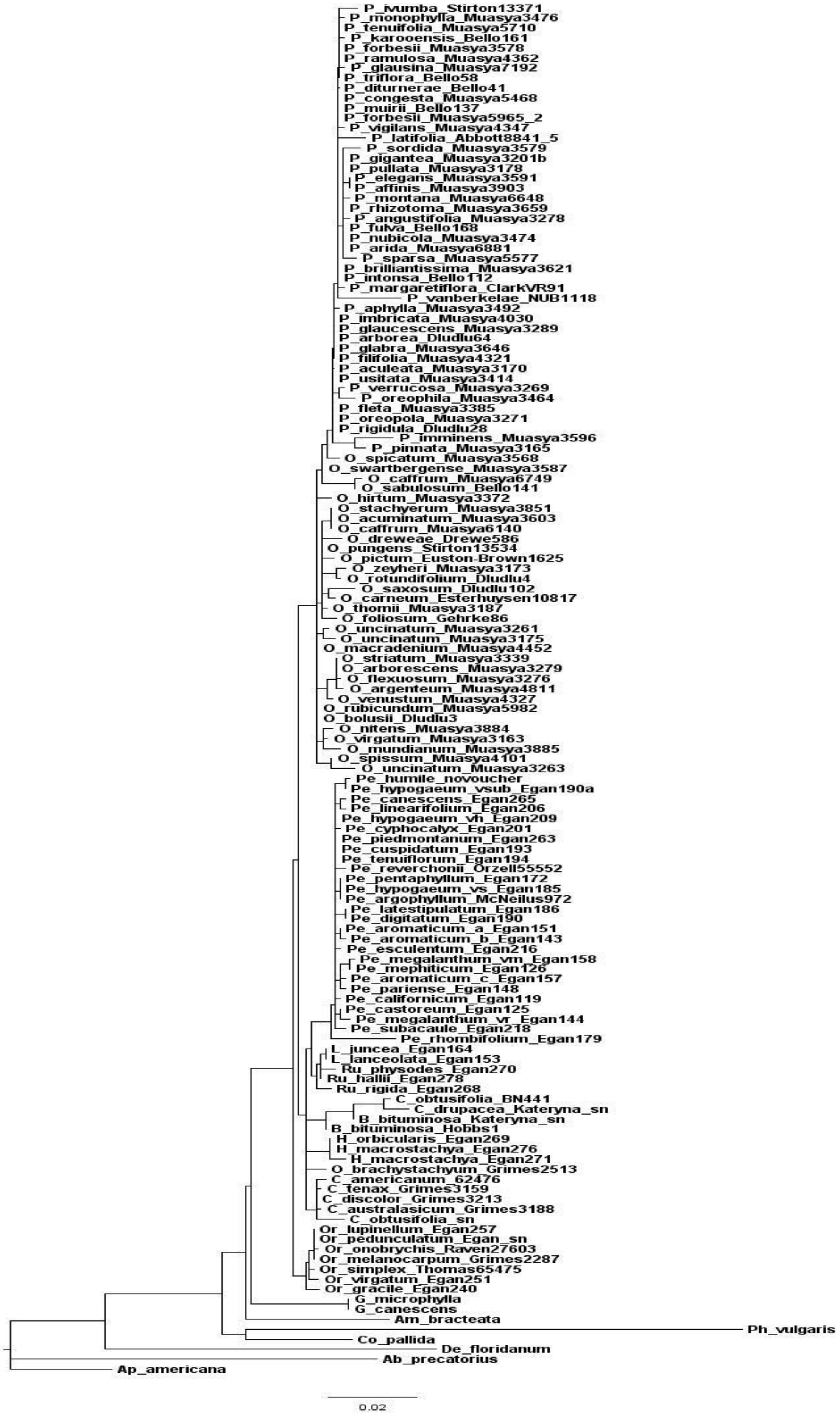
Appendix B10



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