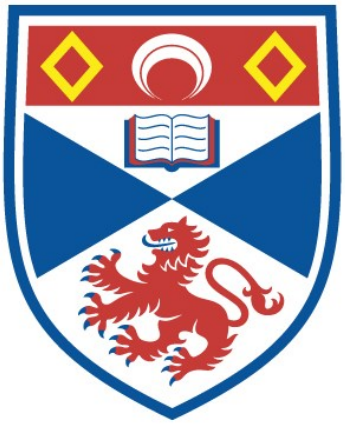
A TAXONOMIC REVISION OF 'HYPENIA' (MART. EX BENTH.) HARLEY (LABIATAE)

Ruth Atkinson

A Thesis Submitted for the Degree of PhD
at the
University of St Andrews



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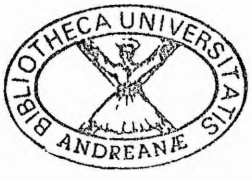

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iii) I hereby certify that the candidate has fulfilled the conditions of the Resolution and Regulations appropriate for the degree of Ph. D. in the University of St Andrews and that the candidate is qualified to submit this thesis in application for that degree

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Abstract

A monograph is presented of *Hypenia* in the plant family Labiatae. *Hypenia* was formerly a genus but in this study it is treated as 23 species in six sections of the genus *Eriope*. 22 of the 23 species treated in this study occupy relatively restricted ranges in the savannas of the central Brazilian plateau and neighbouring parts of the Serra do Espinaço in eastern Brazil. The remaining species occurs in dry scrubland in northeastern Brazil with a disjunction to savanna in northern Venezuela.

The previous classification and taxonomic history of *Hypenia* are outlined followed by a discussion of morphology and cytology in the group. Observations were made on pollination biology of selected species. Cladistic investigations were conducted using morphological characters and the ITS region of the nuclear genome. The morphological and molecular analyses were then combined in a single analysis. The cladistic analyses indicated that *Hypenia* was a paraphyletic genus and that generic boundaries between *Hypenia* and *Eriope* needed to be reconsidered. The classification of *Hypenia* was revised in the context of the phylogeny generated by the combined analysis and with consideration given to the theory of classification with particular reference to recent literature on the subject.

The largest number of species in *Hypenia* belonged to the 'macrantha complex', a group of red-flowered species endemic to central Brazil. Considerable revision of the species in this group was required. The 'macrantha complex' was recognised as *Eriope* section *Hypenia* with two subsections. One of the subsections was characterised by resupination of the corolla, a previously overlooked character. Five new species of *Hypenia* are presented in this account. Distribution maps of all the species described were followed by a discussion of biogeographic patterns in the group.

Acknowledgements

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My field work in Brazil was made possible by the kind attention of many people. Ana Maria Giulietti, Manoel Claudio da Silva and Carol Proença were all extremely generous with their hospitality and gave considerable help with visits to the field. Felipe Ribeiro and Lenise Dolores both took responsibility for my fieldwork in Brazil, a duty for which I am very grateful. In addition the staff of the herbaria at the Universities of São Paulo and Brasília and in the Reserva Ecológica do IBGE all provided much help during my stay at these institutions. I would also like to thank the members of the Neves family in Rio de Contas who kindly allowed me to stay in their house during my stay there. In addition, my stay in Bahia was enlivened by the company of Alison McInroy who was a cheerful companion throughout the trip.

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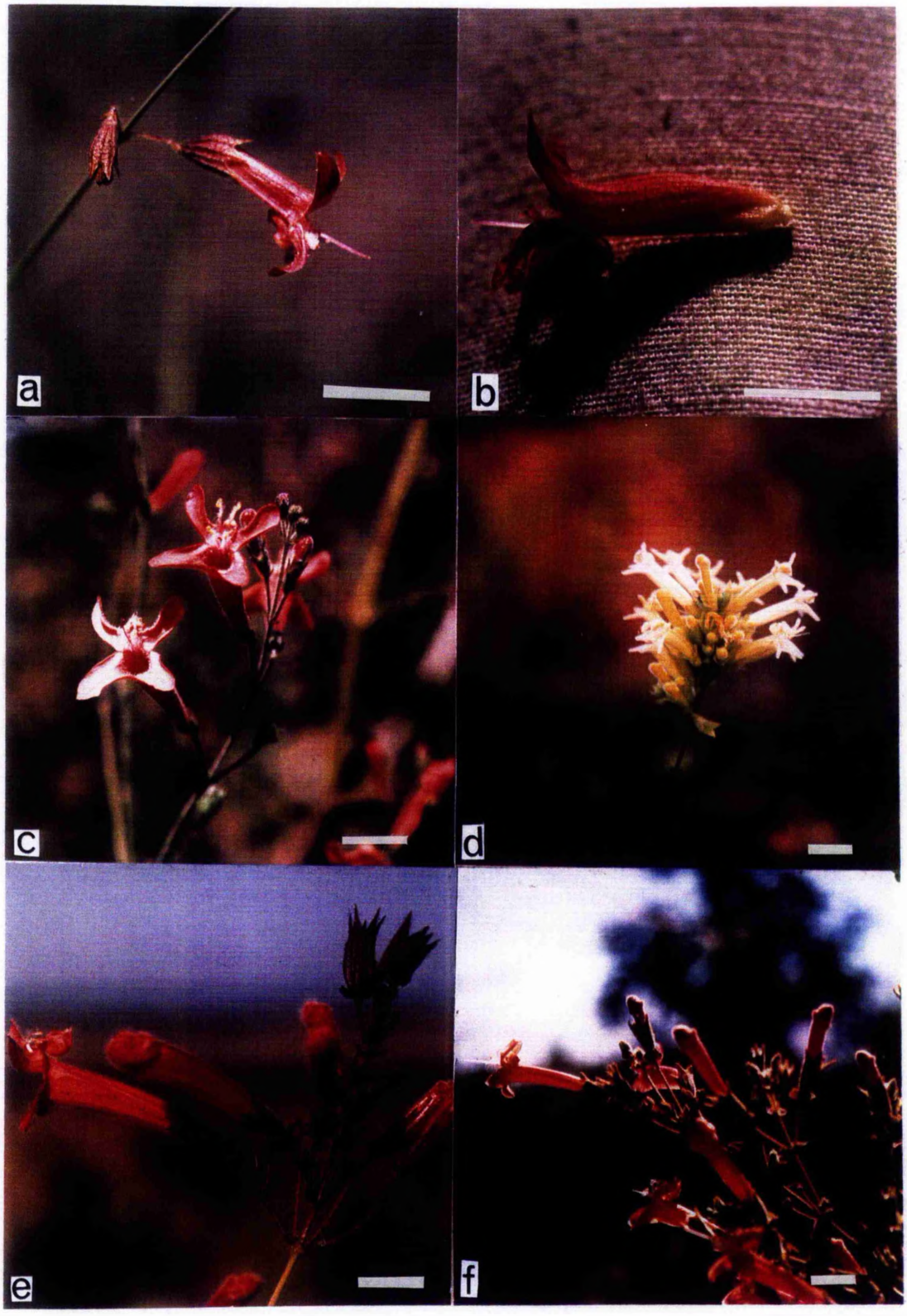


Plate I: Corolla morphology in subsection *Laxiflorae*

a) *E. reticulata;* b) *E. reticulata* showing grooved corolla tube; c) *E. niquelandiensis*; d) *E. crispata*; e) *E. macrantha*; f) *E. calycina* (scale bar = 1cm)

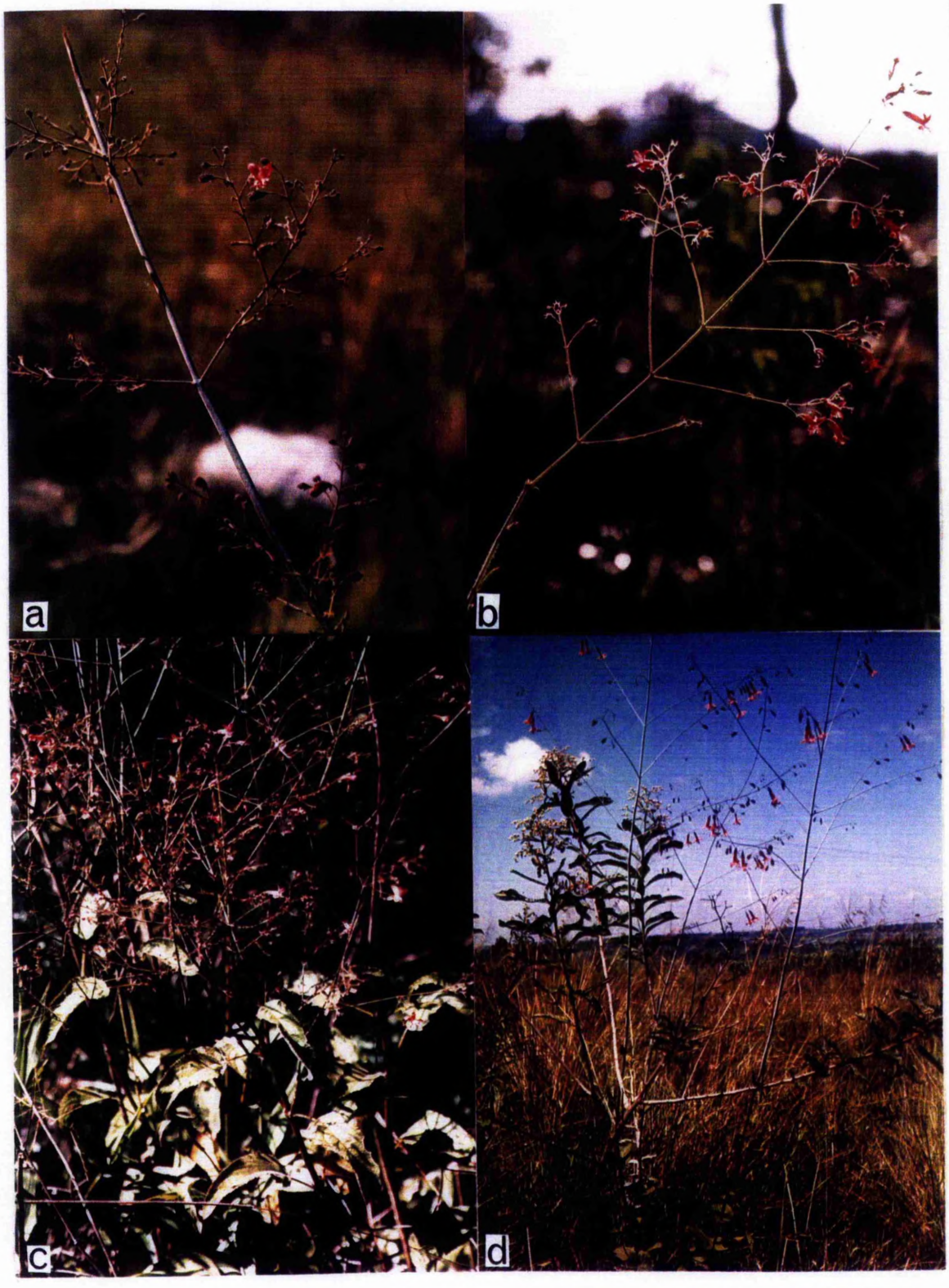


Plate II: Inflorescence morphology in subsection *Laxiflorae*

a) *E. reticulata,* Serra do Cipó; b) *E. aristulata*; c) *E. bracteolata*; d) *E. reticulata* São Paulo

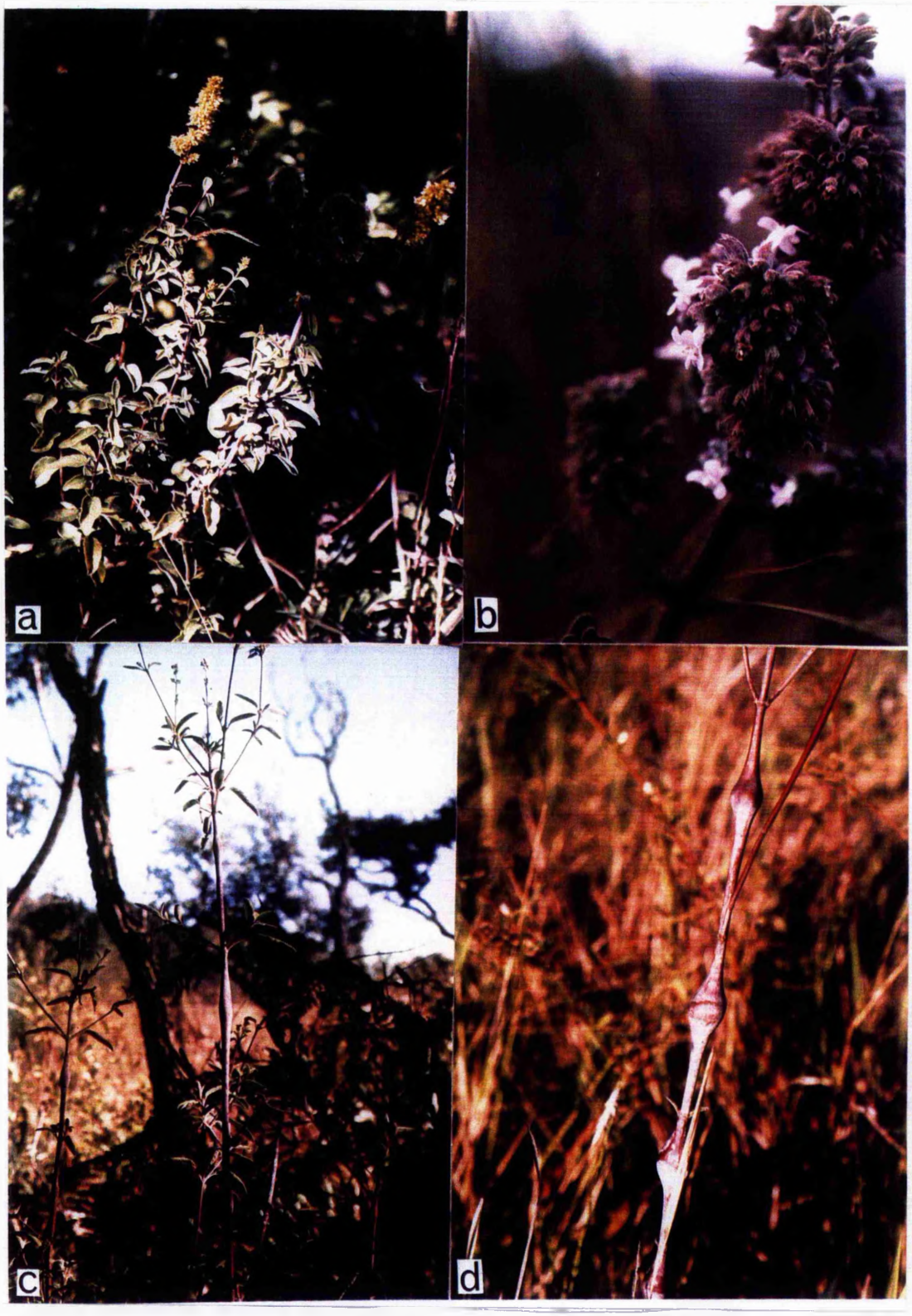


Plate III: Inflorescence and stem of *E. densiflora* and *E. brachystachys*

a) Inflorescence of *E. densiflora*; b) inflorescence of *E. brachystachys*; c) stem of *E. densiflora*; d) stem of *E. brachystachys*

Plate IV: Habit in *Eriope* and inflorescence variation in the Hyptidinae

a) *E. angustifolia;* b) *E. hypoleuca*; c) *Hyptis suaveolens*; d) *Hyptis rugosa*; e) *E. salzmannii* (scale bar = 1 cm)

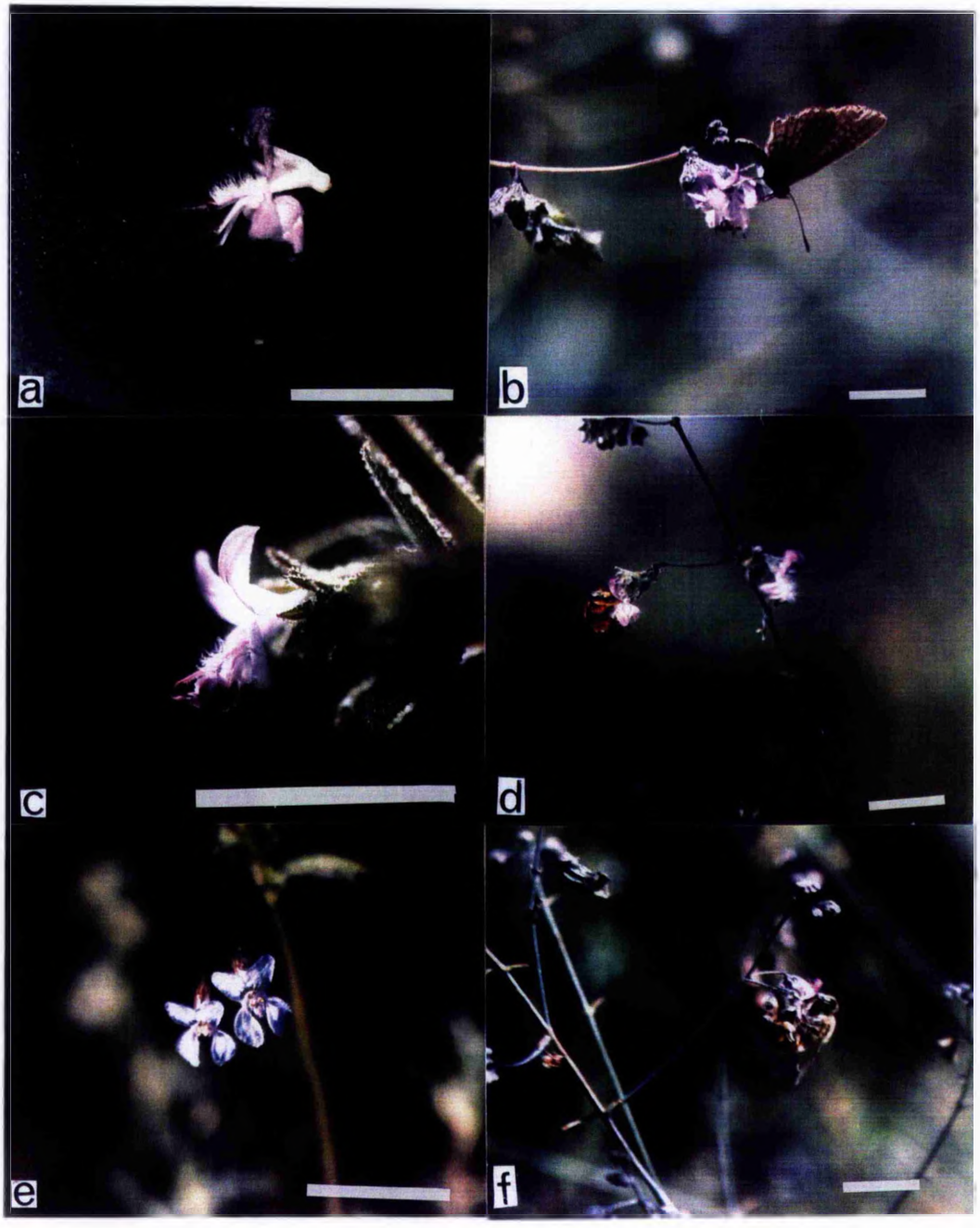


Plate V: Flowers of *E. vitifolia, E. salzmannii* and *Hyptidendron vepretorum* and visitors to *E. vitifolia*

a) *E. vitifolia*; b) butterfly feeding on *E. vitifolia*; c) *Hyptidendron vepretorum*; d) *Paratetrapedia* sp. feeding on *E. vitifolia*; e) *E. salzmannii*; f) *Apis mellifera* feeding on *E. vitifolia* (scale bar = 1 cm)

CONTENTS

Chapter 1: Introduction

This chapter includes a discussion of major classifications of the Labiatae. The place of *Hypenia* in the Labiatae and its previous classification is then introduced. The chapter concludes with an introduction to the taxonomic problems encountered in *Hypenia* and includes a brief outline of the taxonomy used in this work.

i) The classification of *Hypenia*

Hypenia belongs to family Labiatae (preferred to Lamiaceae for its descriptive quality), subfamily Nepetoideae , tribe Ocimeae, subtribe Hyptidinae. The classification of these increasingly exclusive groups is discussed below. Within the Hyptidinae *Hypenia* is closely related to *Eriope* and *Hyptis* and was considered part of the latter until its recent separation (Harley 1988a). Subtribe Hyptidinae is largely confined to South America and *Hypenia* is endemic to the savannas of central and northern South America. Further discussion of the taxonomic history of *Hypenia* and its broad geographic distribution follows the exposition of the taxonomy of the Labiatae.

ii) Circumscription of the Labiatae

The Labiatae was considered by Bentham (1832-1836, 1848 and 1876) to be one of the most natural of flowering plant families. It was distinguished by the bilocular ovary with each locule deeply lobed and divided by a septum to produce four uni-ovulate lobes which give rise to a fruit of four nutlets. The style rises between the lobes from the base of the ovary. The most closely related family, and the one from which the Labiatae are thought to be derived, is the Verbenaceae (Cronquist 1981). The ovary in the Verbenaceae is very variable but it is basically bilocular and can be very similar to that described for the Labiatae. Thus, the traditional definition of the Labiatae was not exclusive. Pollen exine structure indicated that different groups in the Labiatae evolved independently several times from within the Verbenaceae (Raj 1983). A cladistic analysis of the two families using 85 palynological, phytochemical, embryological, anatomical and morphological characters supported this finding (Cantino 1992a). As a result, the Labiatae was subject to major reappraisal and many genera previously placed in the Verbenaceae were transferred into the Labiatae (Cantino 1992b, Cantino et al. 1992).