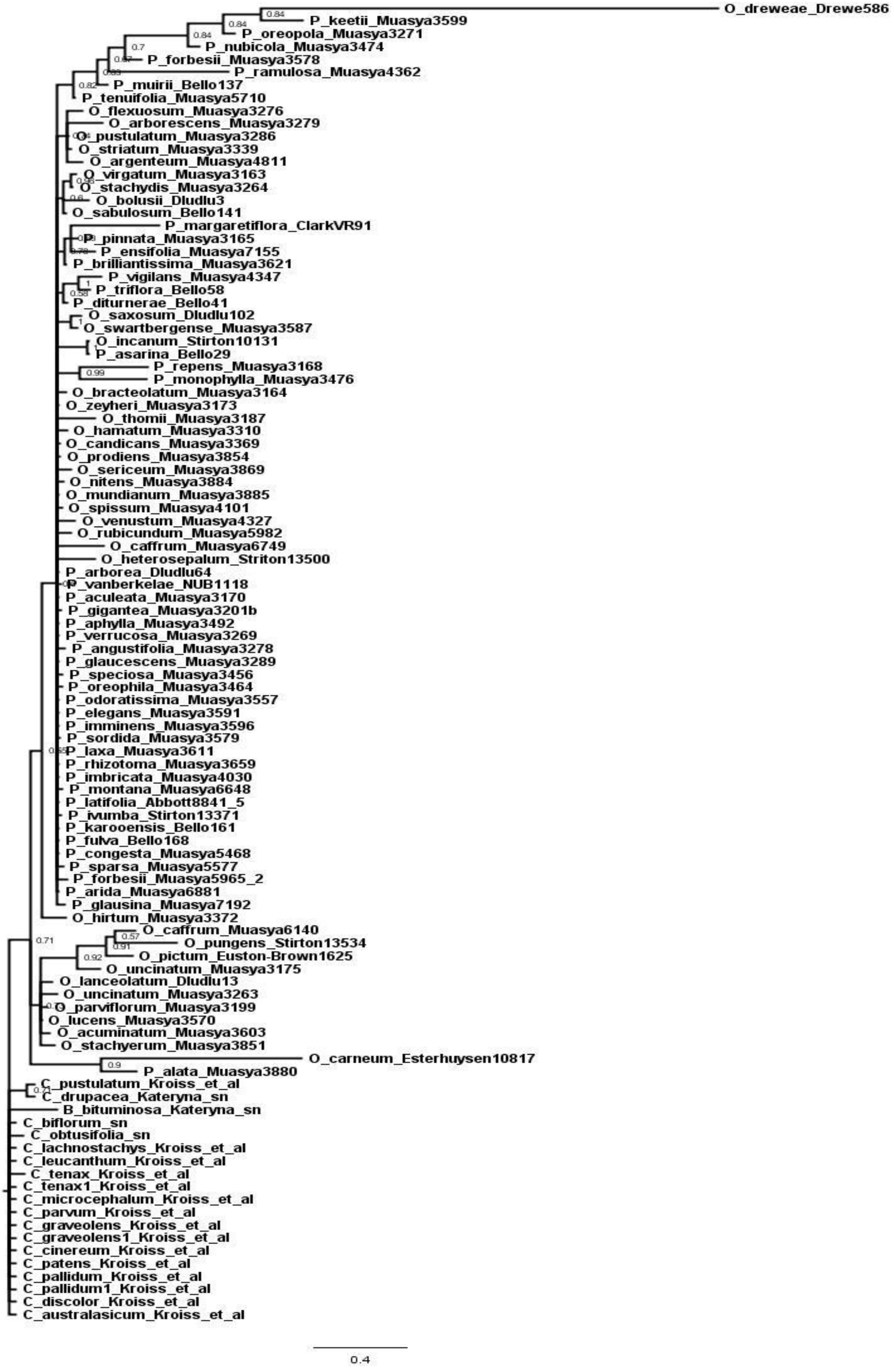
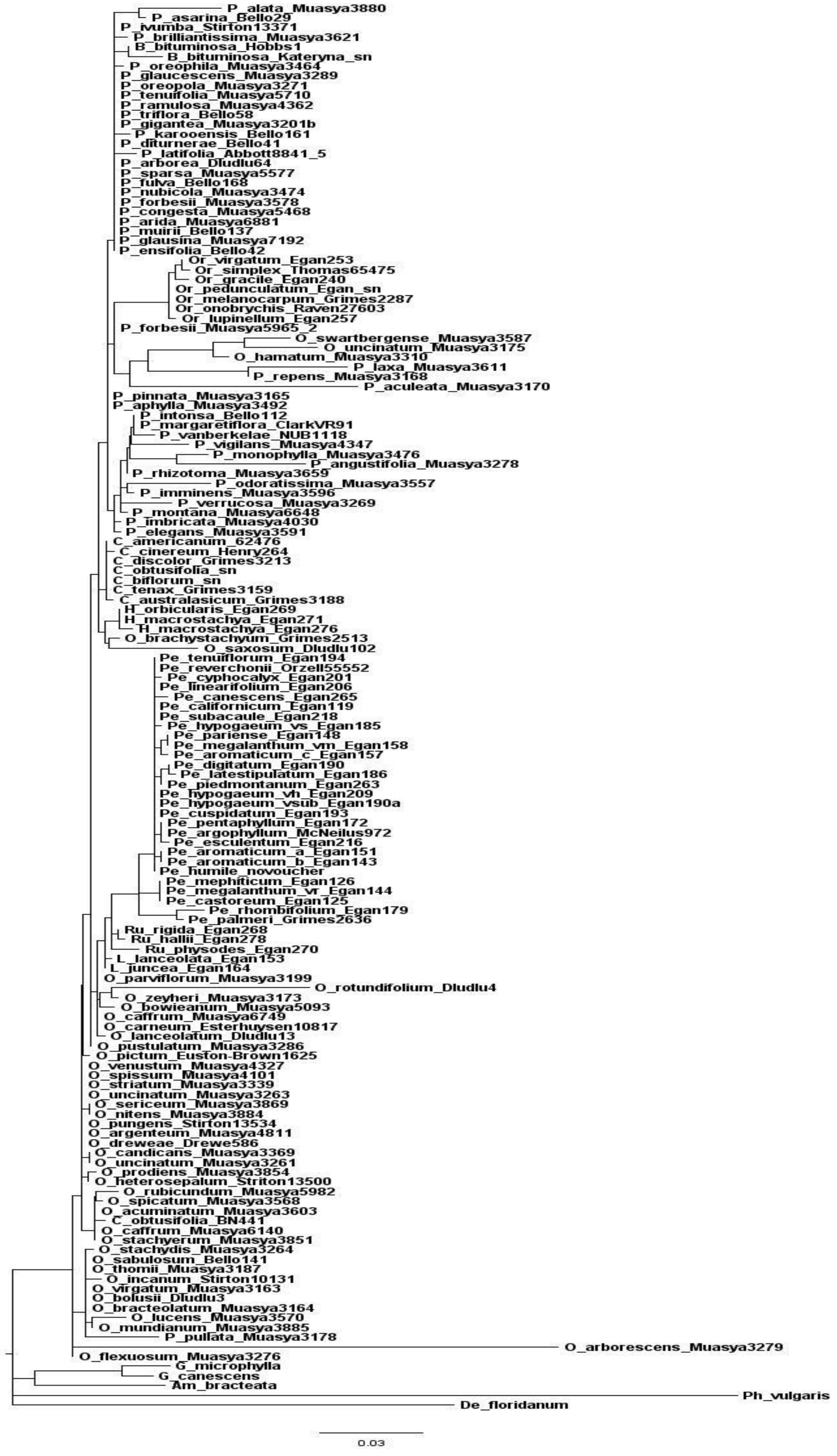


Appendix B12

Appendix B13

Appendix B14



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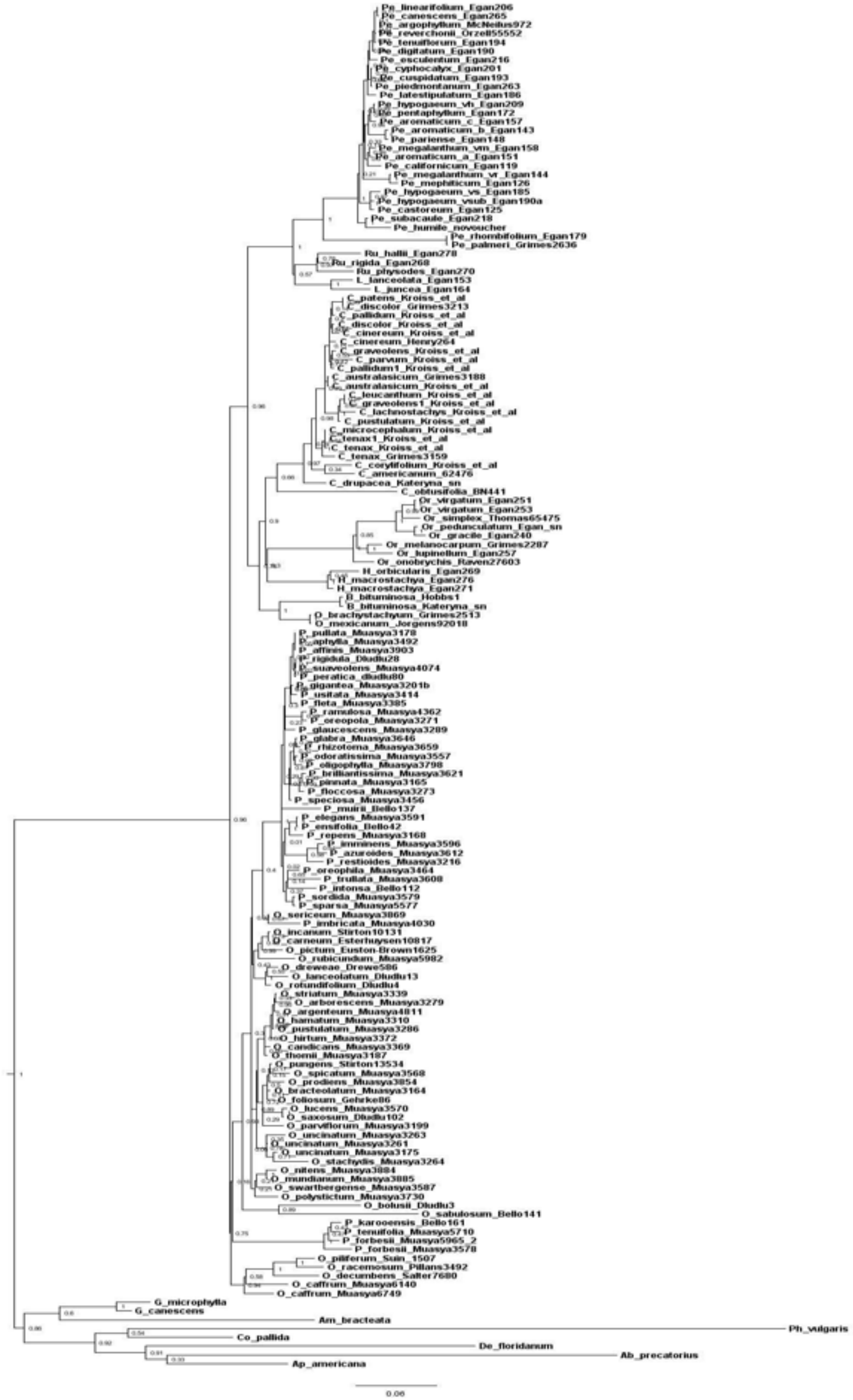
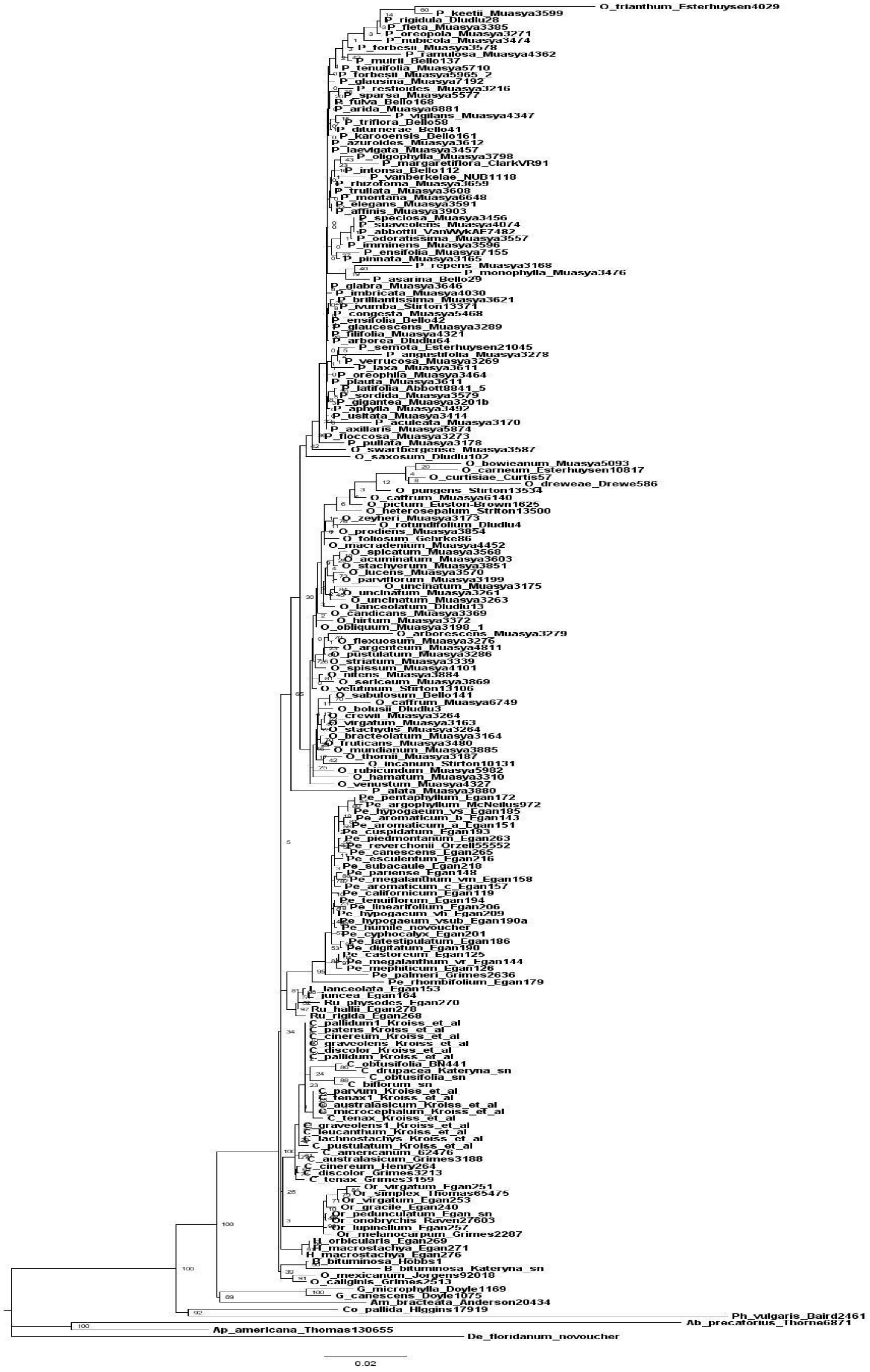