Classifications of the angiosperms based on morphology have treated the Labiatae as a member of the order Lamiales in the superorder Asteridae (Cronquist 1981); or as a member of the subclass Lamiidae which includes the superorders Solananae, Gentiananae and Lamianae (Takhtajan 1996); or alternatively as a suborder of the Scrophulariales in the superorder Gentiananae (Thorne 1992) (higher taxon endings are those used in the works cited).

Molecular analysis of the *rbc*L gene in the chloroplast genome (Olmstead et al. 1992a, 1992b, Wagstaff & Olmstead 1997) indicates that the Labiatae belongs to the Asteridae. The evidence from *rbc*L sequence data suggests that the Lamiales should be united with the Scrophulariales, which is paraphyletic by the exclusion of the Lamiales. The Lamiales *sensu stricto*, i.e. Labiatae and Verbenaceae, show little total variation in *rbc*L sequences but the data supports a polyphyletic origin of the Labiatae and that the Verbenoideae is the sister group of the rest of the Verbenaceae and Labiatae (Olmstead et al. 1992b, Wagstaff & Olmstead 1997). They also suggest that the Avicenniaceae and Symphorematacеae, which are closely allied to the Verbenaceae and often included within it (Cronquist 1981), justify recognition at the family level.

iii) Classification of the Labiatae

The Labiatae is distributed throughout the world and is found in a wide range of habitats with major centres of diversity in China, Southwest Asia, Mexico and South America. It is a large family of about 3000 (Heywood 1993) or 5000 (Cantino 1992b) species. Different estimates of numbers of species in the Labiatae reflect different concepts of family limits discussed below.

The Labiatae has been subject to several different classifications. Four major classifications by Bentham (1876), Briquet (1896), Erdtman (1945) and Wunderlich (1967) were reviewed by Cantino (1992b) and compared to current understanding of the family.

Bentham's (1876) classification had eight tribes but Briquet (1896) elevated five of them to subfamily level, combined three as subfamily Lamioideae and elevated Bentham's subtribe Scutellariinae to subfamily Scutellarioideae. Erdtman's (1945) classification was solely based on palynological features and recognised just two subfamilies, the Lamioideae, with tricolpate pollen shed in a two-celled stage and the Nepetoideae, with hexacolpate pollen

shed in a three-celled stage. Wunderlich (1967) found other morphological and embryological characters which correlated with palynology and her classification continued to recognise the large subfamily Nepetoideae proposed by Erdtman (1945). In other respects Wunderlich's classification resembled that of Briquet (1896). Cantino (1992a and 1992b) found considerable support for subfamily Nepetoideae and congruence with many features of Wunderlich's (1967) system.

None of the four classifications considered by Cantino (1992b) accounted for the multiple, polyphyletic, origin of the Labiatae from the Verbenaceae (Cantino 1992a). Junell's (1934) suggestion that the Verbenaceae be restricted to subfamily Verbenoideae was supported by Cantino (1992b). The transfer of 50 or so non-Verbenoideae genera to the Labiatae provided support for single, monophyletic, origins for the Verbenaceae and the Labiatae. In this case the Verbenaceae was supported by several characters including the absence of multi-cellular trichomes (El-Gazzar & Watson 1970, El-Gazzar 1974) and pollen exine thickening near the apertures (Raj 1983, Chadwell et al. 1992). The Labiatae was less well-supported although a possible defining character was the manner of attachment of the ovules to the carpel (Junell 1934).

The classification of the family presented by Cantino et al. (1992) attempted to reconcile the evidence for multiple origins of the traditionally circumscribed Labiatae with the evidence from phylogenetic and morphological studies. Their classification is followed in this thesis.

iv) Subfamily Nepetoideae

The Nepetoideae is the largest subfamily in the Labiatae and is characterised by: hexacolpate pollen shed in a three-celled stage; exalbuminous seeds with an investing embryo; the absence of iridoid glycosides (Kooiman 1972); the presence of rosmarinic acid; a high volatile terpenoid content, causing many species to be aromatic; highly saturated seed oils; and frequently a mucilaginous pericarp (Ryding 1992a). The pollen type, exalbuminous seeds and absence of iridoid glycosides are all unique to the subfamily (Wagstaff et al. 1995) and the Nepetoideae is considered to be the most advanced group in the Labiatae (Cantino & Sanders 1986). In the RFLP analysis of the chloroplast genome by Wagstaff et al. (1995) the Nepetoideae were the sister group to the rest of the Labiatae.

The Nepetoideae is the only subfamily in the Labiatae to be divided into tribes, of which Cantino et al. (1992) recognise Elsholtzieae, Lavanduleae, Mentheae and Ocimeae. *Hypenia* belongs to the tribe Ocimeae and this largely tropical tribe can be recognised by the position of the stamens. In most other Labiatae the two pairs of stamens are held under the posterior lobe of the usually zygomorphic corolla, or sometimes are spreading as in *Mentha* L., but in the Ocimeae this position is altered so that the stamens are held against the anterior lobe of the corolla, i.e. they are declinate. The Lavanduleae also has declinate stamens but has several unique features, including gynophore lobes opposite rather than alternate to the ovary lobes. Recent molecular analysis of the subfamily using restriction fragment length polymorphisms (RFLPs) indicate that the Ocimeae, including *Lavandula* Tourn. ex L., is the sister group to the rest of the Nepetoideae (Wagstaff et al. 1995). This suggests that *Lavandula* and its close relatives in the Lavanduleae may be better placed in the Ocimeae. It also indicates the early divergence of the Ocimeae from the rest of the Nepetoideae.

v) Tribe Ocimeae

Floral characters are numerous in the Labiatae (Hedge 1992) and characters associated with the calyx and the anterior lobe of the corolla provide the basis of the classification of the Ocimeae into subtribes.

Bentham (1833 and 1848) and Briquet (1896) split the Ocimeae into three subtribes which are distinguished by the position of the stamens against the anterior lobe. Genera in subtribe Ociminae have their stamens exserted from the corolla, members of subtribe Plectranthinae hold their stamens against the boat-shaped anterior lobe, whilst the Hyptidinae enclose the stamens in the anterior lobe prior to anthesis. The distinction between the Ociminae and the Plectranthinae is poorly made and some genera (e.g. *Orthosiphon* Benth.) have intermediate corolla morphology and stamen position. Three genera, *Isodon* (Benth.) Schrad. ex Spach, *Hanceola* Kudô and *Siphocranion* Kudô have divergent pericarp structure and their position in the tribe is uncertain (Paton & Ryding, in press).

vi) Subtribe Hyptidinae

The Hyptidinae is a group of about 450 species, more than half of which belong to the genus *Hyptis* Jacq.. Most variation within the subtribe is associated with vegetative characters and the structure of the inflorescence. Vegetative characters are very variable between species whereas inflorescence structure and some floral characters are more conservative and have

been used as the basis for classification of the subtribe. Morphological variation in *Hypenia* and its relatives is discussed in chapter 2 with particular emphasis on inflorescence development in the Hyptidinae.

Members of subtribe Hyptidinae possess a boat-shaped anterior corolla lobe which is modified by being thickened at the base and laterally compressed with interlocking fimbriate margins holding the stamens under pressure (see figure 2-7). If the flower is visited the anterior lobe releases the stamens and flexes backwards, hinging on the thickened base. This releases pollen in an explosive action against the lower side of the visitor's body. Thus the Hyptidinae are characterised by a precise pollination mechanism. The genus *Isodon* has similar calyx morphology and stamen position to the Hyptidinae but lacks the explosive corolla mechanism. Within the Hyptidinae the corolla structure varies in the details of size and shape of the tube and in the size and position of the lobes. Past taxonomic interpretations of morphological variation are discussed below.

vii) Generic circumscription in the Hyptidinae

Bentham (1833) laid the foundations for the classification of the genera in the Ocimeae. He described *Hyptis*, *Peltodon* Pohl, *Marsypianthes* Mart. ex Benth., *Eriope* Kunth ex Benth. and *Raphiodon* Benth. Bentham (1848) later elaborated this classification by introducing further infra-generic taxa. Briquet (1896) introduced a more complex infra-generic classification but this was simplified by Epling (1936) who largely returned to Bentham's (1848) classification. In addition Epling (1932) had separated three Mexican and Central American species of *Hyptis* to create a new genus, *Asterohyptis* Epling. Harley, in his revision of *Eriope* (1976), created the monotypic genus *Eriopidion* Harley and in a later publication Harley (1988a) elevated *Hyptis* section *Hypenia* to generic rank and also combined two sections of *Hyptis*, *Umbellaria* Benth. and *Buddleoides* Benth., to make the new genus *Hyptidendron* Harley.

The currently accepted genera of the Hyptidinae are *Hyptis, Peltodon, Marsypianthes, Raphiodon, Asterohyptis, Eriope, Eriopidion, Hypenia* and *Hyptidendron.* The classification of the Hyptidinae followed here is the unpublished scheme proposed by Harley and presented in appendix I. Those genera of the Hyptidinae which have never been considered close to *Hypenia,* i.e. *Asterohyptis, Marsypianthes, Rahpiodon* and *Peltodon* are described briefly

below followed by a more detailed discussion of the relationships of the remaining genera of the Hyptidinae with *Hypenia.*

Asterohyptis, Raphiodon, Marsypianthes* and *Peltodon

The three species of *Asterohyptis* lack the conspicuously differentiated anterior lobe characteristic of the remainder of the Hyptidinae. This introduces some doubt as to the place of this genus in the Hyptidinae since it lacks the definitive characters of the subtribe. However, other aspects of floral morphology, notably the calyx, suggest the allegiance of *Asterohyptis* with the Hyptidinae. This suggests that the differentiated anterior lobe may have been lost in *Asterohyptis*.

Raphiodon, Marsypianthes and *Peltodon* are small genera which were first described by Bentham (1833). He defined *Raphiodon* by its calyx which bears spines between the lobes; *Marsypianthes* by its nutlets which fuse to the base of the style leaving a jagged scar on their adaxial face after abscission; and *Peltodon* by the cochleariform apices of its calyx lobes. Bentham (1833) and Epling (1949) both noted the close affinities of all these genera, particularly *Peltodon* and *Raphiodon,* with *Hyptis,* but they have maintained their status in all classifications of the Hyptidinae. They have never been regarded as closely allied to *Hypenia.*

Hyptis

The largest genus in the Hyptidinae is *Hyptis* with 277 species in 26 sections. *Hyptis* has very diverse vegetative form and inflorescence structure. Variation in inflorescence characters is particularly great and was used to split the genus into a large number of more or less easily identified sections. Epling (1949) noted the possibility of recognising sections of *Hyptis* as separate genera although he rejected this approach on the grounds of causing excessive name changes. Section *Hypenia* was separated from *Hyptis* by Harley (1988a) because of its divergent morphology (see below) and most sections of *Hyptis* are morphologically distinct from *Hypenia.* However, there has been some confusion between *Hypenia* and the former section *Siagonarrhen* Benth. discussed below.

Hyptis* section *Siagonarrhen

Bentham (1833) first described section *Siagonarrhen* but noted the difficulty of separating sections *Hypenia* and *Siagonarrhen.* Epling (1936) abandoned the section and transferred a number of species into *Hyptis* section *Hypenia* subsection *Densiflorae* Benth. These species were moved again by Harley (1976) into *Eriope.* The remaining species in section *Siagonharren* were transferred by Epling (1936) into *Hyptis* section *Buddleoides* Benth. (now *Hyptidendron*).

Hyptidendron

Bentham (1833) described two sections of *Hyptis, Buddleoides* and *Umbellaria,* which Harley (1988a) combined as the genus *Hyptidendron* although he maintained the two sections. Harley (1988a) indicated the possibility of close relationships between *Hypenia, Eriope* and *Hyptidendron.* This was supported by Bentham's inclusion of species, now recognised as belonging to *Hyptidendron,* in *Hyptis* section *Siagonarrhen* together with species now assigned to *Eriope* and *Hypenia.*

Eriope

The boundary between *Eriope* and *Hypenia* (or *Hyptis* section *Hypenia*) has been subject to reappraisal from Bentham (1833) and Epling (1936 and 1949) to Harley (1974, 1976, 1988a) and a consideration of *Eriope* is essential to the understanding of *Hypenia.*

Bentham (1833) recognised *Eriope* by its turbinate calyx with fruit obscured by dense white hairs in the throat and by the deflexed peduncle. Harley (1976 & 1988a) clarified the generic boundary of *Eriope* by recognising the importance of the corolla characters and the stylopodium, the base of the style which projects above the nutlets and persists after stylar abscission, in defining the genus. This caused him to transfer a number of species into *Eriope* from *Hyptis* sections *Siagonarrhen* and *Hypenia* (Harley 1976 & 1988a). Harley has continued to work on *Eriope* and has described several new species (Harley 1992) since the publication of his original monograph (Harley 1976). Confusion between *Hypenia* and *Eriope* has occurred because of their shared possession of habit characters, collectively known as the 'greasy pole syndrome' (Harley 1991), and the shared inflorescence morphology of some species of both genera.

Eriopidion

Harley (1976) separated the monotypic genus, *Eriopidion* from *Eriope* on the basis of the modified corolla, hygroscopic calyx and lack of a stylopodium of its sole member *E. strictum* (Benth.) Harley. It had previously been treated by Epling (1936) as section *Tubiflorae* Epling in *Eriope*. *E. strictum* was included in a morphological cladistic analysis which suggested it was closely allied to *Eriope* section *Eriope* (Ross unpublished).

viii) Species circumscription in *Hypenia*

The species included by Bentham (1833 & 1848) in *Hyptis* section *Hypenia* were described on the basis of a limited number of specimens and Bentham appeared to have had little difficulty in circumscribing species. Subsequent taxonomic accounts of *Hypenia* continued to

recognise most of Bentham's original species with the addition of several new ones (Moore 1895, Briquet 1896, Epling 1949). Bentham (1848) split *Hyptis* section *Hypenia* into two subsections, *Densiflorae* and *Laxiflorae* according to whether the flowers were arranged in a loose panicle or held in dense cymose heads. This classification was maintained by Epling (1936, 1949). However Epling had problems in delimiting species in subsection *Laxiflorae* and this has proved to be the biggest species-level problem in *Hypenia.*

Subsection *Laxiflorae*

Subsection *Laxiflorae* sensu Bentham (1848) and Epling (1936 and 1949) constituted the largest group in *Hypenia.* Both Bentham and Epling recognised 15 species. The subsection was characterised by the large, open inflorescence of all the species. Most have large, red flowers but several species with small, blue or lilac flowers were also included because of their similar inflorescence morphology. For the purposes of this discussion the red-flowered species, known as the 'macrantha complex' (Harley 1988b) are separated from the lilac and blue-flowered species.

The 'macrantha complex'

Many of the red-flowered species recognised by Epling (1949) were represented by very few specimens, sometimes from widely diverging localities. In his later work Epling noted the taxonomic difficulty in subsection *Laxiflorae* (Epling 1951): 'The red flowered species of section *Hypenia* represent a puzzling complex and not improbably will prove to be conspecific; in this case *Hyptis macrantha* St. Hilaire is probably the most descriptive name ....'. Later, Epling and Mathias (1957) lumped all the red flowered species in subsection *Laxiflorae* under the name *Hyptis macrantha* St. Hil. ex Benth. They note that any past attempt to describe species in the group '... was a futile attempt to distinguish between the frequently localized, and sometimes remarkably different, aspects of what is probably one variable taxon of the campos throughout Minas Gerais, Goyaz and Mato Grosso.' Epling and Jativa (1968) commented subsequently on the 'puzzling subsection *Laxiflorae*' and alluded to the difficulty of characterising species: 'one may sort a pile of specimens into several modal groups but these will consist mostly of specimens from widely scattered localities. Opposed are the relatively constant characters of habit, corolla and calyx structure and shape, the relatively larger size of the latter within the genus, and the relatively stable indument of the stems.' Epling's concept of one variable red flowered species in subsection *Laxiflorae* was referred to as the 'macrantha complex' (Harley 1988b). The 'macrantha complex' was not a formally recognised taxon but the names *Hyptis macrantha* or *Hypenia macrantha* (St. Hil. ex Benth.) Harley were used as a synonym for all red-flowered *Hypenia* species. The 'macrantha complex' posed the biggest species delimitation problem in *Hypenia.*

Not all the taxa in the 'macrantha complex' were difficult to assign to species. One relatively recently described species, *H. subrosea* (Harley) Harley was very distinctive. It was placed with other species of the 'macrantha complex' on the basis of its open inflorescence (Harley 1974), despite having white flowers.

Non red-flowered species of subsection *Laxiflorae*

The treatment by Epling and Mathias (1957) of the non red-flowered species in subsection *Laxiflorae,* i.e. *H. vitifolia* Pohl ex Benth., *H. salzmannii* Benth., *H. micrantha* Benth. and several taxa now consigned to *Eriope* was confusing. However, these taxa are relatively well-defined. As a result the specific epithets have continued in use.

The non red-flowered species previously included in the *Laxiflorae* presented relatively few problems of species delimitation. *H. vitifolia* (Pohl ex Benth.) Harley has many characters to define it and was not subject to problems of species delimitation. However, *H. longicaulis* Harley ined. is a newly recognised species which was separated from *H. vitifolia* by its habit and chromosome number, but is otherwise morphologically very similar. *H. micrantha* Benth. in DC. presented few problems in species delimitation but the fact that it is the only *Hypenia* species to have been previously placed in *Eriope* indicates its distinctiveness. It is an isolated species both morphologically and geographically and shows a restricted geographic range. It is morphologically distinct in the possession of branched trichomes, exceptionally regular branching in the inflorescence and leaf-like phyllomes. Its corolla size, shape and colour is similar to that of species of *Eriope* sensu stricto. *H. salzmannii* (Benth.) Harley was equally well characterised by floral, habit and leaf characters.

Subsection *Densiflorae*

The main taxonomic problem for previous workers in section *Hypenia,* subsection *Densiflorae* was circumscription of the subsection itself. A number of species were transferred from it into other sections of *Hyptis* and *Eriope* (Harley 1976, 1988a).

At the species-level *H. brachystachys* (Pohl ex Benth.) Harley presented the most problems and was the most variable species in the *Densiflorae.* As circumscribed here it includes four synonyms which represent a wide range of morphological variation. *H. densiflora* (Pohl ex Benth.) Harley, which has a similar distribution to *H. brachystachys* but appears to be ecologically restricted, was far less variable and relatively morphologically isolated. *H. concinna* (Benth. in DC.) Harley was represented by one geographically and morphologically isolated collection.

H. irregularis (Benth. in DC.) Harley was placed in the *Densiflorae* but according to the cladistic analysis it was misplaced in this group because of its lack of wax and fistulose stems and its divergent inflorescence structure. In addition, *H. brachystachys* and *H. densiflora* have connate posterior calyx lobes which are lacking in *H. irregularis* and *H. gracilis*. *H. gracilis* is a new species which was separated from *H. irregularis* because of its inflorescence morphology which shows more resemblance to the 'macrantha complex' than section *Densiflorae*. However, the main difference between *H. irregularis* and *H. gracilis* is in the development of the indeterminate axes. Both species have one-flowered cymes but in *H. irregularis* they are crowded on the indeterminate axes and in *H. gracilis* they are widely spaced. *H. gracilis* is a puzzling taxon and is only represented by two specimens from widely diverging localities and with somewhat different morphologies.

ix) Classification of *Hypenia* used in this thesis

The initial aim was to revise *Hypenia* according to Harley's (1988a) generic boundaries and the species revised in this account correspond to Harley's concept of the genus. However, as a consequence of this taxonomic revision at the species level I have concluded that *Hypenia* should be subsumed within *Eriope* (see chapter 8). Therefore my species-level taxonomic conclusions are presented briefly at the beginning of this work since the species and their classification will be referred to before the detailed taxonomic accounts are given in chapter 10. For the sake of clarity, in the first nine chapters of this thesis I have continued to use the name *Hypenia* to refer to the species which fall within Harley's (1988a) concept of the genus and in these chapters *Eriope* is used in accordance with Harley's classification presented in appendix I. In chapter 8 I discuss the arguments for sinking *Hypenia* as a genus, followed by a synopsis of my revised concept of *Eriope* including *Hypenia*. In chapter 10 the species are described using nomenclature as it applies to *Eriope*.

The infra-generic classification I have used in the first part of this study recognises the red-flowered *Hypenia* species previously lumped by Epling and Mathias (1957) in the 'macrantha complex' as section *Hypenia*. The section is split into subsections *Hypenia* with non-resupinate flowers and *Ellipticae* with resupinate flowers (see chapter 2). Other infra-generic taxa which are required for nomenclatural reasons are presented in chapter 10 but are otherwise not referred to in the general discussion which follows. 14 species were described in this study in the 'macrantha complex'. Species delimitation in this group is discussed in more detail in chapter 9.

x) Species recognised in this thesis

The taxonomic accounts include all the species recognised by Harley (1988a) as belonging to *Hypenia* but inevitably there have been some changes. Several new species are described and several are placed in synonomy. Harley listed 24 species of *Hypenia.* I recognise 22 species with many differences in the circumscription of the taxa. Species recognised in my revision and those in Harley (1988a) are listed in table 1-1.