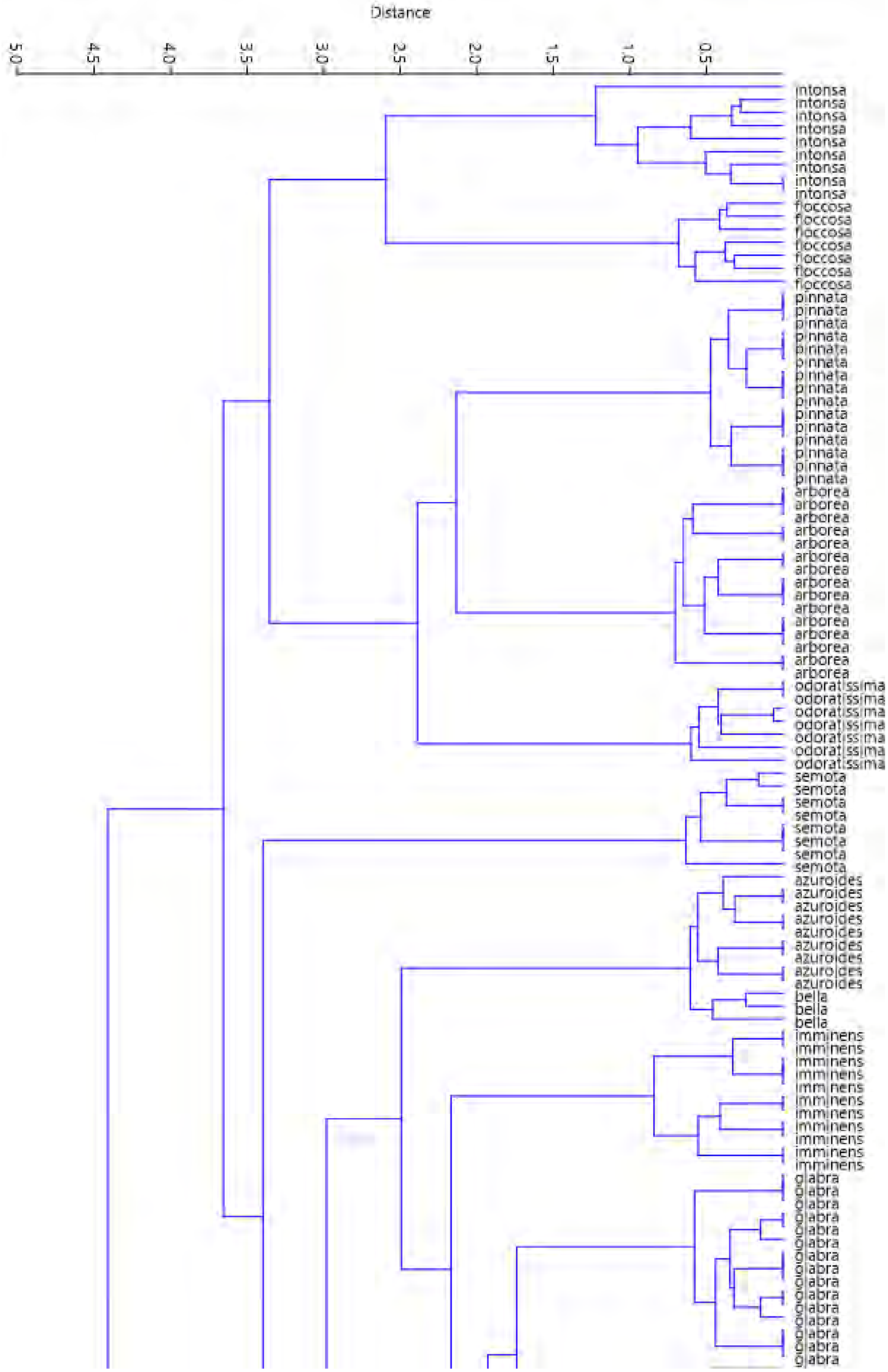
Appendix B16

Appendix B17

Appendix B18



Appendix C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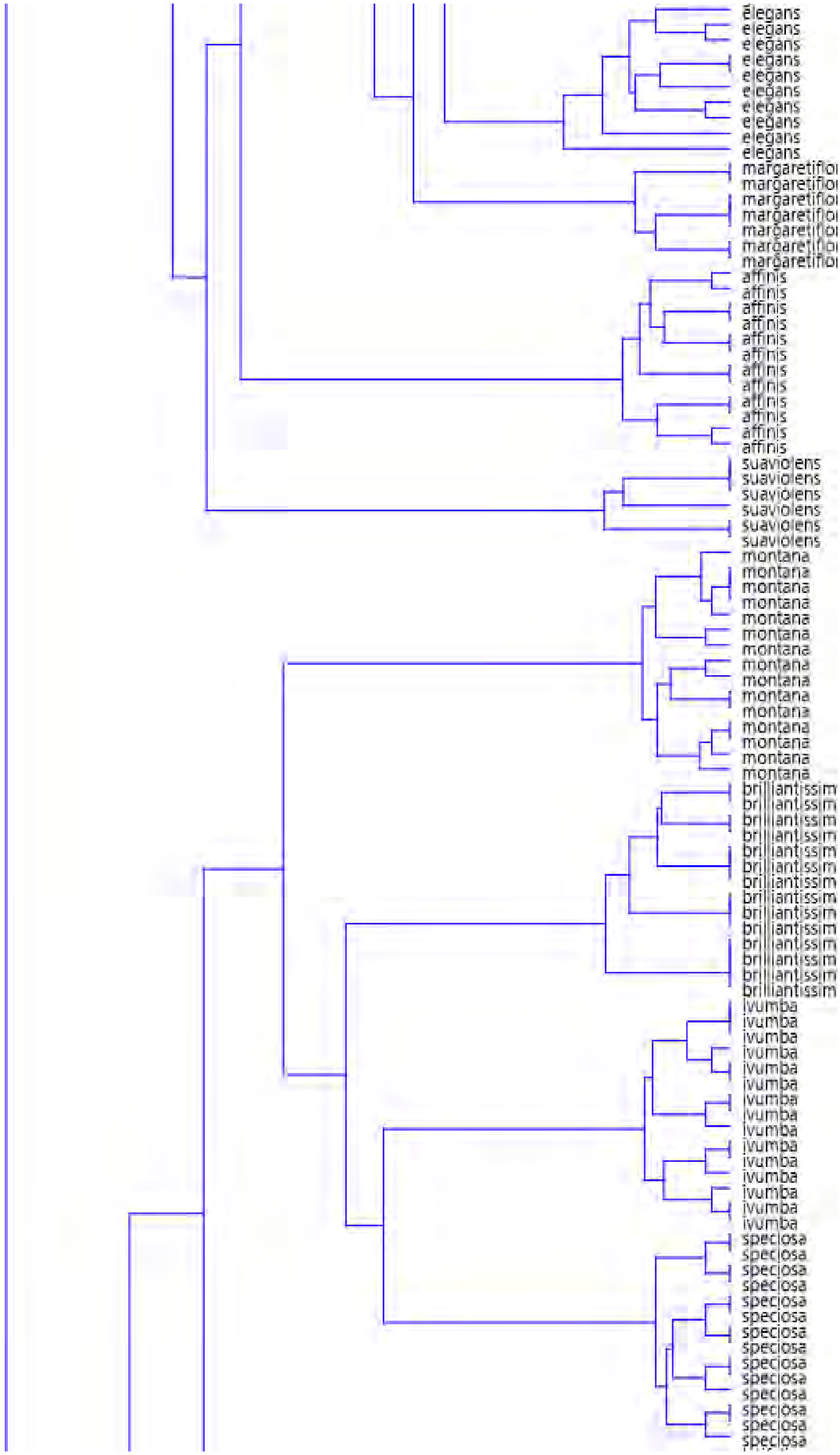