Table 1-1: Species of *Hypenia* cited by Harley (1988a) and species recognised in this account

Harley (1988a)*	**Atkinson (unpubl.)**
densiflora (Pohl ex Benth.) Harley	*densiflora*
inelegans (Epling) Harley	= *brachystachys*
pruinosa (Pohl ex Benth.) Harley	= *brachystachys*
irregularis (Benth. in DC.) Harley	*irregularis*
-/-	*gracilis* sp. nov.
brachystachys (Pohl ex Benth.) Harley	*brachystachys*
paradisi (Harley) Harley	= *brachystachys*
marifolia (Benth. in DC.) Harley	= *brachystachys*
concinna (Benth. in DC.) Harley	*concinna*
vitifolia (Pohl ex Benth.) Harley	*vitifolia*
-/-	*longicaulis* sp. nov.
micrantha (Benth. in DC.) Harley	*micrantha*
salzmannii (Benth.) Harley	*salzmannii*
subrosea (Harley) Harley	*subrosea*
aristulata (Epling) Harley	*aristulata*
perplexa (Epling) Harley	= *reticulata*
reticulata (Mart. ex Benth.) Harley	*reticulata*
glauca (St. Hil. ex Benth.) Harley	= *reticulata*
calycina (Pohl ex Benth.) Harley	*calycina*
crispata (Pohl ex Benth.) Harley	*crispata*
durifolia (Epling) Harley	*sclerophylla*
macrantha (St. Hil. ex Benth.) Harley	*macrantha*
gardneriana (Benth. in DC.) Harley	= *reticulata*
macrosiphon (Briq.) Harley	*macrosiphon.*
pauliana (Epling) Harley	= *reticulata*

paniculata (Benth.) Harley	*paniculata*
-/-	*coccinea*
-/-	*caiaponiensis* sp. nov.
-/-	*hatschbachii* sp. nov.
-/-	*niquelandiensis* sp. nov.
-/-	*indaiensis* sp. nov.

* Authorities are given for species of *Hypenia sensu* Harley (1988a). Full authorities following the taxonomic results of this study are given in chapter 10.

xi) Background to the present study

New material is rapidly accumulating from central Brazil, until recently one of the least botanically explored parts of the world, and it is now possible to attempt a comprehensive revision of *Hypenia* based on Harley's (1988a) revision of generic boundaries. Previous difficulties with the taxonomy of the genus have arisen partly because much of the geographical range of *Hypenia* has only been sparsely collected. Many of the species in Bentham's (1833) first account of *Hypenia* were named on the basis of only one or a very few specimens collected by the earliest botanical explorers in Central Brazil, notably St. Hilaire. Bentham, in his later work (Bentham 1848) had access to a greater range of specimens collected by Gardner, Burchell, Pohl and others which enabled him to extend the number of species he recognised and to mark species boundaries with more certainty.

The work by Epling (1949) was published nearly a century after Bentham but there was still relatively little new material to work with. Although there are more species than described by Bentham (1848), few of them have many more specimen citations, but additional material made it increasingly difficult to draw boundaries between species in the *Laxiflorae*. Eventually this led to the use of the name *Hyptis macrantha* for all the red-flowered taxa (Epling & Mathias, 1957). *H. macrantha* sensu lato (i.e. the 'macrantha complex') is a very widespread taxon and occurs over much of the Brazilian savannas, or cerrado, and extends into a related vegetation in the uplands of eastern Brazil known as campo rupestre and into Paraguay and Bolivia. Cerrado is one of the most widespread vegetation types in Brazil and formerly covered two million square kilometres of central Brazil (Ratter et al. 1996). *H. macrantha* sensu lato is very characteristic of cerrado and the previous state of its taxonomy meant that it was impossible to assess the range of variation in *Hypenia* across the cerrado region and much diversity has been obscured by taxonomic uncertainty.

Since the late 1950s there has been a considerable increase in the number of collections available from Central Brazil, notably those made by the expeditions of Irwin et al. in the early 1970s. The building of Brasília in the 1950s, in the heart of the cerrado region, has also led to a major influx of collections from an area which is an important centre of diversity for *Hypenia.* With the specialist interest of Harley and publication of revised generic limits in the Hyptidinae it has become possible to attempt a revision of *Hypenia* based on greatly augmented collections and knowledge of the group.

xii) Taxonomic problems investigated in *Hypenia*

1. The relationship between *Hypenia* and the other genera of the Hyptidinae.
2. Infra-generic classification of *Hypenia.*
3. Species delimitation in *Hypenia,* particularly in the 'macrantha complex'.

The relationship between *Hypenia* and the other genera of the Hyptidinae is clearly important in assessing the status of *Hypenia* and its infra-generic classification. In particular, the taxonomic confusion between *Hypenia* and *Eriope*, together with many morphological and cytological similarities, indicated a close relationship between these two genera. The morphology of *Hypenia* and its relatives in the Hyptidinae are discussed in chapter 2 and cytology in chapter 3. Observations on pollination biology were helpful in clarifying the significance of the morphology of the flower and inflorescence in *Hypenia* and the results are presented in appendix II.

To aid the construction of a classification a cladistic analysis was conducted. The theory of cladistics is discussed in chapter 4 and the morphological analysis of *Hypenia* is presented in chapter 5. This is followed in chapter 6 by a molecular analysis based on sequencing of the ITS region. The data sets from the morphological and molecular analysise were combined in chapter 7. Chapter 8 considers the relationship between classification and phylogeny and includes an outline of my sectional and generic classification of *Hypenia.*

Species concepts are discussed in chapter 9 and the chapter also includes a discussion of species delimitation in the 'macrantha complex'. Chapter 10 presents the formal species accounts, keys and maps. Finally, evolutionary patterns in *Hypenia* are discussed in relation to geographical distributions in chapter 11.

Chapter 2: Morphological variation in *Hypenia*

A. Introduction

There is considerable morphological diversity within *Hypenia* and an assessment of morphological variation is essential to understanding the taxonomy and evolutionary relationships of the genus. The chapter is divided into two sections. The first is concerned with vegetative characters. The second part of the chapter is concerned with reproductive characters, particularly those associated with the inflorescence. The inflorescence in the Hyptidinae is very variable and an understanding of the nature of variation within it is essential to an understanding of character evolution in the Hyptidinae.

Morphological diversity in the Hyptidinae, particularly *Eriope,* is discussed. There is a summary of observed variation at the end of the chapter applied to all morphological characters in *Hypenia* which is used as the basis for the selection of characters for the morphological cladistic analysis in chapter 5.

B. Vegetative characters

i) Habit

Habit in the Hyptidinae is very variable. Several species of *Hyptidendron,* e.g. *H. canum* (Pohl ex Benth.) Harley are large shrubs or trees with perennial above-ground woody parts. *Hyptidendron arboreum* (Benth. in DC.) Harley is a tree of forest margins and can reach 20 metres. Most species of *Hyptis* are small shrubs with perennial, woody, above-ground parts although several species, e.g. those in section *Gymneia* Benth. and *Raphiodon echinus* (Nees & Mart.) Schau. are small herbs.

All *Hypenia* species are geoxylic suffrutices. This growth habit is characterised by one to ca. 30 short-lived, erect stems rising from a persistent, woody rootstock known as the xylopodium. A more extreme form of this growth habit was described, rather aptly as 'trees which live underground', by White (1976) from savannas of Africa. In these species the underground rootstock is massively developed and the many shoots observed above the ground are apparently distinct from each other. A number of cerrado species, belonging to genera which are otherwise exclusively trees, have adopted the geoxylic habit, e.g. *Cochlospermum regium* (Schrank) Pilger, a member of the Cochlospermaceae. Several, such as *Caryocar brasiliense* Cambess. (Caryocaraceae) and *Kielmeyera rosea* Mart. (Clusiaceae),

are plastic in their habit and infra-specific variation encompasses both trees and geoxylic suffrutices. The growth habit is presumably an adaptation for surviving periodic fires.

In *Hypenia* the xylopodium is small and stems tend to be few, often persisting for more than one year. The geoxylic habit is also found in *Eriope* (e.g. *E. angustifolia,* plate IVa) but large shrubs or small trees with permanent above ground woody parts are equally common in the genus (e.g. *E. hypoleuca,* plate IVb). In most species of *Hypenia* long, virgate stems bearing large, terminal inflorescences rise from the xylopodium. Some species are more shrubby and intricately branched, notably *H. irregularis* and some forms of *H. brachystachys*. *H. brachystachys* is very variable in habit and populations around Distrito Federal have very long, unbranched stems whereas populations observed in northwestern Goiás around Niquelândia, were intricately branched shrubs with short inflorescence branches.

ii) Stems

In most members of the Hyptidinae, apart from many species of *Hypenia* and *Eriope,* there is no differentiation of stem morphology between the vegetative and flowering parts. One exception is *Hyptis subtilis* Epling in section *Umbellatae* (Epling) Epling which has glabrous upper stems. Most species of *Hyptis* and *Hyptidendron* have more or less four-angled stems.

In *Hypenia* and many geoxylic members of *Eriope* there is a clear separation between the vegetative lower stem and the flowering upper parts, reflected in anatomy (Rudall 1979) and the character of the indumentum. Stems are always erect and woody at the base and usually have the long setose trichomes associated with the 'greasy-pole syndrome' (see below), shorter simple trichomes and variously glandular trichomes and sessile glands on the lower parts.

The upper part of the stem is very variable within *Hypenia.* It is usually rounded and terete but is sometimes angled (in *H. irregularis* and *H. micrantha*). Often the internodes are hollow, i.e. fistulose, and may also be swollen. These swellings are formed during normal development of the stem (Rudall 1979). The internode swellings may be elongate, e.g. *H. densiflora* (plate IIIc) or globose, e.g. *H. brachystachys* (plate IIId). The shape is usually constant within species, although *H. reticulata* is variable for this character. Sometimes the entire internode is swollen (e.g. *H. calycina* and *H. macrantha*). *H. vitifolia* has very large swellings which are irregular in shape and easily compressed. Most species of *Eriope* lack

fistulose, swollen stems but *E. hypenioides* Mart. ex Benth. has elongate swellings, similar to those found in *H. densiflora,* and *E. tumidicaulis* Harley has conspicuous globose swellings.

Most species of *Hypenia* have glabrous upper parts of the stem, usually with a plate-like waxy coating (Rudall 1979) and often with an indumentum of short simple and glandular trichomes on the upper inflorescence branches.

'Greasy-pole syndrome'

All members of *Hypenia* show at least some characters associated with the 'greasy-pole syndrome' (Harley 1991) and in some species, e.g. *H. vitifolia,* they are conspicuously developed (see figure 2-1). These characters are: long setose trichomes at the base of the stem; slender, virgate stems which sway in the wind; and waxy stems with well-developed swellings. All these characters supposedly combine to form a deterrent to ants reaching the inflorescence where they may otherwise trigger the explosive pollination mechanism of the flower without effecting pollination (Harley 1991).

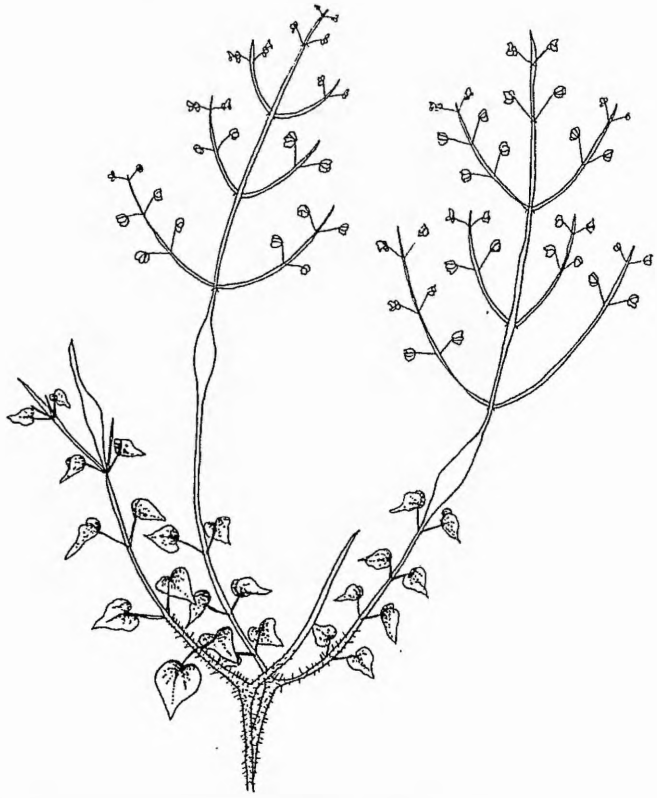


Figure 2-1: The 'greasy-pole syndrome' in *H. vitifolia*

Long, virgate stems with setose trichomes at the base, waxy coating and swellings.

iii) Leaves

Leaf morphology in the Hyptidinae is variable at the species level. Most species have lanceolate or ovate leaves but deeply dissected, almost pinnate, leaves are found in *Hyptis tagetifolia* Harley. The basal leaves in *Hypenia* are commonly lanceolate or ovate with a regular serrate or crenate margin. The apex is usually truncate or cordate. Most species are constant in leaf size and shape but *H. macrosiphon* and *H. vitifolia* sometimes have lobed leaves of variable shape and size with dentate margins.

H. brachystachys, H. irregularis and *H. concinna* have small, more or less sessile (petiole < 2mm long), ovate leaves, usually with a crenate margin and cordate base. *H. densiflora,* placed with the preceding species by Bentham (1848) and Epling (1936) in the *Densiflorae,* shows some similarity in its leaf morphology with species of section *Hypenia* (the 'macrantha complex'). Both *H. densiflora* and section *Hypenia* have larger, petiolate, lanceolate leaves with serrate margin and cuneate or truncate base. The petiole is well-developed in all species of section *Hypenia* except for *H. crispata* which has shortly petiolate leaves with lobed bases which clasp the stem. *H. vitifolia* has broadly ovate, usually lobed, petiolate leaves, *H. micrantha* has large, broadly ovate leaves and *H. salzmannii* has small, ovate to lanceolate leaves.

Basal leaves of most species of *Hypenia* are thickened. This is a character associated with water retention (Rudall 1980b) which is essential in the seasonally dry habitats where *Hypenia* occurs. The vegetative characteristics probably vary between the marked wet and dry seasons found in the savannas of Central Brazil. I did not visit these regions during the wet season but Harley has made a collection of *Hypenia* from Paraguay during the wet season when only the vegetative parts are present. In this sterile collection (Harley 28034, probably *H. reticulata*) the leaves are larger than those found on flowering specimens of *H. reticulata* from Paraguay which have been collected during the dry season.

Leaves in the inflorescence (= phyllomes) in *Hypenia* and *Eriope* are reduced and narrowly lanceolate, or subulate, resembling peduncle bracts rather than the basal leaves. Several species of *Hyptis* from sections *Laniflorae* Epling and *Umbellatae,* including *H. subtilis,* have similarly differentiated phyllomes. *H. micrantha* is unusual in *Hypenia* because it has phyllomes with a similar morphology to that of the basal leaves.

Leaf anatomy is of limited taxonomic value above the species level although it can be used, in conjunction with morphological evidence, to support taxonomic changes. For example the misplacement of *Hyptis fruticosa* Salzm. ex Benth. and *Hyptis cuniloides* Epling in section *Umbellaria* Benth. was indicated by leaf anatomy (Rudall 1980b). Harley (unpubl.) suggests they should be placed together in a new section (see appendix I). Variation in leaf anatomy in *Eriope* is mostly associated with adaptations to edaphic conditions, particularly xeromorphy (Rudall 1979 & 1980b). Variability is found in a number of characters including: trichome type and frequency; dorsiventral or isobilateral leaves; presence of adaxial stomata; presence of a hypodermis; number of layers of adaxial palisade mesophyll cells; occurrence of large bundles of phloem fibres at main veins; and various venation characters (Rudall 1979).

iv) Indumentum

The indumentum of most Labiatae is well-developed and the glabrous condition is a rare one in the family. In a survey of the trichomes only one tenth of genera of Labiatae were scored as glabrous (Cantino 1990). Multicellular, unbranched (uniseriate) trichomes are a feature of the Labiatae and all genera in the Verbenaceae which have multicellular trichomes were transferred by Cantino et al. (1992) into the Labiatae. The Verbenaceae are commonly glabrous or have branched, multicellular or simple, unicellular trichomes (El-Gazzar & Watson 1970, El-Gazzar 1974). An exception is *Stachytarpheta* Vahl, which has multicellular trichomes but is otherwise a typical member of the Verbenoideae. In the Labiatae glandular trichomes are very common and display a range of variation in the family.

In *Hypenia* all species have well-developed indumentum and there is a wide range of different trichome types within the group. Trichomes were classified into five types. They are described below and are illustrated in figure 2-2.

1. Setose trichomes (figure 2-2 a). Stiff, straight trichomes are very conspicuous and occur in most species of *Hypenia.* They are usually found at the base of the stem and frequently extend onto the petiole of basal leaves. Setose trichomes form an important character in the 'greasy-pole syndrome', acting as an effective barrier to ants attempting to climb the base of the stem. *H. aristulata* is unusual in having long setose trichomes on all parts of the stem and on the leaves.

2. Eglandular, simple trichomes (figure 2-2 b). Found in all *Hypenia* species and can occur on all parts of the plant.

3. Branched trichomes (figure 2-2 c). In *Hypenia* branched trichomes are only found on the lower leaf of *H. micrantha* but they are a feature of a number of other taxa in the Hyptidinae, notably members of *Hyptidendron* and *Eriope* as well as some *Hyptis* species, e.g. in section *Laniflorae.*

4. Glandular trichomes ('capitate glandular trichomes' of Cantino 1990) are conspicuously stalked glands. This type of hair is very common in *Hypenia* and is found on all species. They are very variable in size (see figure 2-2 d, e and f). In general they are too widespread on the plant to constitute well-defined characters, although populations of *H. reticulata* can be identified by the presence or absence of glandular trichomes on the peduncle. Some species have minute glandular trichomes with a tiny glandular tip barely expanded beyond the diameter of the stem below. Cantino (1990) notes the high degree of infra-generic variation in glandular trichomes throughout the Labiatae and their corresponding lack of phylogenetic utility.

5. Subsessile glands (figure 2-2 g) are large glands apparently sitting directly on the organ surface but in fact borne on foot cells sunken below the leaf surface (Abu Asab & Cantino 1987). Abu Asab and Cantino (1987) and Cantino (1990) describe a number of types of subsessile glandular trichomes based on the number of cell divisions within the gland. This has not been investigated in detail in this study but it appears that the subsessile glands in *Hypenia* are few-celled. They do not appear like scales as do the many-celled glands that are described by Abu Asab and Cantino (1987). Subsessile glands are common throughout *Hypenia* and can be found on all parts of the plant.

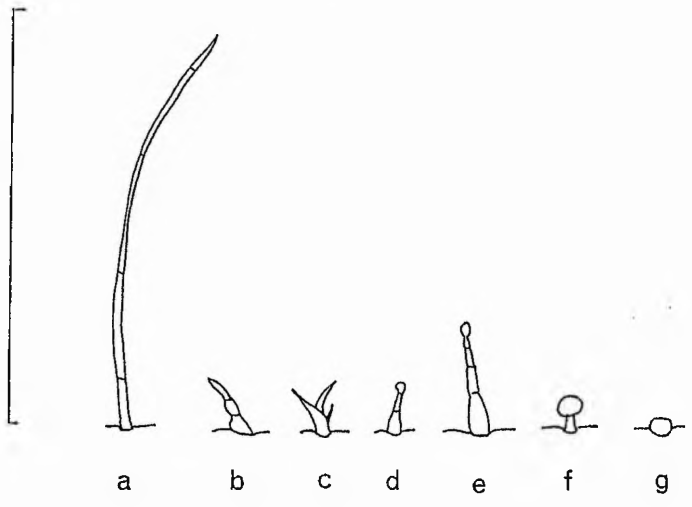


Figure 2-2: Trichomes in *Hypenia*

(see text for descriptions, scale bar = 6.5 mm)

The distribution of indumentum and the relative proportions of different hair types can provide valuable characters for species delimitation. However, such high potential for variation can be extremely confusing and the indumentum is difficult to characterise for phylogenetic studies because of the quantitative nature of the variation and the large number of potential classes.

Leaf indumentum is particularly variable. For example in *H. reticulata* some specimens have a tomentose indumentum on the lower leaf surface whereas others have sparse indumentum on both surfaces. The most characteristic feature of *H. marifolia* (Benth. in DC.) Harley was the tomentose indumentum on most parts of the plant, but continuous variation made it impossible to maintain the distinction and *H. marifolia* is now a synonym of *H. brachystachys.*

More simply defined characters can be found and the presence or absence of glabrous inflorescence branches is an important character in *Hypenia. H. aristulata, H. irregularis* and *H. micrantha* are all characterised by indumentum on the inflorescence branches. Subsessile glands on the lower leaf surface are a feature of a number of species and their distribution, i.e. concentrated around the margin of the lower surface, is characteristic of *H. gracilis.* In addition two hair types, setose trichomes and branched trichomes, can be used as simple presence / absence characters.

v) Systematic distribution of vegetative characters

Vegetative characters in the Hyptidinae are variable and not always clearly correlated with taxonomic divisions. Large woody plants are characteristic of *Hyptidendron* and some species of *Eriope.* Some species of *Eriope* also share a similar growth habit with *Hypenia,* i.e. they are geoxyilic suffrutices. Stem characters, particularly those associated witli the 'greasy-pole syndrome', are significant in *Hypenia* and *Eriope.* These characters include the possession of glabrous, waxy fistulose stems which are often swollen. The common occurrence of some 'greasy pole' characters in some species of *Eriope* and many species of *Hypenia* indicates a close relationship between the genera.

Leaf morphology has limited application at sectional or generic level and is most valuable at the species level throughout the Hyptidinae. However, the differentiation of basal and inflorescence leaves (phyllomes) is an important character to distinguish both *Hypenia* (but

see *Hypenia micrantha* and *Hypiis* sections *Laniflorae* and *Umbellatae*) and *Eriope* from the rest of the Hyptidinae.