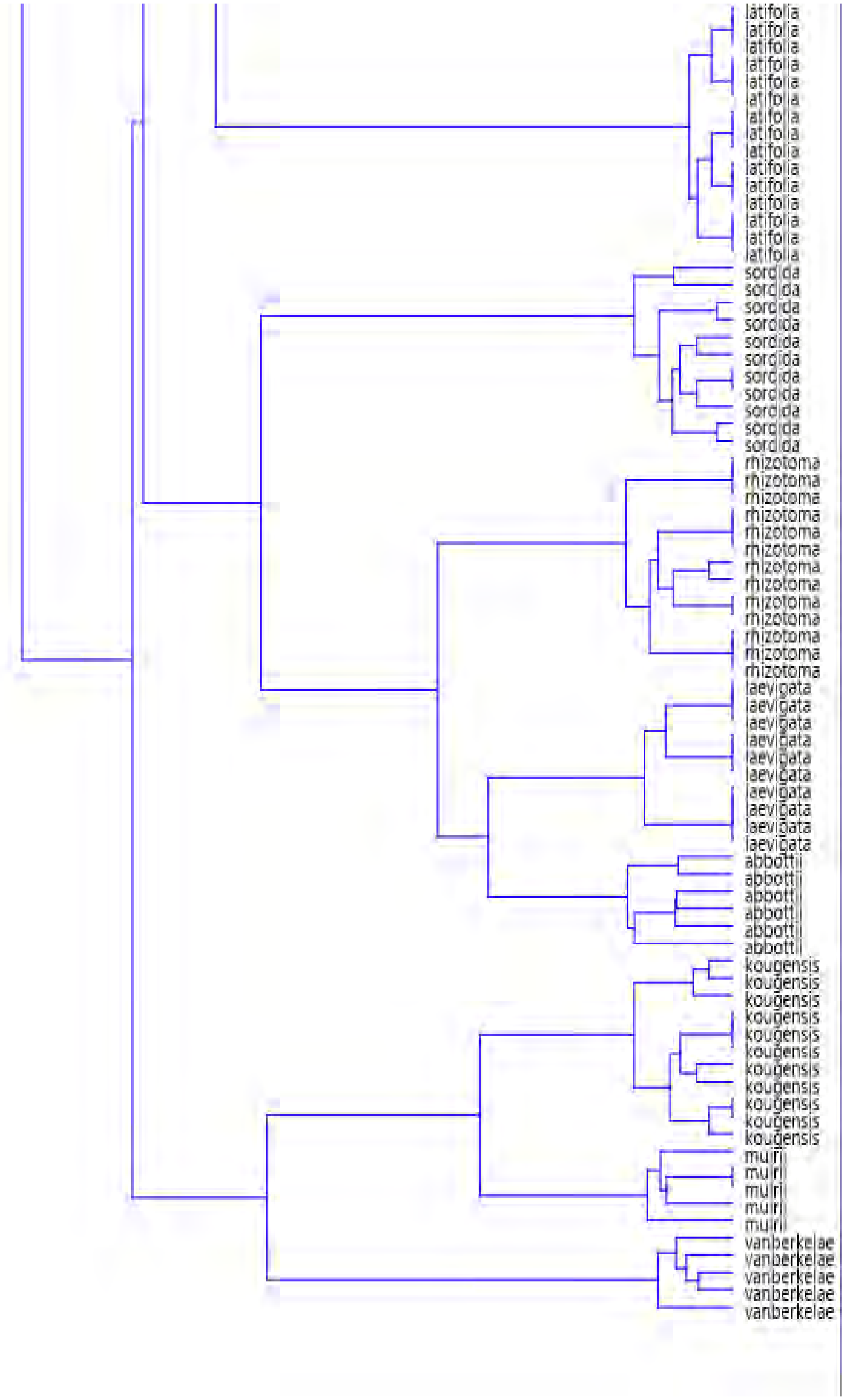
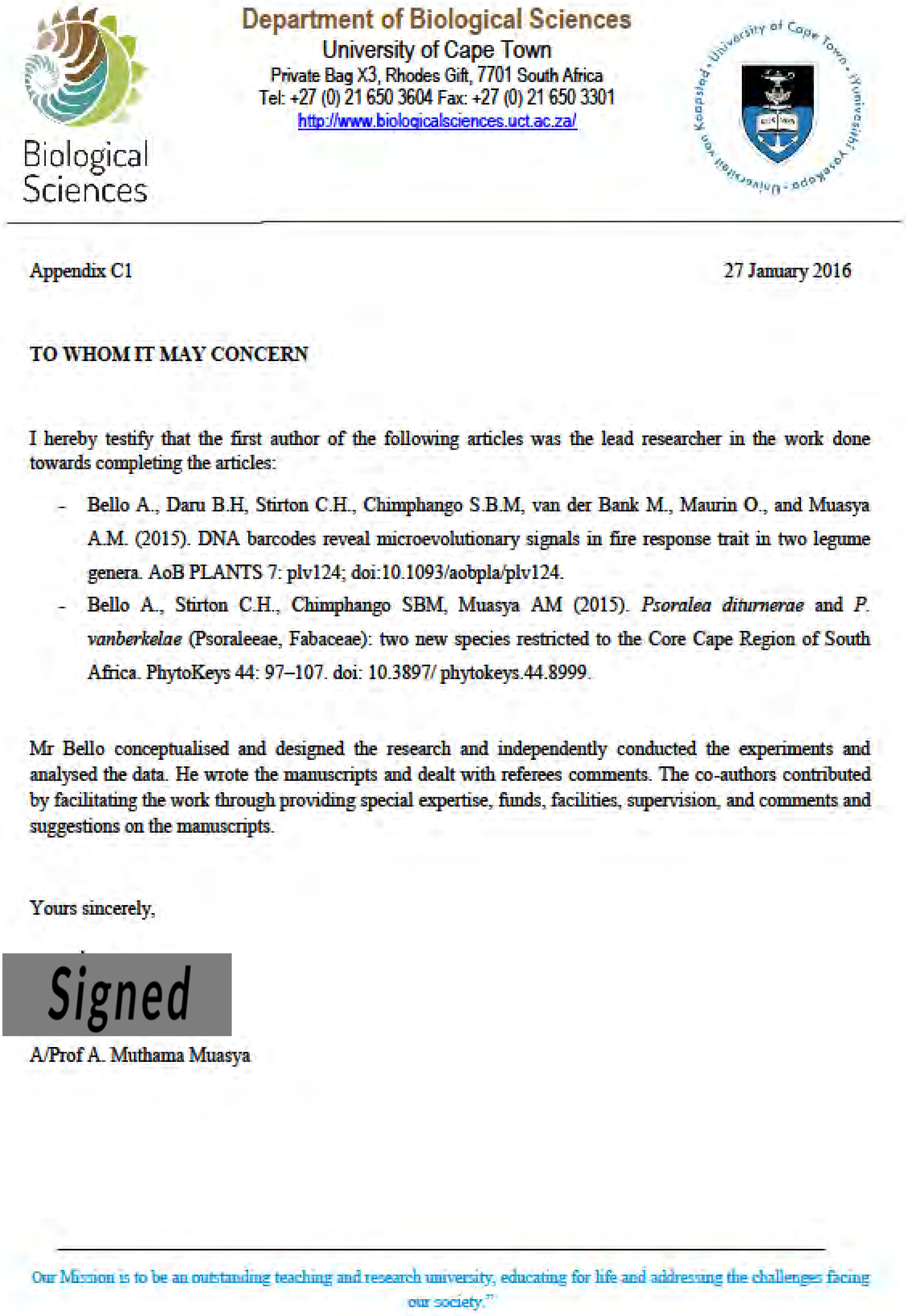
Appendix C. continued