C. Reproductive characters: inflorescence

Reproductive characters are an important source of characters for cladistic analysis and classification of *Hypenia.* In particular the inflorescence provided many characters for the cladistic analysis in chapter 5 and a detailed analysis was required to accurately establish homologies. The close relationship between *Eriope, Hyptidendron,* parts of *Hyptis* and *Hypenia* makes it essential to consider these groups to understand the variation on which to impose a taxonomic framework in *Hypenia.*

i) Inflorescence classification and terminology

The terminology used to describe inflorescences is profuse and confusing. Different authors frequently introduce new terms with varying degrees of precision or use well-known words with variable meanings. I have tried to limit the number of terms used in the circumscription of the inflorescences of the Hyptidinae and have chosen those terms which seem most applicable from current literature, resisting the temptation to coin new ones. The vocabulary employed in this study is mostly derived from previous monographers of the group (Bentham 1833, 1848, Epling 1936, 1949, Harley 1976). However, the analysis of the inflorescence that follows and the circumscription of characters for the cladistic analysis required additional precisely defined terms which could be used to establish homology and additional terms were selected from existing literature. Most of this literature owes a great debt to the work of Troll (1964 & 1969) and his follower Weberling (1989). Troll and Weberling established exactingly defined terminology, greatly expanding that established by earlier workers such as Lindley (1830) and Rickett (1944). However, Troll relied on a typological interpretation of inflorescence development which is dependent on the selection of the appropriate *Bauplan,* or structural plan, for an accurate assessment of homology and evolutionary development. For this reason his interpretation of the significance of changes in inflorescence structure has largely been rejected (Briggs & Johnson 1979). Nevertheless, the work of Troll remains widely influential, although it is frequently subject to modification. Terms used in the discussion of inflorescence morphology and in the species descriptions are defined below and the most commonly used terms are illustrated in figure 2-3.

Anthopodium: after Briggs and Johnson (1979) is defined as the internode between the flower and the ultimate node of the axis that it terminates (sometimes this internode is not elongated and can be difficult to see).

Bract: a general term usually applied to leaves subtending flowers (i.e. phyllomes). It is used specifically here to describe the phyllomes subtending the floral axis but below the bracteoles, within a cyme. It is retained in this specific sense to differentiate phyllomes within a cyme from phyllomes which subtend a cyme.

Bracteoles: this term has been used in the species descriptions presented here and has been widely used by Harley (1976) and Epling (1949). The term has been criticised because of the flexible way it has been applied by a variety of authors and more precisely defined alternatives have been proposed (Briggs & Johnson 1979). In my species descriptions and discussion of inflorescence morphology the term bracteoles refers to the paired bracts below a single flower terminating an axis. Following the precise terminology of Briggs and Johnson these phyllomes are called **metaxyphylls** (adapted from Troll's term *Zwitschenblätter*) and are defined as empty phyllomes (i.e. with no flowers in their axis) which are distal to the last non-empty phyllomes. The term bracteole is retained here because it has been used by Epling and Harley in the same sense as Briggs and Johnson's metaxyphylls.

Capitulum: following Epling's (1949) usage of the term; flowers arranged in dense heads so that the bracts subtending individual flowers are displaced to form an involucre at the base. The involucre may be obscured by the flowers in mature capitulae.

Cincinnus: after Weberling (1989); cymes with branches only at one axil, successive branches developing alternately.

Cyme: inflorescence subunit in which the axis is terminated by a flower.

Determinate axis: axis with determinate growth terminated by a flower or cyme.

Dichasium: after Weberling (1989); cymes with branches developing from both axils below the terminal flower.

Indeterminate axis: axis with indeterminate growth not terminated by flower or cyme.

Peduncle: is used in the same sense as Harley (1976) to describe the axis below the bracteoles in single-flowered cymes, i.e. the penultimate internode of the axis terminated by a flower, called by Conn (1995) the **propodium**.

Phyllome: a leaf or modified leaf (usually synonymous with bract) within the inflorescence. I have applied the term more specifically to those leaves in the inflorescence which subtend cymes or indeterminate branches.

Thyrse: following Troll (1964) and subsequent work (Weberling 1989); this term is defined as an inflorescence with cymose partial inflorescences and is *determinate* if the main axes

terminate with a flower or *indeterminate* if they lack a terminal flower. Troll recognises a series of variations based on the degree of branching and development of the partial inflorescences, e.g. pleio-thyrse (*Pleiothyrsen*), with three orders of branches and double-thyrse (*Doppelthyrsen*), with two orders of branches. The inflorescences of *Hypenia* are commonly described as paniculate (Epling 1949). However, Troll defines a panicle (*Rispe* in German) as a branched inflorescence where the axes are terminated by a flower, in which there is at least one degree of branching and there are no thyrsoid sub-units present. Using this definition the application of the term panicle is erroneous and the inflorescences of the Hyptidinae are described as indeterminate thyrses.

Verticil: a commonly used term in the Labiatae to describe opposite, many-flowered, more or less sessile cymes forming a whorl around the inflorescence axis.

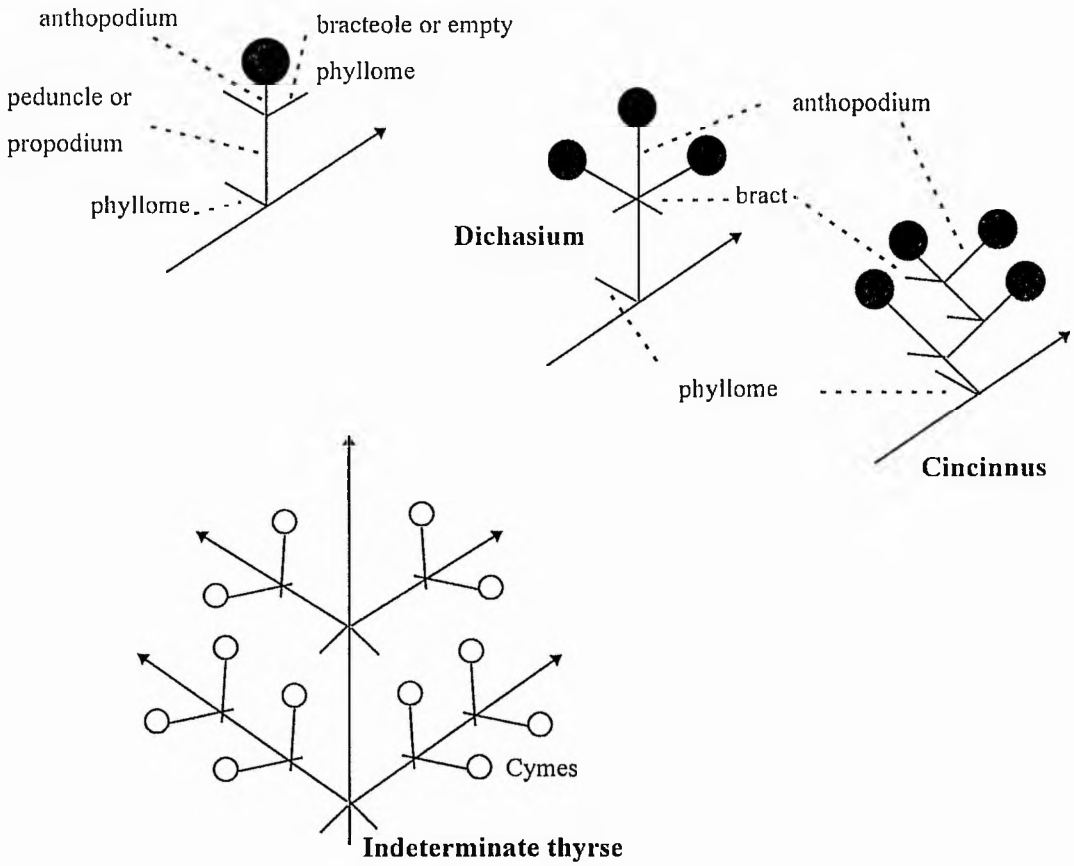


Figure 2-3: Terms used to describe the inflorescence in the Hyptidinae

ii) Inflorescence variation in the Hyptidinae

The basic inflorescence structure in all members of the Hyptidinae is the same as that found throughout the Labiatae, i.e. an indeterminate thyse following the definition of Troll (1964) where the indeterminate axes of the inflorescence bear determinate subunits (cymes). Variation is found in either the indeterminate or determinate axes. Overall patterns which can be determined in the Hyptidinae and their relatives in the Ocimeae are discussed in terms of the two axis types and then the characteristics of the inflorescence structure of *Hypenia* is presented.

Indeterminate axes

Indeterminate axes are important in determining the overall character of the inflorescence. Variation is associated with position of the inflorescence, extent of branching, differentiation between the vegetative and flowering stem morphology and development of the phyllomes subtending inflorescence branches and cymes. The inflorescence can be erect or prostrate depending on the habit of the plant and degree of woodiness.

Variation in branch development between taxa can be important, e.g. the main difference between the very dissimilar appearing inflorescences of *Eriope salviifolia* (Pohl ex Benth.) Harley (figure 2-4 f) and *E. angustifolia* Epling (plate IVa) occurs in the branching of the indeterminate axes: *E. salviifolia* has multiple short branches with closely spaced single-flowered cymes giving rise to a densely clustered inflorescence whereas *E. angustifolia* has unbranched indeterminate axes and widely spaced, single-flowered cymes borne in a simple, open raceme.

Species in the Hyptidinae that become trees or large shrubs often have multiple inflorescences and the degree of branching within a single inflorescence cannot be assessed, particularly from herbarium specimens. In *Hypenia* and some species of *Eriope* there is a single large inflorescence in which branch development is variable between individuals and which is too large to collect in its entirety making assessment of the degrees of branching difficult. For these reasons the rigorous approach taken by Troll (1964) to define different thyrse types according to the extent of branching has not been followed in this account.

The extent of development of the indeterminate axis between the cymose sub-units can produce a degree of convergence in the overall structure of the inflorescence in taxa which have divergent cyme structure, e.g. *Eriope salviifolia, Hyptis pectinata* (L.) Poit. (section

Mesosphaeria Benth.) and *Hypenia brachystachys* all have similar appearing inflorescences derived from very different cyme structures (see discussion under determinate axes).

Differentiation between the vegetative stems and the inflorescence stems is a feature of *Hypenia* and *Eriope* and is strongly associated with the 'greasy-pole syndrome' and the development of glabrous, waxy stems and the occurrence of fistulose swellings. The reduction and change in morphology of phyllomes in the inflorescence compared to the basal leaves is also most characteristic of species with 'greasy-pole syndrome' characters. In *Hypenia, H. micrantha* is the only species that has phyllomes which share the same morphology as the basal leaves. This species also lacks many 'greasy-pole syndrome' characters, notably waxy, glabrous stems. Many species of *Hyptis* have phyllomes which are reduced compared to the non-inflorescence leaves but they are rarely morphologically distinct. However, there are some exceptions including *Hyptis subtilis* and *Hyptis* section *Laniflorae* which have subulate phyllomes.

Determinate axes: cymes

Cyme structure is very diverse in the Hyptidinae. Hedge (1992) states that the evolutionary tendency in the Labiatae is the reduction of flower size and their proliferation in multi-flowered cymes. Within the Hyptidinae however, there are two tendencies in floral and inflorescence development leading in opposite directions. One developmental line leads to dense, capitulate heads where there are a large number of small flowers in each cyme following the pattern outlined by Hedge (1992). The other contrasting line is increasing flower size and the reduction of the number in each cyme to one. The distribution of cyme types in the Hyptidinae is summarised in table 2-1.

Multi-flowered cymes

Multi-flowered cymes in the Hyptidinae are very variable but they can be divided into a number of classes depending on the degree of development of various axes within the cyme. Examples of each multi-flowered cyme type are illustrated in figure 2-4. The cyme types identified are outlined below and their distribution and development in the Hyptidinae are discussed.

a) Sessile cymes: usually less than 15 flowers and the first axis and all internal axes are very short and obscured by the flowers, e.g. *Hyptis pachyphylla* Epling, section *Pachyphyllae* (Epling) Harley (figure 2-4 a).

b) Few-flowered capitulate cymes: usually less than 15 flowers, a well-developed first axis and short internal axes. The anthopodia are variously developed, e.g. species in *Hyptis* section *Laniflorae* and *Hyptis subtilis* in section *Umbellatae* have long anthopodia which are clearly visible. Many other species have short anthopodia obscured by the flowers, e.g. *Hypenia vitifolia, Hyptis fruticosa* (figure 2-4 b) and *H. suaveolens* (L.) Poit. in section *Mesospheria.*

c) Spherical capitulate cymes: usually more than 30 flowers, a well-developed first axis and all internal axes are short and obscured. The overall shape of the eyme is more or less spherical. The bracts subtending individual flowers are often filiform and are displaced in an involucre which is often obscured, e.g. *Hyptis rugosa* Benth. in section *Cyanocephalus* Pohl ex Benth. (plate IV d).

d) Hemispherical capitulate cymes: less than 30 flowers, a well-developed first axis and all internal axes are short and obscured. The overall shape of the cyme is hemispherical and the bracts are linear or oblong and are displaced to produce an involucre which is frequently conspicuous, e.g. *Hyptis lanceolata* Poit. (section *Hyptis) and Peltodon rugosus* Tolm. (figure 2.4 c).

e) Open cyme with dichasial divisions: more than 30 flowers, all the axes are well-developed and visible so the overall size of the cyme is very large, e.g. *Hyptis tafallae* Benth. in section *Umbellatae* (figure 2.4 d).

f) Cyme with cincinnate divisions: usually more than 30 flowers, the first axis is well-developed and dichasial but subsequent axes are cincinnate. In *Hyptidendron amethystoides* (Benth. in DC.) Harley in section *Umbellaria* (figure 2-4 e) the cymes are few-flowered and the branches originate from the same axil, i.e. the cyme is scorpiodal. The length of internal axes is variable, in *Hyptis asperifolia* Standl. the axes are well-developed. In *Hyptis irwinnii* Harley (section *Mesosphaeria)* the internal axes are short.

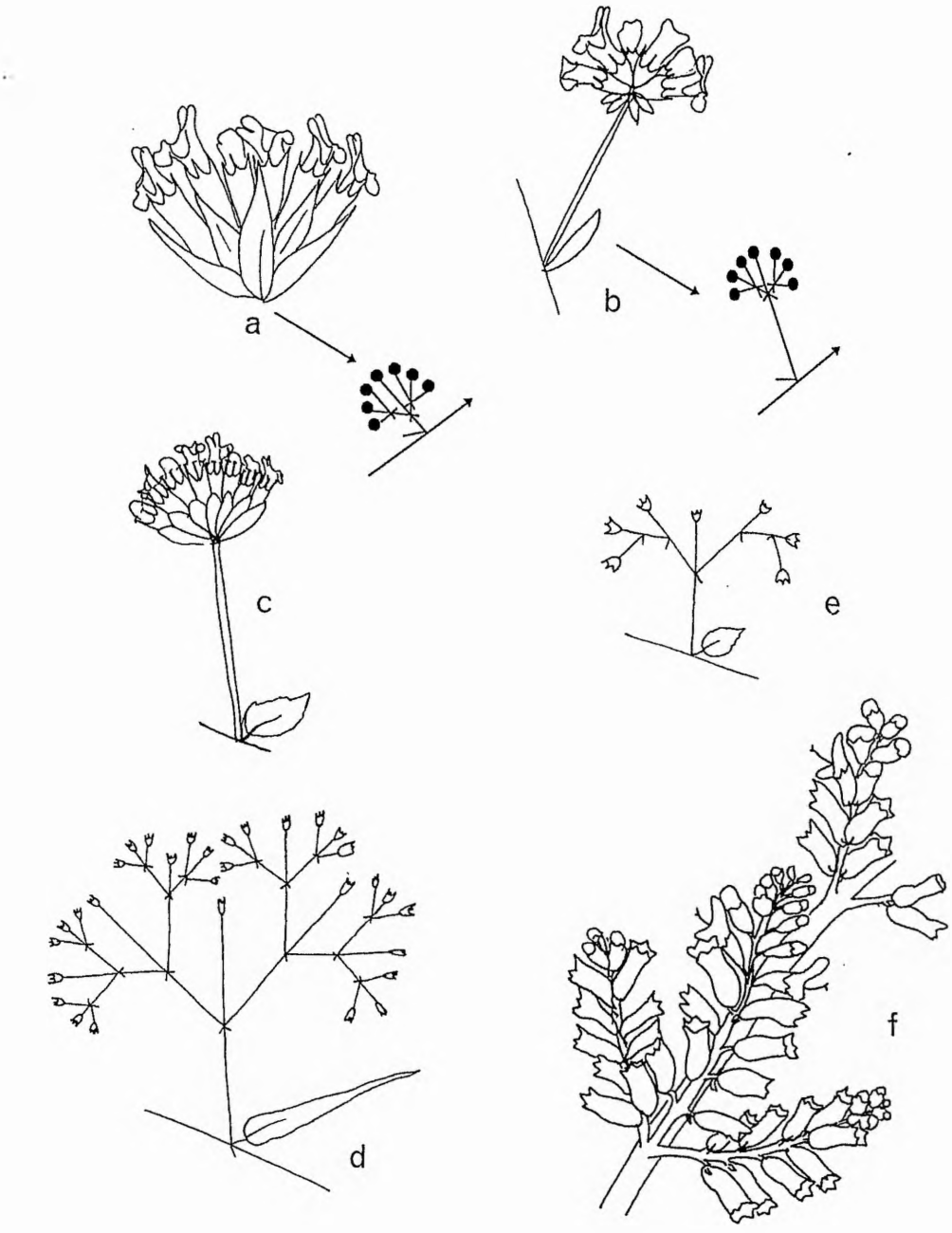


Figure 2-4: Examples of cyme types in the Hyptidinae

a) Sessile, e.g. *Hyptis pachyphylla*: b) few-flowered capitulate, e.g. *Hyptis fruticosa*: c) hemispherical capitulate, e.g. *Peltodon rugosus*: d) open dichasial, e.g. *Hyptis tafallae*: e) cincinnate, e.g. *Hyptidendron amethystoides*: f) single-flowered with bracteoles, e.g. *Eriope salviifolia.* Not drawn to scale.

Sessile, verticillate, cymes are common in the Labiatae. In the Hyptidinae they can be seen in *Hyptidendron, Hyptis* and *Asterohyptis* and can also be found in *Plectranthus* and *Isodon.* Sessile cymes are comparable to few-flowered capitulate cymes but the dichasial divisions have occurred above a reduced first axis so that the cyme is more or less sessile (e.g. *Hyptis pachyphylla* figure 2-4 a).

In *Hyptis section Umbellatae* the first axis of the cyme is well-developed and the branching in the cyme is dichasial. Variation occurs in the extent of development of internal axes in the cyme. In *H. tafallae* the axes throughout the cyme are clearly visible and branch frequently to give rise to large, umbellate cymes. In *H. tafallae* all divisions are clearly dichasial and this seems to be the predominant tendency throughout the remainder of *Hyptis,* the main variations occurring in the development of the axes and in the number of the flowers per cyme. However, there are a number of examples of species with cincinnate divisions, *Hyptis irwinnii, Hyptis* section *Muellerohyptis* Briq. and *H. eriocephala* in *Hyptis* section *Mesosphaeria* subsection *Eriocephalae* have densely-packed cymes with cincinnate branches radiating out from the first dichasial divisions. Most species of *Hyptis* with dense capitulate cymes however, as well as *Raphiodon and Peltodon,* have their flowers developing dichasially within them. It is possible that cincinnate divisions are present in these capitulate cymes. Harley (pers. comm.) has observed aberrant plants with lobed capitulae which could be a result of basal cincinnate divisions in the capitulum. Although this is not a typical morphology it does indicate the propensity with which cincinnate divisions can replace dichasial ones.

Development of the first axis of the cyme and reduction of subsequent axes is a common tendency in the Hyptidinae and *Hyptis subtilis,* also in section *Umbellatae,* shows clearly reduced internal cyme axes above a long first axis. The axes above the first axis are reduced so that the anthopodia of all the flowers in the cyme are reduced and appear to rise from more or less the same point. The same pattern can also be seen in *Hyptis* section *Laniflorae, H. fruticosa* and *Hypenia vitifolia.* This type of few-flowered capitulum is common in *Hyptis* and can be seen in *sections Polydesmia* and *Mesosphaeria,* e.g. *H. suaveolens* (plate IV c). Bracts are associated with individual floral axes and the involucre, characteristic of capitulae with densely packed flowers, is not present.

Increasing proliferation of dichasial branching and maintenance of reduced axes produces the many-flowered spherical capitulate cymes characteristic of *Hyptis* section *Cyanocephalus* and *Raphiodon.* Capitulate cymes are also seen in *Hyptis* section *Hyptis* and *Peltodon* but in these taxa the capitulum is flattened and hemispherical (e.g. *Peltodon rugosus* figure 2-4 c). In both types of cyme the floral axes are reduced and the flowers so tightly packed in the cyme that the bracts are displaced from individual flowers and form an involucre at the base of capitulum. In spherical cymes this involucre may be obscured by fully developed flowers but can usually be seen when all the flowers are in bud (see *H. rugosa,* plate IV d). In hemispherical capitulae there is frequently a conspicuous involucre of broad bracts at the base of the cyme which are similar in appearance to ray florets in Compositae inflorescences and may be functionally analogous. However, the inflorescence in the Compositae is derived from a highly contracted raceme where the axis has become contracted into a disc-like capitulum (Weberling 1989). The capitulate cymes found in the Hyptidinae are derived by proliferation of divisions within a cyme and therefore are not structurally analogous to Compositae inflorescences. *Lantana* L., in the Verbenaceae, also has hemispherical capitulae, but in common with the capitulum found in the Compositae, they are derived from a contracted raceme.

The order of proliferation of dichasial divisions can be traced in spherical cymes. In *Hyptis rugosa* (plate IV d) the first flowers to mature are those at the top of the cyme, forming the tuft of flowers on the capitulum at the left, the capitulum in the middle is more mature and has open flowers around its equator. This indicates a pattern of development where the first dichasial division gives rise to flowers at the top of the cyme and subsequent divisions are displaced downwards.

Single-flowered cymes

The alternative developmental line to proliferation of flowers in a dense head is the reduction of flower number to one per cyme. Two types of single-flowered cymes are found in the Hyptidinae.

a) Single-flowered cyme with bracteoles present: found in all species of *Eriope* (e.g. *E. salviifolia,* figure 2-4 f) and most species of *Hypenia.*

b) Single-flowered cyme with bracteoles absent: found in *Hyptis elegans* (Briq.) Briq. and *H. floribunda* Briq., both in section *Minthidium* Benth.

The process of reduction, with the production of single flowers subtended by a pair of bracteoles can be seen in all species of *Eriope* (although they are frequently caducous) and their derivation in *Hypenia* is discussed in the next section.

A different process of reduction is seen in *Hyptis elegans* and *H. floribunda* which have single-flowered cymes but lack bracteoles. Missing bracteoles suggest that there has not been a suppression of flower development below existing flowers, rather that floral development and axes have been suppressed below the anthopodium. However, there are no residual bracts to indicate this route of development and it is difficult to make conclusive statements about the production of single-flowered cymes in *H. elegans* and *H. floribunda.*

Table 2-1: Distribution of cyme types in the Hyptidinae

(After Harley's classification of the Hyptidinae, see appendix I)

Genus	Cyme type
Hyptis	Sessile, few-flowered capitulate, spherical and hemispherical capitulate, open with cincinnate and dichasial divisions, single-flowered without bracteoles.
Marsypianthes	Open with cincinnate divisions.
Hyptidendron	Sessile, open with cincinnate divisions.
Eriope	Single-flowered with bracteoles
Hypenia	Single-flowered with bracteoles (but maybe > one-flowered in *H. brachystachys,* see below), few-flowered capitulate.
Raphiodon	Spherical capitulate
Peltodon	Hemispherical capitulate
Asterohyptis	Sessile

iii) Inflorescence structure in *Hypenia*

The inflorescence in *Hypenia* species shows a range of variation from the extreme of simple, single-flowered racemes in *Hypenia* section *Hypenia,* and the few-flowered capitulate type found in *Hypenia vitifolia* and *H. longicaulis.* These extremes are also found outside the genus. Single-flowered cymes with bracteoles are typical of *Eriope* species and few-flowered capitulate cymes are common in *Hyptis.* The inflorescence in *Hypenia,* therefore is a potentially important indicator of the systematic position of the group.

Indeterminate axes

Hypenia species have large inflorescences which are often conspicuously distinct from the vegetative lower parts. Fistulose swellings and wax are often present on the stem and the inflorescence is 0.1 to 3 metres above the vegetative parts. The phyllomes throughout the inflorescence are reduced with a markedly distinct morphology from that of the vegetative leaves. They are usually subulate, although some species have broad, ovate phyllomes and bracteoles, e.g. *H. calycina.* The lowest node of the inflorescence sometimes has phyllomes which are morphologically undifferentiated, although these are frequently caducous. However, *Hypenia micrantha* has leaf-like phyllomes at all the nodes on the indeterminate axes of the inflorescence.

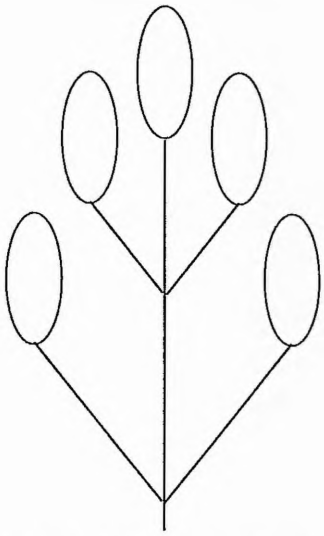
The indeterminate axes of the inflorescence are well developed in all species of *Hypenia.* In *H. brachystachys* and *H. densiflora* the cymes are often clustered on short indeterminate branches (see plate III a and b and figure 2-5) off a single, long axis. In *H. vitifolia, H. salzmannii, H. micrantha* and section *Hypenia* the branches are of more or less uniform length and the cymes (which are frequently single-flowered) are distantly spaced along the branches. If single-flowered cymes are present this gives rise to a simple, branched, racemose inflorescence. The exact spacing of the cymes can have a considerable impact on the overall appearance of a species and in *H. reticulata* it contributes much of the considerable morphological diversity in the species (see section *Hypenia,* plate II and *H. salzmanni,* plate IV e). In section *Hypenia* supernumerary branches, borne in the axils of the main branches, are common and *H. sclerophylla* and *H. caiaponiensis* have characteristic inflorescence morphologies derived from their habit of producing irregular supernumary branches.

Cymes

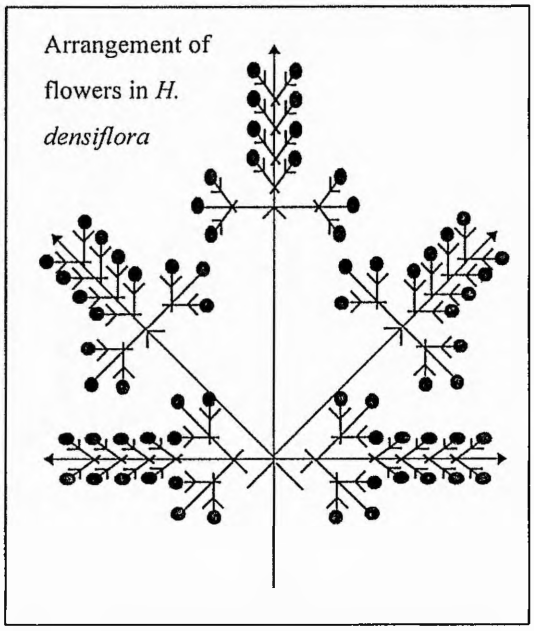
In *Hypenia* the inflorescence structure is very variable and a series can be traced from *H. vitifolia,* with several flowers per cyme to *H. micrantha* with just one flower per cyme. These one-flowered cymes are found throughout *Hypenia* section *Hypenia* (although *H. sclerophylla* and *H. crispata* sometimes have two flowers per cyme). The peduncle is usually more than 5 mm long. In *H. vitifolia* the anthopodium is very short with the flowers densely arranged in the cymes above a well developed first axis.

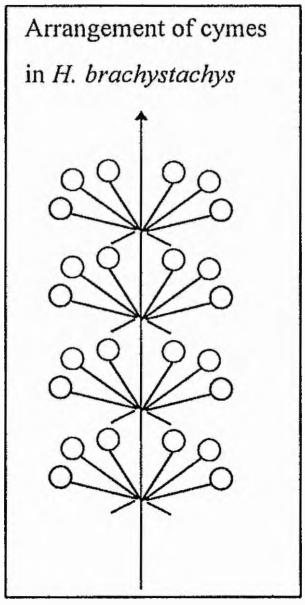
In *H. irregularis, H. densiflora* and *H. gracilis* one-flowered cymes are predominant, although dichasially branched cymes are also present. However, there is a great range of variation in the structure and arrangement of the cymes in *H. brachystachys* due to variable development of the flowers and axes. In both *H. brachystachys* and *H. densiflora*

suppression of axes may occur so that more than three branches appear to arise from one node. Figure 2-5 illustrates the arrangement of the cymes in *H. densiflora* and *H. brachystachys* and the variation found in cyme structure in *H. brachystachys*. Photographs of the inflorescence of both species are shown in plate III. *Eriope salviifolia* has a similar inflorecence to *H. densiflora* but the cymes are always single-flowered (figure 2-4 f).



Overall inflorescence structure of *H. densiflora* and *H. brachystachys*





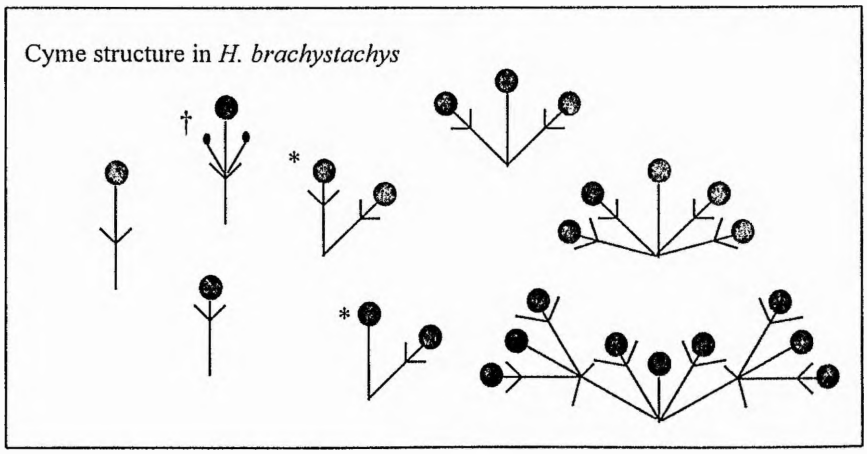


Figure 2-5 : Inflorescence structure in *H. densiflora* and *H. brachystachys*

The basic branching pattern of the cymes in all species of *Hypenia* is dichasial, although cincinni may appear in *H. brachystachys* (figure 2-5, marked *) when one branch does not develop out of the axis of the bracteoles. However, two bracteoles are always present in this case, instead of one associated with a cincinnus, and it has been interpreted as a dichasium.

Bracteoles are absent in *H. vitifolia.* They are also absent from some flowers in *H. brachystachys, H. densiflora* and three-flowered cymes in *H.salzmannii,* depending on their position in the cyme. Bracteoles are present below all flowers in the remaining species. The variable presence of bracteoles in *H. brachystachys* and *H. densiflora* helps to indicate the derivation of this character. Harley (1976) suggested that in *Hypenia* and *Eriope* they were derived by the lack of development of flowers in their axils. This occurs commonly in many taxa and their derivation has been indicated in the Myrtaceae (Briggs & Johnson 1979) and in the Loganiaceae (Conn 1995). The middle flower of the three-flowered dichasia present in *H. densiflora* and *H. brachystachys* lacks bracteoles and the anthopodium is well developed (see cyme marked † in figure 2-5). The lateral flowers however, always have bracteoles, indicating that suppression of lateral flower development has occurred and the anthopodium has become reduced. The first axis of the cyme, if present, is less than 5 mm long so that the cymes are more or less sessile. In single-flowered cymes the peduncle is also less than 5 mm long.

iv) Systematic distribution of inflorescence characters

A conspicuous tendency in *Hyptis* is the development of dense, capitulate, multi-flowered cymes borne on well-developed first axes. Cymes vary in the number of flowers and the regularity of their arrangement. Relatively few-flowered cymes are seen in *Hyptis fruticosa* and *Hyptis* sections *Laniflorae* and *Umbellatae*. The same arrangement can also be seen in *Hypenia vitifolia.* An increase in the number of flowers per cyme is associated with an increase in the regularity of their arrangement. Spherical cymes are particularly developed in *Hyptis* section *Eriocephala* and hemispherical cymes are characteristic of *Peltodon* and *Hyptis* section *Hyptis.*

Hypenia section *Hypenia, H.micrantha,H. salzmannii* and *H. irregularis* have basically the same inflorescence morphology as all species of *Eriope,* i.e., one-flowered cymes arranged on variously-branched indeterminate axes. The inflorescence is terminal and the branches are frequently morphologically distinct.

The cymes of *H. densiflora* are mostly one-flowered but multi-flowered, dichasial cymes are present on the lower parts of the indeterminate axes. *H. brachystachys* also has multi-flowered, dichasial cyines which are very variable and may be complex. Both species have more or less sessile cymes which are crowded on the indeterminate axis. More or less sessile, multi-flowered cymes are common in the Hyptidinae and in the Ocimeae in general.

D. Reproductive characters: flowers

Floral morphology in the Hyptidinae was considered to be more or less constant by Epling (1949) but detailed studies of the corolla, particularly the length and shape of the tube, are important in the taxonomy of *Hypenia.* In addition, at the generic and sectional level there is considerable taxonomic value in calyx morphology.

i) Flower orientation

In section *Hypenia* subsection *Ellipticae* the flower is upside-down, or resupinate, in comparison to the flower of all other species in the Hyptidinae. In this group the anterior lobe occurs on the upper side of the corolla and all the characters which make the flower symmetrical along one plane are inverted. Four resupinate-flowered species are illustrated in plate I.

In all the resupinate-flowered species observed in the field the bracteoles were deflexed upwards whereas the bracteoles in the non-resupinate subsection *Hypenia* were deflexed downwards. This suggests that there is a twist in the peduncle. If the twist occurred in the anthopodium the bracteoles may be expected to remained displaced downwards (see *H. macrantha,* plate I e). Observations of pollinator behaviour on *H. reticulata* (non-resupinate flowers, subsection *Hypenia*) and *H. macrantha* (resupinate flowers, subsection *Ellipticae*) suggested that *H. macrantha* was hummingbird pollinated and that *H. reticulata* was pollinated by large bees (appendix II).

The arrangement of the flowers in the Ocimeae has been discussed in relation to pollination. Most non-ocimoid members of the Labiatae have nototribic flowers, i.e. the reproductive parts are held against the posterior lobe of the corolla. The reversed condition, sternotriby is found in the Ocimeae and has been postulated as a the result of resupination of the corolla with an exchange of position between the upper and lower lips (van der Pijl 1972, Faegri & van der Pijl 1979). However, Tanaka (1972, cited in Huck 1992) suggested that sternotriby in the tribe does not result from resupination but is due to the transference of the anthers and

style from the upper to the lower part of the flower. Ocimeae flowers are described as reversed gullet flowers by Faegri & van der Pijl (1979) on the basis of the proposed resupination in the flower, although they are sternotribic.

Some species of Labiatae have resupinate flowers where all the parts, not just the stamens and style, are inverted, e.g. *Teucrium resupinatum* Balansa ex Coss. (Teucrioideae) and all members of the genus *Cyclotrichium* (Boiss.) Manden & Scheng. (Nepetoideae, tribe Mentheae). In these non-ocimoid taxa the flowers change from nototriby to sternotriby.

iii) Calyx morphology in the Hyptidinae

The calyx in the Hyptidinae is cylindrical with five sub-equal lobes. There are ten nerves in the calyx, five extend into the lobes and alternate with the other five which extend only to the margin of the calyx tube. The length of the tube varies from 2 to ca. 15 mm, and the diameter. varies relative to the length giving rise to some variation in shape i.e. from cylindrical to campanulate. Most taxa have straight calyces but some species of *Hyptis* have sigmoid calyces, e.g. *H. rugosa* Benth. in section *Cyanocephalus* Pohl ex Benth.

In all species the calyx is accrescent, i.e. enlarged, when fruiting, increasing by two to three times in length and diameter. Flowering calyces display much denser indumentum relative to the fruiting calyx which is sparsely covered in glandular and eglandular trichomes. Many of the characters associated with the calyx refer to the fully mature condition and the descriptions and illustrations that follow concentrateon fruiting calyx morphology. The calyx of species mentioned in the text is illustrated in figure 2-6.

Species of *Eriope and Eriopidion* have the most distinctive calyces in the subtribe and, although the calyx of *Eriopidion strictum* is unique (see below), it shows clear links with *Eriope* section *Eriope* (see classification in appendix I). In all species of *Eriope* the three posterior lobes are slightly connate at the base making the calyx zygomorphic. This condition can also be found to a lesser extent in *Hyptidendron* section *Hyptidendron* (e.g. *H. canum* figure 2-6 k) and some species of *Hypenia* (e.g. *H. brachystachys* figure 2-6 c). Members of *Eriope* section *Eriope* have a distinctive calyx morphology with very short posterior lobes which are connate at the base and reflexed to a vertical position at the mouth of the campanulate calyx tube (e.g. *E. hypenioides* figure 2-6 j). *Eriopidion strictum* has a similar morphology but the middle of the three connate lobes on the upper side is enlarged and the

upper lip formed by these three lobes closes over the mouth of the calyx when it is dry, becoming reflexed when wet due to a hygroscopic mechanism (Harley 1976).

Most species of *Hyptis,* e.g. section *Hyptis* plus *Hypenia, Eriope, Hyptidendron* and *Marsypianthes* have deltoid lobes where the membrane extends down in a triangle from just below the tip of the medial nerve of the lobe, meeting the calyx tube at the intermediate nerves. In several sections of *Hyptis,* e.g. *Cyanocephalus,* the membrane is reduced and the lobes apparently consist of the central nerve only. In the genus *Peltodon* (e.g. *P. rugosus,* figure 2-6 m), which is distinguished from *Hyptis* by its seemingly peltate, but actually spoon-shaped, or cochleariform, calyx lobes, there is a very narrow marginal membrane which extends in parallel to the nerve until its tip where it expands into a spoon shape. *Raphiodon echinus* (Nees & Mart.) Schau. has a calyx that is unique in the subtribe (figure 2-6 n). The calyx has spines between the lobes so that it has ten lobes instead of the five elsewhere in the Hyptidinae. The spines make the cyme into an easily transported burr-like mass which is abscissed as a single unit. They are presumably a dispersal adaptation associated with the habit of shedding the entire cymose sub-unit as a single unit and may account for the relatively widespread distribution of this species especially along roadsides.

In most members of the subtribe the lobes are straight, but *Hyptidendron* section *Hyptidendron*
species (e.g. *H. canum* figure 2-6 k) have spreading calyx lobes as do members of section *Cyanocephalus.*

Many species of *Hyptis,* some populations of *H. reticulata,* all members of *Eriope* section *Eriope* and *Eriopidion strictum* have an annulus of hairs in the calyx tube which can be dense enough to obscure the nutlets and prevent their dispersal until the calyx is subject to some physical force. The hairs may also act as a barrier to water entering the calyx and triggering myxocarpy in the nutlets. Myxocarpic nutlets covered in the sticky mucus produced after wetting are very difficult to dislodge from the calyx. However, most species of *Eriope* and *Hypenia* have the mouth of the calyx pointing downwards in fruit and are not vulnerable to wetting from rain. They also produce mature nutlets at the driest time of year.

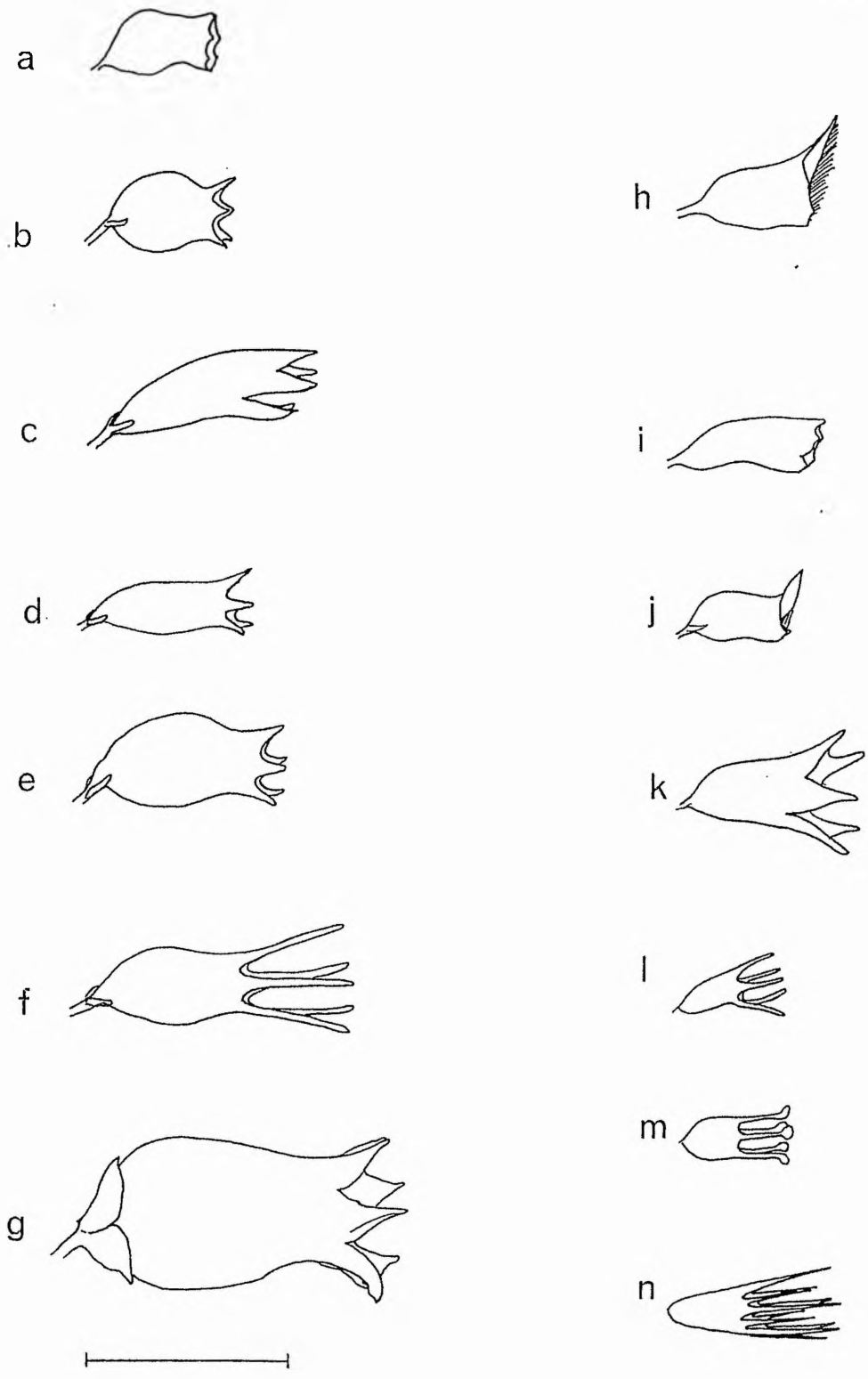


Figure 2-6: Calyx morphology in *Hypenia* and the Hyptidinae

a) *Hypenia vitifolia*; b) *H. salzmannii*; c) *H. brachystachys*; d) *H.irregularis*; e) *H. reticulata*; f) *H. aristulata*; g) *H. calycina*; h) *Eriope hypenioides*; i) *E. salviifolia*; j) *Eriopidion strictum*; k) *Hyptidendron canum*; l) *Hyptis rugosa*; m) *Peltodon rugosus*; n) *Raphiodon echinus*. All illustrations are lateral views of fruiting calyces. Scale bar = 10 mm.

iii) Calyx morphology in *Hypenia*

Hypenia vitifolia* and *H. longicaulis

The calyx is more or less actinomorphic, shortly cylindrical or campanulate in flower and cylindrical in fruit with very short (< / = 1 mm in flower and fruit), equal teeth (figure 2-6 a). The fruiting calyx expands from 2 or 3 mm in flower to 6 or 7 mm and has a distinctly bulbous base. There is slight zygomorphy evident in the calyx in fruit with the calyx lobes arranged as three posterior over two anterior. The bilateral shape is mostly conferred by the bulbous base extending on one side more than the other. The indumentum on the calyx of *H. vitifolia* consists of simple, egiandular trichomes and subsessile glands. The throat is glabrous. The fruiting calyx texture in *H. vitifolia* is somewhat leathery. The calyx of *H. longicaulis* is very similar to that of *H vitifolia* but is generally less bulbous at the base. The population of *H. vitifolia* at Morro do Chapéu in Bahia lacks bulbous calyces and this is not a reliable character to distinguish the two species.