Appendix C. continued



Appendix D1



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Appendix C1

27 January 2016

TO WHOM IT MAY CONCERN

I hereby testify that the first author of the following articles was the lead researcher in the work done towards completing the articles:

- Bello A., Daru B.H, Stirton C.H., Chimphango S.B.M, van der Bank M., Maurin O., and Muasya A.M. (2015). DNA barcodes reveal microevolutionary signals in fire response trait in two legume genera. AoB PLANTS 7: plv124; doi:10.1093/aobpla/plv124.
- Bello A., Stirton C.H., Chimphango SBM, Muasya AM (2015). *Psoralea diturnerae* and *P. vanberkelae* (Psoraleeae, Fabaceae): two new species restricted to the Core Cape Region of South Africa. PhytoKeys 44: 97–107. doi: 10.3897/ phytokeys.44.8999.

Mr Bello conceptualised and designed the research and independently conducted the experiments and analysed the data. He wrote the manuscripts and dealt with referees comments. The co-authors contributed by facilitating the work through providing special expertise, funds, facilities, supervision, and comments and suggestions on the manuscripts.

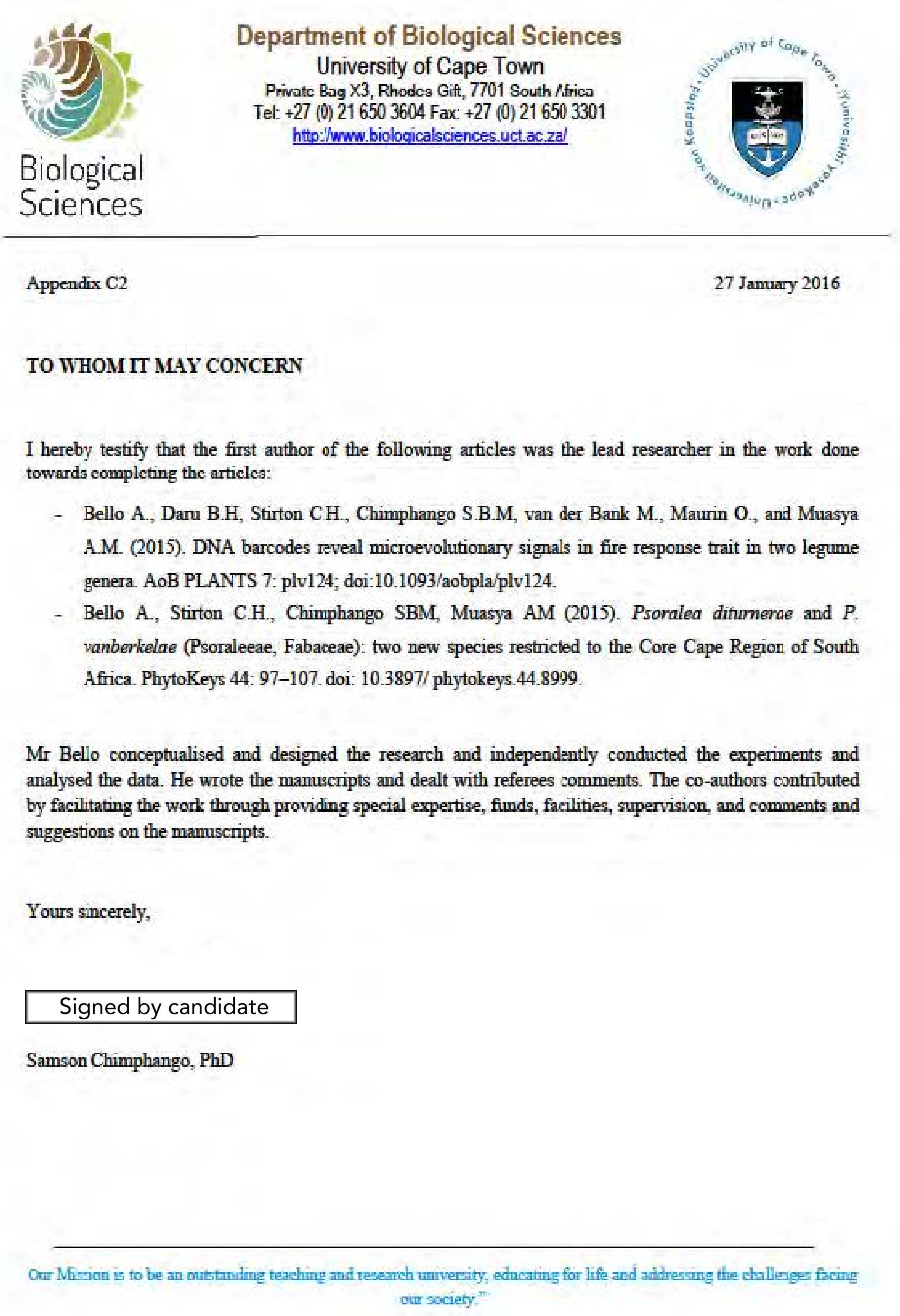
Yours sincerely,

Signed

A/Prof A. Muthama Muasya

Our Mission is to be an outstanding teaching and research university, educating for life and addressing the challenges facing our society.”

Appendix D2



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Appendix C2 27 January 2016

TO WHOM IT MAY CONCERN

I hereby testify that the first author of the following articles was the lead researcher in the work done towards completing the articles:

- Bello A., Daru B.H, Stirton C.H., Chimphango S.B.M, van der Bank M., Maurin O., and Muasya A.M. (2015). DNA barcodes reveal microevolutionary signals in fire response trait in two legume genera. AoB PLANTS 7: plv124; doi:10.1093/aobpla/plv124.
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Yours sincerely,

Signed by candidate

Samson Chimphango, PhD