H. salzmannii* and *H. micrantha

The morphology of the calyx in *H. salzmannii* (figure 2-6 b) and *H. micrantha* is similar to that of *H. vitifolia,* but the teeth are more developed. In the fruiting calyx of *H. salzmannii* there is considerable enlargement at the base and the calyx may be almost spherical below a constriction at the throat. The throat in both species is more or less glabrous in fruit. Indumentum consists of sparse eglandular trichomes and subsessile glands. The fruiting calyx of both *H. salzmannii* and *H. micrantha* is rather papery in texture and is generally displaced so that the month points downwards.

H. densiflora, H. irregularis* and *H. brachystachys

Both *H. densiflora* and *H. brachystachys* (figure 2-6 c) have similar calyces which are cylindrical and somewhat sigmoid. The three posterior lobes are connate at the base, conferring distinct zygomorphy to the flowering and fruiting calyces of these species. The sigmoid calyx shape is possibly the result of packing constraints in the dense inflorescence. Flowers from individuals of *H. brachystachys* with relatively lax inflorescences have straighter calyces. *H. irregularis* (figure 2-6 d) has a straight, cylindrical calyx with symmetric lobes. The flowering calyx in all three species is 4 to 8 mm long and the corolla is exserted less than one third its length. Enlargement of the mature calyx involves an increase of about one and a half times the flowering length. Indumentum consists of simple, eglandular trichomes and subsessile glands. The calyx of species placed by Harley in *Eriope* section *Nudicalyx* Harley sect. nov. (see appendix I) have long, cylindrical calyx tubes,

slightly connate posterior lobes and a naked throat (e.g. *E. salviifolia,* figure 2-6 i) and are similar to those of *H. brachystachys* and *H. densiflora.*

Section *Hypenia* (the 'macrantha complex')

Species in section *Hypenia* have cylindrical or campanulate calyces which are often slightly gibbous and more or less symmetric with deltoid lobes. The lobes may be slightly rostrate towards the apex and in *H. aristulata* (figure 2-6 f) the lobes are conspicuously long with a very narrow membranous margin to the central nerve. Conversely, in *H. calycina* (figure 2-6 g) the lobes are broad and sometimes slightly winged. In all species in the section the fruiting calyx increases in size by about one and a half to two times, from its length in flower of 3.5 to 14 mm.

iv) Corolla morphology in the Hyptidinae

The morphology of the anterior corolla lobe is an important character in the Ocimeae and helps to delimit the subtribes (see chapter 1). In the Hyptidinae the boat-shaped anterior lobe is laterally flattened and the upper margins are held tightly together by interlocking fimbriate projections. After a mechanical stimulus the lobe rapidly reflexes, hinging on a thickened portion of tissue at its base (see figure 2-7). This morphology is shared by all members of the Hyptidinae, other than *Asterohyptis.* The morphology of the corolla has been correlated with pollination mechanisms in *Eriope* (Harley 1971) and in *Hyptis* (Burkhart 1939) and the explosive mechanism of the anterior lobe flicks pollen onto the underside of visitors. The insects reported visitng *Eriope* were short-tongued bees (Harley 1971). An apparently independently derived explosive mechanism in the Ocimeae has been reported in *Aeollanthus* Mart. ex Spreng. (Hedge 1972) but only the lower two stamens are held under tension by the anterior lobe. *Plectranthus vestitus* Benth. retracts its anterior lobe (Nilsson et al. 1985) as does *Plectranthus inflexus* Vahl ex Benth. (Huck 1992). In both these species the anterior lobe is not retracted in response to mechanical pressure exerted by visitors to the flower but automatically occurs after anthesis. Such mechanisms indicate a relatively high degree of specialisation of floral biology in the Ocimeae.

Within the Hyptidinae the corolla is relatively uniform in shape and consists of a tube with two posterior lobes, two lateral lobes and one anterior lobe. The posterior lobes are usually held erect and the lateral lobes are either held at 90° to the corolla mouth or point forwards. Although relatively invariable in shape, the corolla in the Hyptidinae varies greatly in size. The tube can be from ca. 3 mm long in *Hyptis* section *Minthidium* (e.g. *H. elegans* figure 2-8 k) to 26 mm in *Hypenia* (*H. macrantha*). Shape varies less than size and variation is

generally rather subtle but can have a profound effect on pollination (see appendix II). *Hyptis* species usually have cylindrical corolla tubes like those of most members of *Hypenia,* whereas *Hyptidendron vepretorum* (Mart. ex Benth.) Harley (figure 2-8 j), *Eriope, Hypenia vitifolia* (figure 2-8 a) and *H. micrantha* have flared corolla tubes. However, the shape of the corolla differs between these species: that of *Hyptidendron vepretorumi* and *Hypenia vitifolia* and *H.micrantha* flares from the base in a uniform funnel shape but that of *Eriope* opens out suddenly above a constriction over the nutlets (e.g. *E. hypenioides* figure 2-8 g). This constriction can also be seen in the cylindrically-tubed species of *Hypenia* (e.g. *H. reticulata* see plate I b). The broad mouth to the funnel-shaped corolla seems to be an adaptation to pollination by short-tongued insects, particularly bees. *Raphiodon echinus* (figure 2-8 l) has a cylindrical corolla tube with a narrow mouth which tapers in the lower half of the tube to a long constricted tube. This tube can only be entered by long-tongued insects such as butterflies, which I observed frequently visiting this species. Similar narrow corolla tubes can be seen in *Hyptis* section *Cyanocecphalus* (e.g. *H. rugosus* figure 2-8 j) and in *Hyptis* section *Trichosphaeria* Benth. but in these species the corolla tube and mouth is even narrower. Harley (pers. comm.) has frequently observed butterflies visiting the slightly perfumed flowers (unusual in the Labiatae) of species of *Hyptis* section *Trichosphaeria.*

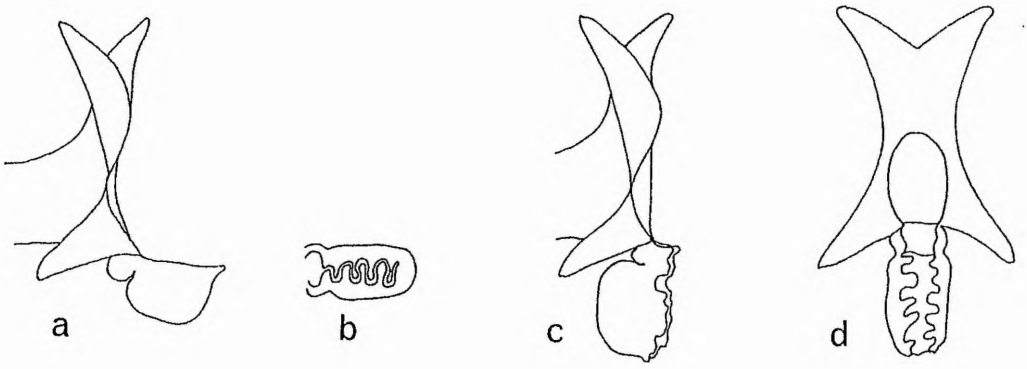


Figure 2-7: Anterior corolla lobe morphology in the Hyptidinae

a) Corolla lobes prior to triggering, lateral view; b) anterior lobe prior to triggering, view from above; c) Corolla after triggering, lateral view; d) Corolla after triggering, view from front.

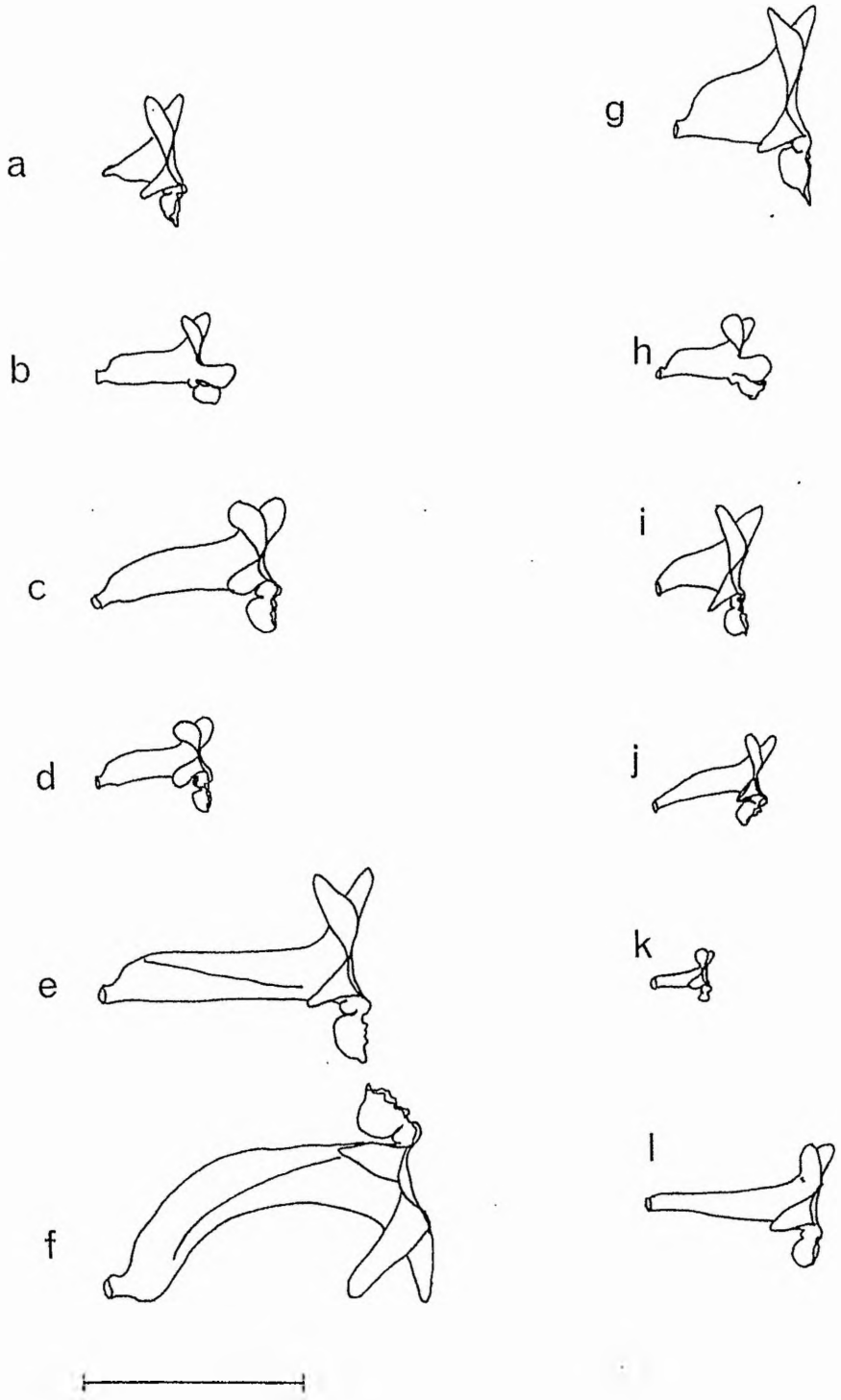


Figure 2-8: Corolla morphology in *Hypenia* and the Hyptidinae

a) *Hypenia vitifolia*; b) *H. salzmannii*; c) *H. brachystachys*; d) *H. irregularis*; e) *H.reticulata*; f) *H. calycina*; g) *Eriope hypenioides*; h) *Eriopidion strictum*; i) *Hyptidendron vepretorum*; j) *Hyptis rugosa*; k) *Hyptis elegans*; l) *Raphiodon echinus*. All illustrations are lateral views. Scale bar = 10 mm.

v) Corolla morphology in *Hypenia*

Within *Hypenia* there is considerable diversity in respect to pollinators exploited by members of the genus and much of this diversity can be correlated with changes in corolla morphology as well as inflorescence structure and the orientation of the flower.

Hypenia vitifolia, H. salzmannii and *H. micrantha*

Colour

Corollas are blue in *H. salzmannii* and lilac in *H. vitifolia* and *H. micrantha* (see plate V for corolla morphology and colour in *H. salmannziii,, H. vitifolia* and *Hyptidendron vepretorum*). In *H. vitifolia* the throat of the corolla tube is pale and has dark purple lines on the interior of the posterior lobes leading into the throat. *H. salzmannii* has a white throat to the corolla tube. In both *H. salzmannii* and *H. vitifolia* there is a striking change in colour between the bud stage and the flower at anthesis. Buds of *H. salzmannii* are primrose yellow and change to sky blue when the lobes open out. Buds of *H. vitifolia* are brown and change to lilac as the flower matures. I also observed a change from yellow or brown to lilac / blue / pink as the flowers opened in the corollas of *Eriope latifolia* Mart. ex Benth. and *E. parvifolia* Mart. ex Benth..

Form

H. vitifolia (figure 2-8 a) has a corolla tube which is very similar in shape to those of species of *Hyptidendron* section *Umbellaria* (e.g. *Hyptidendron vepretorum*). The corolla is funnel-shaped, flaring abruptly from a very narrow base to a broad mouth in the 3 to 5 mm length of the tube. The posterior and lateral lobes are slightly reflexed back from the mouth and the anterior lobe projects forward until it is triggered, after which it reflexes 90°. This form of corolla is also shared by *H. micrantha. H. salzmannii* (figure 2-8 b) has a cylindrical corolla tube and forward pointing lateral lobes which are very similar to the corolla of *Eriopidion strictum.* The anterior lobe of *H. salzmannii* is relatively small.

H. brachystachys, H. densiflora, H. irregularis and *H. gracilis*

Colour

These four species have cream or lilac corollas, often with some marking around the mouth of the tube, either in a contrasting colour or as spots. No colour change has been observed between different stages of floral development.

Form

H. densiflora and *H. brachystachys* (figure 2-8 c) have corolla tubes 6 to 12 mm long which are usually slightly sigmoid. *H. irregularis* (figure 2-8 d) and *H. gracilis* have a less crowded inflorescence and the corolla tube is straight and smaller, 4 to 6 mm long. The lobes of all four species are small, rounded at their tips and held at ca. 90° to the mouth of the corolla tube. In common with species of *Eriope,* there is a constriction in the corolla around the ovary.

Section *Hypenia* (the 'macrantha complex)

Colour

Most species have red corollas, although *H. subrosea* has a white corolla with lilac spots on the front face of the lobes and the corolla of *H. crispata* is yellow (see plate I d). However, red is used in rather a broad sense on many herbarium labels and field observations indicate that there is considerable variation in colour, often within species. Different populations of *H. aristulata* and *H. reticulata* (at Serra do Cipó, Minas Gerais) varied in colour from scarlet to purplish-red, probably as a result of different degrees of yellow pigmentation in the corolla at different ages of the flower.

In subsection *Hypenia* the hinge of the corolla, the point at which the anterior lobe retracts from the tube of the corolla on its thickened base, lacks red pigment and provides a contrast with the colour of the rest of the corolla. The colour of the hinge depends on the levels of yellow pigments in the flower. In scarlet / orange flowers the hinge is yellow indicating high levels of yellow pigment. In purplish-pink flowers the hinge is cream.

H.reticulata flowers show progressive changes in colour as the flower, matures (see appendix II). In bud the corolla is red but after triggering of the anterior lobe it changes to pink or apricot, depending on the amount of yellow pigments present. *H. calycina* is very variable in corolla colour and flowers range from shades of apricot and salmon pink to red. The corollas of *H. macrantha* stay a uniform scarlet throughout their life.

Form

Within the section as a whole there is considerable variation in size of the corolla but the shape is relatively uniform. The lateral and posterior lobes are ca. one quarter to one third the length of the corolla tube, held at 90° to the corolla mouth and variously pointed or

rounded, e.g. *H. reticulata* (figure 2-8 e). In *H. aristulata* the lateral lobes are sometimes slightly, twisted.

The corolla tube in subsection *Hypenia* varies between 8 and 18 mm and in subsection *Ellipticae* from 15 to 26 mm. The flowers of subsection *Ellipticae* are amongst the largest in the Ocimeae, certainly the largest in the Hyptidinae. The tube is usually straight, although in *H. calycina* there is a marked curve along its length (see figure 2-8 f and plate I f).

Species in section *Hypenia* have an inward growth along the length of opposite sides of the tube and crossing it diagonally from the anterior side of the mouth of the throat to the posterior side at the base of the corolla (see plate I b). These projections effectively cut the interior of the corolla tube into two halves and direct inserted objects, such as a hummingbird's beak or the tongue of large bees to the base of the corolla but away from the gynoecium. They presumably act to protect the ovules from probing beaks and probosces. Lateral projections in the corolla were also observed by Brantjes and de Vos (1981) in *H. pauliana* (Epling) Harley, a synonym of *H. reticulata.* These projections can be seen in some species of *Plectranthus* and it is probable that they represent the posterior staminal traces (Paton pers. comm.).

vi) Pollination syndromes in *Hypenia*

Much of the diversity of floral structure in *Hypenia* can be related to functional aspects of pollination and preliminary field studies were conducted to investigate the relationship between floral biology and morphology. In addition observations on phenology and fruit set in some species of *Hypenia* were made. The results are presented in appendix II and briefly discussed below.

The Labiatae is considered to be a largely xenogamous family although there are no reports of self-incompatibility within the family (Owens & Ubera-Jiménez 1992). The family has many intricate mechanisms associated with the promotion of outcrossing including protandry, gynodioecy and mechanical aspects of floral morphology (Huck 1992).

Entomophily is common in the Labiatae with many reports of bee pollination (Huck 1992). In addition pollination by flies, butterflies, hawkmoths as well as hummingbirds (Colibridae) in the New World and sun birds (Nectarinidae) in the Old World, are also reported for the family (Meeuse 1992). Bees are the most commonly reported visitors to the Ocimeae (Huck

1992) but bird pollination has been reported in *Hyptis pauliana* (now *Hypenia reticulata*) by Brantjes & de Vos 1981).

Several aspects of floral morphology in *Hypenia* can be correlated with different pollination syndromes. Flower size is obviously important, the small corolla of *H. vitifolia* and *H. salzmannii* can only produce a small reward and provides a suitable landing platform or hovering target for small insects. The funnel-shaped corolla shape of *H. vitifolia* is very similar to that found in *Eriope crassipes* which Harley (1971) observed being visited by short-tongued bees. The larger corolla of *H. densiflora, H. brachystachys* and *H. reticulata* is accessible to larger insects with mouthparts and a tongue able to reach into a corolla tube up to 13 mm long. The xylocopid bees which visited these species have mouthparts over 10 mm long are easily able to reach nectar from these flowers. The corolla tube of *H. macrantha* is too long for bees and nectar is only accessible to hummingbirds such as *Amazilia fimbriata.*

It was previously thought that all species in section *Hypenia* were bird pollinated because of their relatively large, tubular, red corollas (Brantjes and de Vos 1981). My observations suggest that there is more variation in the group and that *H. reticulata* is more likely to be bee pollinated whereas *H. macrantha* is indeed pollinated by hummingbirds. Bird pollination seems to be associated with corolla inversion and increased tube length. In addition the morphology of the inflorescence in *H. macrantha,* where the flowers are held at the ends of the branches, allows for easy access by hovering hummingbirds. The sternotribic flowers found in subsection *Hypenia* provide a more suitable base for bees which need to hold on to the mouth of the corolla. The colour change seen in the maturing corollas of *H. reticulata* may be associated with increased attraction to bees. The red pigments are lost through the life of the flower which may enhance their visibility to bees. Older, triggered flowers, which tend to be pink, may act as the initial attractant. Once the bee is at the plant it moves between flowers on the same plant, visiting flowers at all stages of maturation, including red, untriggered flowers.

vii) Androecium

The androecium of the Hyptidinae consists of two pairs of stamens exserted out of the corolla tube in a declinate position. The anterior stamens are inserted on the corolla tube between the posterior. The anthers are yellow or brown, dorsifixed, bilocular and open by longitudinal slits. In common with all the Ocimeae the anthers are synthecous. The

filaments of all species of *Hypenia* have long, white hairs which are much denser on the posterior pair of stamens.

Pollen

Pollen can be an important source of characters in the Labiatae and Erdtman (1945) proposed the division of the family into two subfamilies based on pollen types (see chapter 1). Rudall (1980a) made a preliminary investigation of pollen in the Hyptidinae as part of a survey of anatomical and palynological variation in the subtribe. Her investigation represented all the genera of the Hyptidinae and all the sections of *Hyptis* and so included *Hypenia* and *Hyptidendron* although these genera were not recognised at the time of her study.

Pollen in the Hyptidinae shows some variation and Rudall (1980a) recognised three classes of pollen type based oil size and shape of the lumina, reticulation and overall size of the pollen grain. Type A pollen, with large, polygonal lumina, numerous, small punctae, thin ridges of reticulum and large size is found in *Eriope, Hyptidendron* section *Umbellaria, Hypenia* and some species of *Hyptis.* Type B is intermediate between A and C and includes *Hyptis fruticosa, Hyptidendron canum* and some other species of *Hyptis.* Type C pollen has small, rounded lumina, few, large punctae, very thick primary reticulum and small grain size and is found in many species of capitulate-cymed *Hyptis.* Pollen therefore provides valuable evidence for evolutionary trends in the Hyptidinae but the continuous nature of tile variation restricts the value of palynological characters investigated. M. M. Harley has investigated pollen in the Ocimeae (1992a and M. M. Harley et al. 1992b) and has found a large numbers of characters (35) although many of them display continuous variation or are subject to variable interpretation, particularly shape. It appears that pollen can be useful at lower taxonomic levels in the Ocimeae and further work on pollen in the Hyptidinae may yield more information for a detailed study of the subtribe.

viii) Gynoecium

Variation in gynoecial morphology is restricted in the Hyptidinae but there are some valuable characters to be found. In the Hyptidinae the ovary is held on a short, lobed gynophore with the lobes alternate to the nutlets. The anterior lobe is larger than the others and functions as a nectary. At the tribal level the linear morphology of the embryo sac found in the Ocimeae is a possible autapomorphy for the tribe (Rudall & Clark 1992).

The stylopodium and its distribution in the Hyptidinae

All species of the Labiatae have an abscission point at the base of the style (Owens & Ubera-Jiménez 1992) and in the Hyptidinae this sometimes occurs above the nutlets so that a persistent stump projects above them (Harley 1973, 1976, 1988a, Rudall 1981). This protecting stump, or 'stylopodium', has been used as all important character in the current delimitation of the genera of the Hyptidinae and its constant occurrence in *Eriope* has been a significant factor in helping to define the genus (Harley 1973, 1976, 1988a). It is a conservative character in *Eriope* but it is present in several sections of *Hyptis* and is also found, in somewhat shorter form, in *Hyptidendron.* The stylopodium is absent from *Hypenia* and *Eriopidion.* In *Hypenia* there is a kink in the style above the nutlets which does not represent an abscission zone and its significance is unclear.

Style

The stigma is bilobed and divergent in the Hyptidinae and in most species the stigmatic lobes are clearly differentiated although in *Hyptis* section *Cyanocephalus* the lobes are poorly differentiated and club-shaped.

Nutlets

Nutlet morphology and anatomy have been investigated by Ryding (1992a, 1992b, 1993) as a source of characters to indicate relationships in the Labiatae. There is a close resemblance between the nutlets of members of the Hyptidinae (Ryding 1992b). All members of the subtribe have a characteristic areole, or depression, above the attachment scar which usually indicates how the nutlets have been inserted into the base of the style. This is extremely developed in *Marsypianthes* which has a very large, jagged scar on the proximal side indicating where the style base was fused to the nutlets (Rudall 1981). Nutlet morphology and anatomy in *Eriope, Eriopidion* and *Hypenia* is similar although *Eriopidion* has distinctive narrowly triquetrous nutlets.Crystals are present in the bone cells of the mesocarp of the Hyptidinae and Ocimeae but are absent from the Plectranthinae apart from the genus *Aeollanthus* (Ryding 1992b).

The nutlets in *Hypenia* are dark brown or black, 2.5 to 4 mm long and 1.5 to 3 mm broad, ovoid or triquetrous and myxocarpic, i.e. they produce mucilage on wetting. Myxocarpy is a common feature in subfamily Nepetoideae but is not found elsewhere in the family (Ryding 1992a). The extent of myxocarpy observed in *Hypenia* is variable. Most species produce little mucilage but some produce a dense layer enveloping the nutlet. Ryding (1992b) notes

a correlation between the maturity of the nutlets and the degree of myxocarpy and my informal observations support this.

Nutlet shape in *Hypenia* varies between species (figure 2-9). The mature nutlets of *H. vitifolia* are ovoid, black and rather shiny when fresh but become somewhat shrivelled when dry. In contrast those of *H. salzmannii* and *H. brachystachys* are triquetrous, sharply angled, brown and maintain a smooth surface when dry. *H. densiflora* has small, ovoid nutlets and section *Hypenia* have nutlets which are a similar shape but larger. A feature of *Hyptidendron* section *Hyptidendron* is the presence of winged nutlets (Harley 1988a). *H. brachystachys* sometimes produces nutlets with a pale, thin margin which could be described as a wing, although its functional significance is unclear.

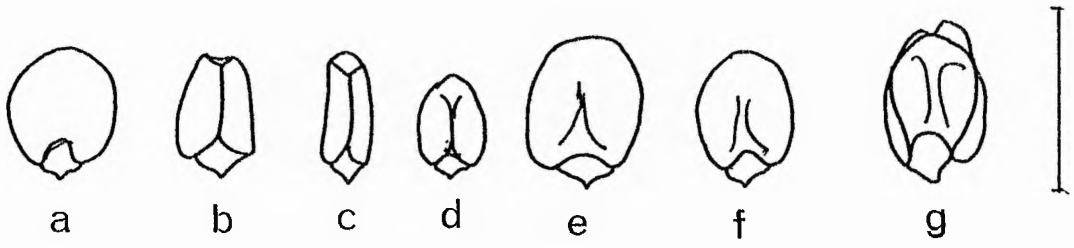


Figure 2-9: Nutlet morphology in *Hypenia* and *Hyptidendron*

a) *Hypenia vitifolia*; b) *H. salzmannii*; c) *H. brachystachys*; d) *H. densiflora*; e) *H. reticulata*; f) *H. macrantha*; g) *Hyptidendron canum*. Scale bar 5 mm.

ix) Systematic distribution of floral characters

Although *Hypenia* species have much in common in their calyx morphology with species of *Hyptis* and *Hyptidendron* there are important connections with *Eriope*. The calyx morphology of *Eriope* section *Nudicalyx* is very similar to *Hypenia brachystachys* and *H. densiflora*. Long hairs in the calyx throat are characteristic of *Eriope* section *Eriope* but are also found in *Hypenia* species.

There is a significant set of characters in the corolla which are correlated with taxonomy, although the association with pollination, a strong selection pressure, must be borne in mind. There is a complex distribution of character states between *Hypenia, Eriope* and *Hyptidendron.* There are many characters associated with the corolla and some unique states are observed in *Hypenia,* notably corolla colour and size, particularly in section *Hypenia.* However, *H. vitifolia, H. micrantha* and *H. salzmannii* display similarities in colour and shape with *Eriope* and *Hyptidendron.* Flower resupination is a potentially valuable character within section *Hypenia* and its distribution may indicate all association between species within the section.

Nutlets have been used to help define *Marsypianthes* and *Eriopidion,* both of which have unusual nutlet morphology. Within *Hypenia* there is less variation although the ovoid, black nutlets of *H. vitifolia* and triquetrous nutlets of *H. brachystachys* and *H. salzmannii* are distinctive.

E. Summary of morphological characters in *Hypenia* and their distribution in the Hyptidinae

Habit: geoxylic suffrutices are found in all species of *Hypenia* (although variably developed in *H. brachystachys* and many species of.*Eriope* but are not conspicuously developed in the rest of the Hyptidinae.

Stems: are very distinctive in many species of *Hypenia.* Fistulose stems with variably shaped swellings are present in several species of *Hypenia* and some species of *Eriope.* Wax shows a similar distribution but is possibly present in *Hyptis subtilis.*

Leaves: morphology and anatomy are very variable and useful at the species level in *Hypenia.* Differentiation is found between the morphology of the basal leaves and the phyllomes in *Hypenia* (except *H. micrantha*), *Eriope* and *Hyptis* sections *Laniflorae* and *Umbellatae.*

Indumentum: several hair types are useful to differentiate species within *Hypenia.* Glabrous inflorescence stems are present in most, but not all, species of *Hypenia,* some species of *Eriope* and *Hyptis subtilis.* Setose trichomes are widely distributed in *Hypenia* and *Eriope* but not outside these genera. Branched trichomes are found in *Hyptis* and *Hyptidendron.*

Inflorescence: all species of the Hyptidinae, including *Hypenia* have indeterminate thyrses. *Hypenia* species have variably developed indeterminate axes. Large thyrses with long indeterminate axes are characteristic of section *Hypenia, H. vitifolia, H. longicaulis, H. micrantha* and *H. salzmannii.* Shorter indeterminate axes are seen in *H. densiflora* and *H. brachystachys* and contribute to the densely clustered inflorescences of these species. Many *Hyptis* species have multi-flowered cymes but the majority of *Hypenia* species, and all species of *Eriope,* have single-flowered cymes with bracteoles. One-flowered cymes are found in *Hyptis,* e.g. *H. elegans,* but these cymes lack bracteoles. Bracteoles are found in all species of *Eriope* and *Hypenia,* except *H. vitifolia* and *H. longicaulis,* and indicate a common origin of one-flowered cymes in the two genera.

Flower orientation: inverted flowers in the Hyptidinae are found only in *Hypenia* section *Hypenia* subsection *Ellipticae.*

Calyx: in most species of *Hypenia* is similar to that found in many other members of the Hyptidinae. Zygomorphy caused by connation of the posterior lobes is found in *H. densiflora, H. brachystachys* and *Eriope* and some species of *Hyptidendron.* Hairs in the calyx throat are present in some species of *Hypenia, Eriope, Eriopidion* and *Hyptis.*

Corolla: provides many useful characters in *Hypenia.* The large red corollas of section *Hypenia* help to define the group, however, *H. subrosea* has white corollas similar in size and colour to those of *H. densiflora* and *H. brachystachys. H. vitifolia, Hyptidendron* and *Eriope* species have lilac corollas witli broad mouths. *Eriope* has a distinct constriction in the corolla around the nutlets which is absent from *H. vitifolia, H. micrantha* and *Hyptidendron vepretorum* but is present in other species of *Hypenia.*

Androecium: no characters in androecial morphology have been found which distinguish *Hypenia* and *Eriope* from the rest of the Hyptidinae.

Gynoecium: the stylopodium is absent from all species of *Hypenia* and most species of *Hyptis* and *Eriopidion.* It is present in all species of *Eriope* and several sections of *Hyptis.* Nutlets are variable at the species level in *Hypenia* and at the sectional and generic level in the Hyptidinae and help to define *Marsypianthes* and *Eriopidion.*

Chapter 3: Cytology

i) Introduction

Cytological investigation in *Hypenia* was undertaken to search for further evidence to investigate the relationship with *Eriope.* The comprehensive study of Harley and Heywood (1992) indicated cytological divergence in *Hypenia vitifolia, H. longicaulis* and *H. salzmannii* but little variation, in an albeit limited sample, of the rest of the genus which were all identical in number to *Eriope.* Further investigation was required to confirm the counts for *H. vitifolia* and *H. salzmannii* and to sample further in the rest of *Hypenia.*

ii) Chromosomal variation

Cytology is the study of cell contents, particularly the heavily staining bodies known as chromosomes. The chromosomes were recognised as the bearers of the genetic material at the beginning of the century and as such have received particular attention ever since (Swanson et al.1981). The karyotype was introduced as a concept to describe the structure of the chromosomes and their basic number. It is based on consistency of chromosome structure within taxa. Previous work in the Hytpidinae indicate that their chromosomes are small and karyotypic variation is difficult to observe (Harley and Heywood 1992).

Change in chromosome number can occur through addition or loss of individual chromosomes, aneuploidy, or, alternatively, the basic number remains constant but there is multiplication of the number of sets of chromosomes in each cell. This process, known as polyploidy, is very common in plants and in many cases series of polyploids can be identified in related species.

Relatively simple techniques based on staining the chromosomes so that they can be viewed under a light microscope can reveal considerable variation in chromosome number which can also be used as indicators of possible evolutionary relationships.

iii) Cytological methods

Counts were made of all available *Hypenia* accessions collected during my trips, or recent Harley expeditions, to Brazil. Existing collections at Kew had been investigated (Harley and Heywood 1992). Root tips were obtained from seedlings grown in petri dishes. Roots were pretreated with 8-hydroxyquinoline (OQ) for 4 hours in the dark at 20°C. They were then

fixed in 3:1 absolute ethanol:glacial acetic acid for a minimum of one hour and stored in the fridge at 4°C. The fixed material was hydrolysed in 1 molar hydrochloric acid at 60°C for 8.5 minutes. It was then stained in Feulgen's reagent for a minimum of 30 minutes and if not used immediately was stored in the reagent and kept in the fridge. The root material was further softened in a mixture of 4% pectinase and 1.5% cellulase for 30 minutes. Root tips were dissected in 2% aceto-orcein and squashed under a cover-slip. Slides were sealed with rubber solution for temporary storage and permanised by freezing with CO_2, dehydrating in absolute ethanol and mounted in Euparal resin. Slides are stored in the Jodrell slide collection.

Observations were made under x40 magnification and photographs taken at x100. Further magnification was acheived with x 1.25 extra magnification ring. Photographs were taken on Ilford Pan F film at ASA 15.

iv) Results of chromosome counts

The chromosome counts made during this study are presented in table 3-1 and photographs are shown in figure 3-1. My chromosome counts confirm those made by Harley and Heywood (1992) for *Hypenia vitifolia,* 2n=28, *H. salzmannii,* 2n=12 and *H. densiflora,* 2n=20. Counts for species not previously reported were made for *H. brachystachys, H. aristulata, H. reticulata* and *H. macrantha* sensu stricto and were all 2n=20. Numbers reported by Harley and Heywood (1992) but not confirmed here due to lack of material, were 2n=22 for *H. longicaulis* and 2n=20 for *H. subrosea.*

Table 3-1: Chromosome counts made during this study

Species	voucher	2n
Hypenia brachystachys	Atkinson et al. 134	20
H. brachystachys	Atkinson & da Silva 150	20
H. densiflora	Atkinson & da Silva 151	20
H. aristulata	Atkinson et al. 135	20
H. macrantha	Atkinson & da Silva 150	20
H. reticulata	Atkinson & da Silva 152	20
H. reticulata	Forzza 111	20
H. salzmannii	Projeto Chapada Diamantina 2501	12
H. vitifolia	Projeto Chapada Diamantina 2500	28
Hyptis lythroides	Atkinson & da Silva 149	28

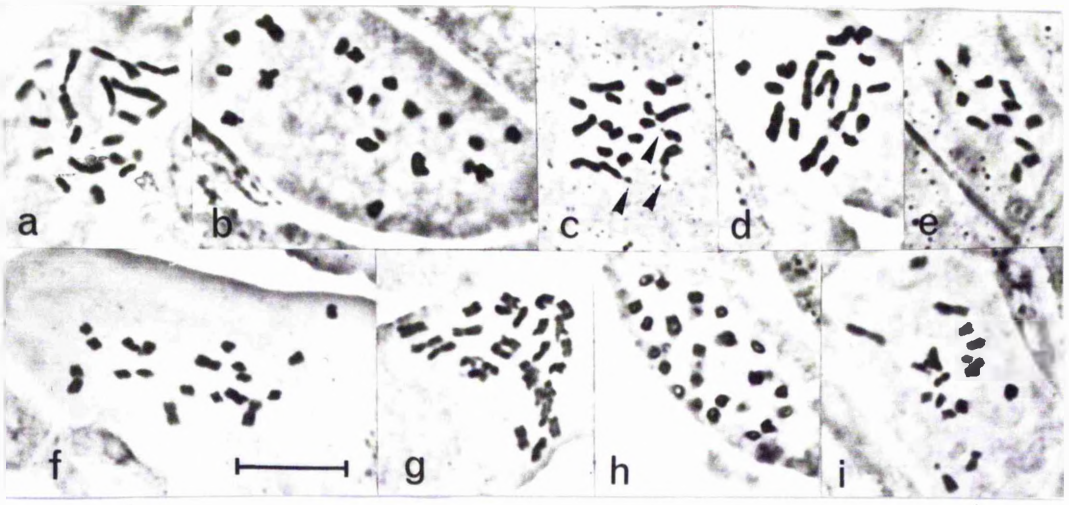


Figure 3-1: Chromosome preparations

a) *Hypenia reticulata*; b) *H. densiflora*; c) *H. macrantha*; d) *H. aristulata*; e) *H. salzmannii*; f) *H. retiuculata*; g) *Hyptis lythroides*; h) *H. vitifolia*; i) *H. brachystachys.* Scale bar = 10 µm. Arrows indicate satellites.

v) Chromosome number in the Hyptidinae

Previous chromosome counts in the Hyptidinae are summarised in table 3-2. Chromosome number varies from 2n=12 in *Hypenia salzmannii* to 2n=ca.96 in *Hyptis villosa* Pohl ex Benth. (section *Xylodontes* (Benth.) Epling.). In *Hyptis, Marsypianthes* and *Peltodon* common numbers are 2n=28, 30 and 32. Harley and Heywood (1992) suggest that the base number of *Hyptis* is x=8, in which case subsection *Eriocephalae* (Epling) Epling of section *Mesosphaeria* Benth., all of which have 2n=16, are the only diploids so far reported in the Hyptidinae. The tetraploid number of 2n=32 is widespread in the Hyptidinae and is found in *Peltodon* and all sections of *Hyptis* investigated except for sections *Leucocephala* Epling, *Xylodontes* and *Pusillae* Epling, sections which are represented by limited sampling. Numbers possibly derived by aneuploidy are equally common, 2n=30 is the only number found in *Asterohyptis, Hyptis* section *Leucocephala* and *Marsypianthes* and is widespread throughout *Hyptis*. 2n=28 is the other reasonably common number and is found throughout the larger sections of *Hyptis*. *Hyptis lythroides* Pohl ex Benth. in section *Polydesmia* Benth., subsection *Glomeratae* Benth., with 2n=28, was counted for the first time in my study. *Raphiodon echinus* is reported to have 2n=26, a unique number in the Hyptidinae.

14 of the 27 counts made in the group prior to Harley and Heywood (1992) refer to *Hyptis suaveolens*, a pantropical weed with chromosome numbers reported of n=14 and 16 and 2n=28, 30, 32 and 24 and indicate the possibility of aneuploidy within species in the Hyptidinae. All eight counts of *H. suaveolens* made by Harley and Heywood (1992) found 2n=28.

High polyploids are unusual in the Hyptidinae counted so far. *Hyptidendron canum* is reported as having 2n=ca.64, suggesting a tetraploid origin from 2n=32. 2n=64 is also reported for *Hyptis floribunda* in section *Minthidium* and *H. conferta* Pohl ex Benth. (section *Hyptis* subsection *Hyptis*). There are a number of other counts of ca.63, 63, 54, 46, ca.96, ca.90, ca.58, all potentially of tetraploid or hexaploid origin from 2n=32 and 28 with subsequent loss of chromosomes.

Harley and Heywood (1992) counted 10 species of *Eriope*, including a wide range of accessions encompassing the variation found in *E. hypenioides* and *E. macrostachya* Mart. ex Benth., and found no deviation from 2n=20. A previous count of n=20, suggesting a tetraploid of 2n=40 had been reported for *E. crassipes* by Coleman (1982).

Table 3-2: Summary of previous chromosome counts in the Hyptidinae (after Harley & Heywood 1992)

Species	Section	2n
Eriope	*Nudicalyx* (5,7)	ca.20, 20
	Eriope (6,12)	20, n=20
Hypenia densiflora (1)		20
H. longicaulis (1)		22
*H. reticulata** (1)		20
H. salzmannii (3)		12
H. subrosea (1)		20
H. vitifolia (1)		28
Hyptidendron (1,1)		ca. 64
Asterohyptis (1,4)		30
Hyptis	*Umbellatae* (1,1)	32
	Laniflorae (3,3)	32
	Minthidium (2,2)	28, 64
	Fruticosae sect. nov. (2,3)	20, 32
	Mesosphaeria (15, 41)	16, 24, 28, 30, 32, 40, 46
	Trichosphaeria (1,2)	32, 24-28
	Polydesmia (8,17)	28, ca.30, 32, ca. 60, ca. 63, 63, 64
	Leucocephala (2,3)	30
	Cyrta (2,2)	32, ca.60
	Cyanocephalus (1,1)	32
	Gymneia (1,2)	ca. 30, 32
	Eriosphaeria (1,1)	32
	Xylodontes (2,4)	32, ca. 90, ca.96
	Apodotes (1,2)	32, ca. 32
	Hyptis (18, 44)	30, 32, ca.32, ca.34, 46, 54, 58,ca.58, 60,64
	Pusillae (1,2)	30, 56
Marsypianthes (3,10)		30
Peltodon (3, 5)		30, 32
Raphiodon (1, 2)		26

* referred to by Harley and Heywood as *H. macrantha,* the voucher quoted by them is referred by me to the very variable *H.reticulata.* Numbers in brackets refer first to the number of species sampled and then to the number of accessions counted. Figures are taken from Harley and Heywood (1992) from

their table I which lists counts they made and from their table II, previous counts in the Hyptidinae. The discussion of chromosome number refers to counts made by Harley and Heywood (1992) unless otherwise indicated.

vi) Karyotype and chromosome size in the Hyptidinae

The chromosomes in the Hyptidinae, including all species of *Hypenia* investigated (figure 3-1), are small (< 3 μm long) and metacentric. Two pairs of chromosomes in *Hypenia* species which have 2n=20 appear to have satellites (figure 3-1 a, c, i). At least one of these pairs of satellited chromosomes are present in *H. salzmannii* (figure 3-1 e). It is difficult to see from available preparations whether satellites are present in *H. vitifolia* (2n=28, figure 3-1 h) or *H. longicaulis* (2n=22). *Hyptis lythroides* has at least one pair of chromosomes with satellites (figure 3-1 g).

vii) Significance of genome size

H. salzmannii has the lowest chromosome number in the Hyptidinae and one of the lowest in the Labiatae. The lowest reported number in the family is 2n=10 (Marrero 1992). The low chromosome number of *H. salzmannii* suggest a reduction in the size of the genome relative to other members the Hyptidinae. A correlation between life form and genome size has been noted in herbaceous plants (Bennett & Smith 1991). Small genomes are associated with small annual species and larger genomes with larger species which have longer growth cycles.

H. salzmannii is found in a wide range of vegetation types such as dry, nutrient poor caatinga, coastal sand dune restinga, upland savanna campo rupestre, and disturbed areas throughout northeastern Brazil and in the savannas of Venezuela and Guyana and it shows a stronger ruderal nature than other species in the genus.

viii) Systematic significance of cytology in *Hypenia*

Cytological evidence supports the position of *Eriope* close to *Hypenia* but also indicates the divergence of *H. vitifolia* and *H. salzmannii.* 2n=28 reported for *H. vitifolia* is a common number in *Hyptis* but 2n=22 for *H longicaulis* is unique in the Hyptidinae. This perhaps indicates a more profound difference between the species than is reflected in their morphology. The low chromosome number of *H. salzmannii* reflects the divergent nature and unusual ecology of this species within the *Hypenia / Eriope* clade. The chromosome number does not help to indicate the affinities of this species.

Chapter 4: Cladistic theory

A. Introduction

The most important taxonomic task above the species-level in *Hypenia* was the construction of an appropriate classification that reflected the distribution of morphological variation outlined in chapter 2 and at the same time was an accurate representation of evolutionary relationships. To this end a phylogeny was required that explained the observed distribution of characters in an evolutionary framework and enabled the construction of an accurate classification. Recent theoretical developments in phylogenetic systematics have had a profound impact on taxonomy and after considerable debate it is widely accepted as a major contribution to taxonomic research, introducing a method with a theoretical base and which introduces the possibility of objective investigation. For this reason it was employed in this study.

The theory of cladistics is discussed in this chapter with an outline of the methods used in this study.

B. The theoretical base of cladistics

The goal of taxonomy is to classify organisms into groupings in a way which conveys the maximum information with maximum stability. A classification which reflects true evolutionary relationships is preferred because predictive information is inherent in an evolutionary system (Stevens 1986). Such a classification is automatically stable because only one system, that which truly represents the course of evolution, is possible (see chapter 8 for further discussion of the theory of classification). Hierarchical systems are logically compatible with the process of evolution because they are produced by phylogenetic processes, i.e. non-reticulate relationships between groups of species (Hennig 1966). Reticulate relationships, or tokogenetic processes, which occur between groups of individuals within species do not produce a hierarchy and must be studied separately from phylogeny. History must be inferred from the evidence available and a method is necessary to recapitulate the pattern of evolution using information gathered from only a fragmentary sample. Cladistics was devised by Hennig (1966) as a method to reconstruct the evolutionary phylogeny of organisms. Cladistics is often used as a synonym for phylogenetic systematics. Cladistics strictly refers to the search for clades, i.e., groups in a dichotomously branching cladogram. Phylogenetic systematics is a more general term used to describe the

investigation of evolutionary relationships but since its use as the title of Hennig's (1966) book in which he develops cladistic methodology it has become synonymous with cladistics.

i) Non-cladistic methods of classification

Two previous schools: phenetics, expounded by Sneath & Sokal (1973); and evolutionary systematics, described by Mayr (1969a) and Davis & Heywood (1963), have attempted to devise methods which can be applied to taxonomy to produce consistent classifications.

Phenetics is based on gathering the maximum number of characters, giving them all equal weight and applying multivariate statistics to the data to produce clusters of similar organisms, termed operational taxonomic units (OTUs). There are no assumptions made about evolutionary history or relationships of the group under investigation. Phenetic approaches are relatively simple to apply since they do not require the distinction to be made between homology and analogy (see below) and they rely on the weight of evidence provided by a large data set to generate measures of overall similarity, regardless of evolutionary history.

The results of phenetic studies depend on the statistic applied. Different clustering methods give different results and because of the lack of a theoretical, evolutionary, base there is no criterion for adopting any one method over another. Phenetics therefore does not provide an acceptable conceptual framework on which to base classification.

Evolutionary systematics is based on the selection of characters which appear to convey maximal information about evolutionary relationships between organisms. Characters are carefully selected and those which appear to be homologous and free of convergence, i.e. they are not analogous as a result of different evolutionary pathways converging on the same functional product, are used as the basis of their classifications. Although this method is employed to some extent by all taxonomists, it is dependent on the knowledge and experience of the individual observer and it is therefore impossible to submit systems derived in this way to objective verification.

Cladistics was developed as an attempt to overcome these problems and has been enthusiastically adopted by many systematists. However, many reservations have been, and continue to be expressed (Cronquist 1987, Hedberg 1995). Despite the controversy and the

frequently acrimonious literature, cladistic methods are now employed by most monographers to illuminate evolutionary relationships in their group.

ii) Defining cladistic terms

Evolutionary biologists such as Mayr (1969a) recognised the undesirability of classifying organisms into groups which do not share a common ancestor, i.e. **polyphyletic groups**. Hennig (1966) went further by highlighting the problem of paraphyly, i.e. groups which do not include all the descendants of a common ancestor. In Hennig's system both polyphyletic and **paraphyletic groups** were disallowed and only **monophyletic groups**, i.e. all taxa derived from a common ancestor that is not also the ancestor of another group, were accepted. Prior to Hennig (1966) monophyly was applied in a much more general sense to indicate common ancestry but this allowed any group to be described as monophyletic if all life is descended from a single common ancestor. The rejection of paraphyletic groups has caused much debate and their place in taxonomy is discussed in more detail in chapter 8.

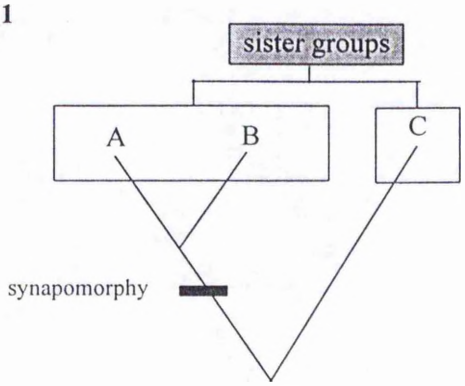
Cladistics is the method by which monophyletic groups are discovered using the distribution of shared, derived characters, or **apomorphies**. Shared characters which are not confined to a group, but are common to all members and may also be found outside it, **plesiomorphies**, convey no information about evolutionary relationships. The status of a character as an apomorphy or plesiomorphy changes according to the taxonomic level being investigated, e.g. the modified anterior lobe of the Hyptidinae is an apomorphy used to define the group within the Ocimeae but it has no value within the Hyptidinae to distinguish evolutionary relationships between *Hypenia, Eriope* and *Hyptis,* since it is common to them all and is therefore plesiomorphic within the subtribe.

A **cladogram** is a representation of clades which are dependent on the order of branching. Branches are generated by speciation events, but there is no indication of the extent of differentiation between clades on each branch. A diagram which demonstrates the amount of divergence between clades is referred to as a **phylogram**. Cladograms are commonly described as **trees** and the terms are used interchangeably in this study. Strictly however, trees are actual representations of phylogeny and can also include information on the ancestral positions of taxa.

Sister groups, defined by Hennig (1966) as 'species groups that arose from the stem species of a monophyletic group by one and the same splitting process' are the basic expression of

phylogenetic relationships. Hierarchies are constructed by the identification of sister groups, identifying the sister group of A and B as C (example 1) by the presence of shared derived characters, **synapomorphies,** will produce a three-taxon statement, the basic unit of a cladogram.

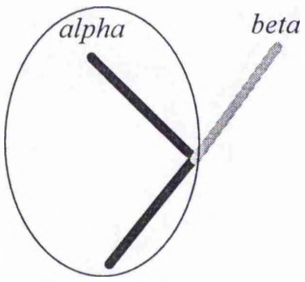
Example 1



iii) The meaning of cladograms

Hennig's (1966) original formulation of phylogenetic systematics insisted that species went extinct at branching nodes. Using this criterion neither A, B nor C in example 1 could be the species which contained the ancestor of the other two species since the ancestral species must become extinct at the branching point. Such an event is essential if the concept of paraphyletic species is to be avoided. If one species, *alpha*, gives rise to another species, *beta* and survives the process, *alpha,* which contains the ancestor of *beta,* becomes paraphyletic by the recognition of *beta*.

Example 2



To avoid this difficulty the school of **pattern cladistics** was developed (Nelson & Platnick 1981, Patterson 1988a, Nelson 1989) whereby the process of evolution in which ancestral taxa could survive beyond branching points was separated from the pattern produced as a result of evolutionary processes. Pattern cladistics has generated considerable controversy because it claims to divorce the pattern found in nature from the process of evolution which generated it. De Queiroz and Donoghue (1990) questioned why Nelson (1989) preferred to

represent relationships in a nested hierarchy if no explanation of common descent was required. Other arrangements may be just as parsimonious and nested hierarchies may also be explained by causes other than common descent. The rejection of the necessity for a link between the theory of evolution by natural selection and the pattern seen in nature was also rebutted by Ridley (1986) who argued that the strength of cladistics is its reliance on evolutionary processes which provide a theoretical base from which cladistic patterns can be explained. However, the investigation of phylogeny relies on the evidence provided by the current distribution of characters, i.e. phylogeny can only be inferred from pattern, not from processes for which we have no evidence.

Despite the importance of developing a consistent model the theoretical arguments about what the trees actually represent have little impact on the practice of cladistics which is mostly used to generate hypotheses to test taxonomic expositions. More practical considerations are concerned with developing an appropriate species concept which is outwith cladistics, but a necessary first step to any analysis. This step is followed by the identification of homologous characters, character coding, finding appropriate outgroups and choosing among many alternative trees.

iv) Homology

A fundamental concept in all evolutionary studies was recognised in the nineteenth century by Owen (cited in Scotland 1992) and is based on the distinction between homology, ' the same organ .... under every variety of form and function' and analogy, ' a part or organ .... which has the same function'. Because **homology** is the basis for recognising monophyletic groups it has been equated with synapomorphy (Patterson 1982) and has the same properties in that every proposal of homology is a hypothesis of monophyletic groups and that paraphyletic groups are not characterised by homologies. Shared characters which appear similar but are found to have originated independently are called **homoplasies** and cannot indicate true evolutionary relationships.

Determining homology is therefore one of the fundamental procedures in any analysis of the evolutionary history of a group of organisms. The process of deciding which characters are homologous is one of observation followed by some test of the hypothesis. The first criterion for deciding on homology is similarity based on position, ontogeny or composition of the organ being observed. **Similarity** has been proposed as a test of homology by Patterson (1982) but as de Pinna (1991) points out it is merely the first step in proposing a hypothesis of homology. Other tests suggested by Patterson (1982) are: **conjunction**, so that two organs

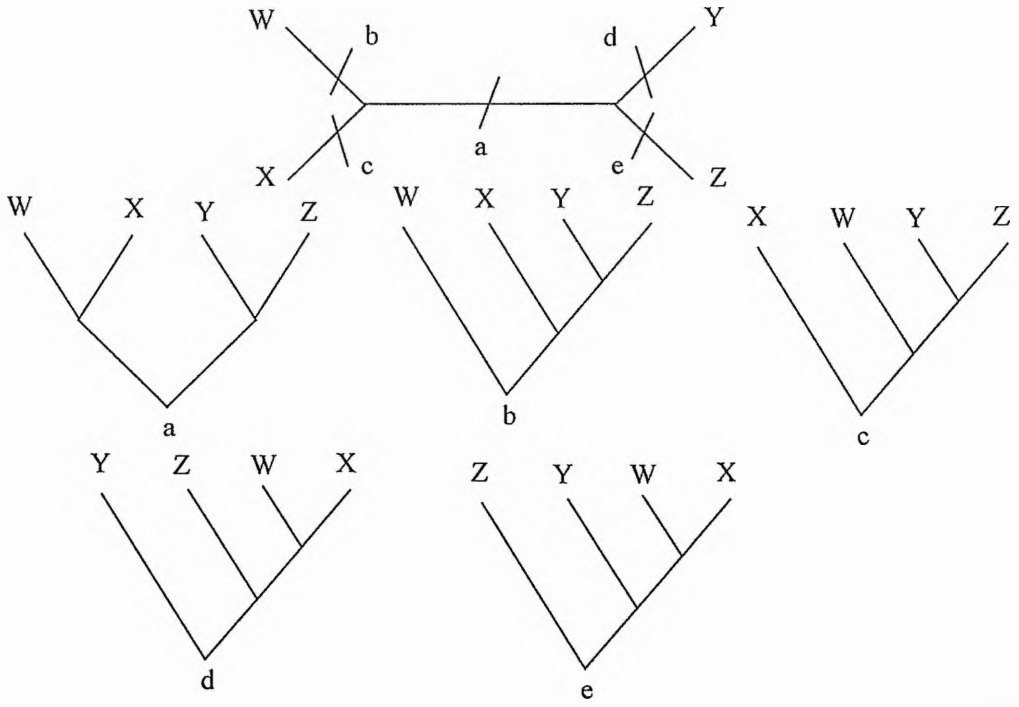
in the same individual cannot be homologous; and **congruence**, where the homology conforms to the same pattern of homologies proposed for other organs.

To illustrate conjunction Patterson (1982 and 1988b) cited the example of the homologous structure of arms and wings in mammals and birds respectively and then went on to consider the case if angels, which have both arms and wings, were discovered. He proposed that the existence of angels would refute the proposed homology of arms and wings. However, the presence of both may be due to one organ being autapomorphic in angels (de Pinna 1991). For example, arms may be homologous with the wings of birds and the arms of angels whereas the wings of angels are a new structure confined to angels with no homologues in birds or mammals. Conjunction therefore is not a test of homology although in most, less hypothetical, examples it is a good base for an initial hypothesis. Congruence has been proposed by de Pinna (1991) as the only effective test of homology since it is based on the pattern produced by a combination of independent characters and is therefore a measure of the fit of the character to the pattern produced by all included characters.

v) Tree rooting

Sister groups can only be identified and a hierarchy established if the network of relationships based on synapomorphies is rooted. In example 3, with four taxa, WXYZ, there are five possible root positions at either a, b, c, d, or e so that the five trees shown are possible. The correct position of the root is dependent on establishing which character state is apomorphic and which plesiomorphic.

Example 3



Direct information on the character state in the ancestral taxon is rarely, if ever, available. The process of speciation can only be observed in retrospect and identification of the individual, or population, which gave rise to a new species is impossible or impracticable, even if there is a good fossil record. To identify which character states are derived and which are of more widespread occurrence therefore requires an indirect method of assessment. The most commonly employed method is that of **outgroup** comparison in which the character state in a selected outgroup is used to enable the conversion of an unrooted network into a rooted tree (Watrous & Wheeler 1981). Other methods have attempted to use fossil groups for comparison, ontogeny or the identification of transformation series within selected characters (Nelson 1985) but these methods rely on *a priori* assumptions about character evolution. The outgroup comparison method does not require *a priori* assessment of the character state found in ancestors but is still subject to problems associated with selecting the correct outgroup (Steussy 1996).

C. Cladistic methodology used in this study

The general discussion presented above gives a broad outline of the theoretical basis of cladistics but is not concerned with the practicalities of constructing a cladogram. The following is an outline of the general methods used in this study.

i) Tree construction and optimisation

For most data sets there is more than one possible tree that can be constructed to represent relationships between the taxa, creating the difficulty of how to choose the optimal tree amongst many conflicting trees. The most commonly applied criterion is that of **parsimony** where the tree (or more usually trees) which require the least number of character changes is chosen. Parsimony is based on the principle that if there are competing scenarios to explain a given event the simplest, i.e. the shortest, is chosen (Camin & Sokal 1965). It has been justified because it describes broad structural properties of the evolutionary process (Sober 1983). Other methods of optimisation have been developed such as three-item statement analysis (Nelson & Platnick 1991) and maximum likelihood which evaluates the fit of a hypothesised model of evolution to the pattern observed from the real data. Maximum likelihood is most often applied to molecular data which is more amenable to the construction of evolutionary models (Felsenstein 1992).

A criticism of cladistic methods concerns the mathematical difficulties which arise in tree construction (Pankhurst 1991). The potential number of trees which can be generated rises at an enormous rate with the addition of more taxa. There is only one possible unrooted tree to describe the relationship between three taxa, three unrooted trees for four taxa and 15 for five. With six taxa the number rises to over 100 and for ten there are millions of possible trees.

There are a number of algorithms, or stepwise procedures, to find possible trees from a matrix of taxa and characters, which vary in their approach to generating trees and searching for the most parsimonious one. For a matrix with a very few taxa it is possible to generate every possible tree and search for the one which requires the least character changes, i.e. is the most parsimonious. Matrices containing up to about 20 taxa can be effectively searched using a **branch and bound** algorithm which eliminates groups of trees which are longer than the minimum length already found. If there are more than about 20 taxa in the data matrix the number of potential trees is so large that it is impossible to search them all to find the most parsimonious tree and **heuristic** algorithms are employed.

Heuristic methods involve the generation of an initial tree which is rearranged in a way that minimises the length (using parsimony as the optimality criterion) of the tree until optimisation can proceed no further. **Stepwise addition** is a heuristic algorithm which generates a tree as a starting point for further rearrangement. Three taxa are chosen for the initial tree then the next taxon is joined and the three possible trees generated by four taxa are evaluated and the shortest chosen. This optimal tree is used as the base in the next round when a further taxon is joined at all five possible places. The optimal tree is again saved and the process is repeated until all the taxa have been added to the tree.

Stepwise addition chooses the solution which looks best under current conditions, i.e. it sets off to climb the nearest optima without considering more distant optima which may be higher. Placement of a taxon may be optimised given the current taxa in the tree but if further taxa are added the initial placement may become suboptimal. Once the decision has been made to connect a taxon at a certain point however, the consequences on further trees must be accepted. Which taxa to use to construct the initial tree and the order in which they are added is therefore important. In Paup 3.1.1 (Swofford 1993) the addition sequence can be set so that the initial tree is generated from taxa incorporated in a random order and multiple replicates help to minimise the risk of merely finding a local optimum.

Branch swapping is an additional algorithmic process which is combined with tree-generating algorithms in PAUP 3.1.1 (Swofford 1993) to try and overcome the problem of getting caught in local optima. Several algorithms can be employed to generate new rearrangements which can then be tested under the optimality criterion. Tree bisection and reconnection (TBR) bisects the tree and reconnects the subtrees by joining a pair of branches, one from each subtree. All possible bisections and reconnections are evaluated.

ii) Outgroups

The outgroup comparison method used in this study follows the procedures outlined by Nixon and Carpenter (1993) and requires the identification of a suitable taxon in which character states can be correlated sufficiently with those of the ingroup (i.e. the group under investigation) to enable the establishment of homology. Denser sampling gives more accurate results so inclusion of a number of outgroups is more likely to produce a reliable topography and may help to identify the sister group if it is unknown. With the inclusion of a number of outgroups, characters which resolve relationships within the outgroup, even if they do not apply to the ingroup, are necessary for the analysis.

The matrix is first analysed to construct a network and then a root is placed between the ingroup and outgroup to convert the network into a tree. If it is not possible to root in this position because taxa become mixed between the two groups it implies that the ingroup is not monophyletic or defining characters have been overlooked. Character polarity can then be inferred directly from the cladogram.

iii) Character optimisation

Character optimisation assigns character states to particular nodes in a cladogram so that their evolution can be traced along branches. It is complicated by ambiguous, i.e. homoplasious characters, since more than one optimisation is possible. A character may show ambiguous evolution either because it has multiple, parallel, origins or because it has been derived at one point in the cladogram and lost by reversal at a subsequent point. Reversals are evolutionarily more parsimonious because they require a single hypothesis of homology rather than the more complex hypothesis favouring multiple parallel origins (de Pinna 1991). However, programs such as PAUP 3.1.1 (Swofford 1993) count the actual number of steps. This means that the origin of a character state which reverts to the previous

state counts as two steps, the same as if the character state had been independently derived in two separate lineages.

iv) Tree comparison and evaluation

Consensus

Applying the principle of parsimony will greatly reduce the number of trees that need to be evaluated from a data set but in most cases more than one shortest tree is likely to be found. Various measures have been proposed to generate a consensus tree which indicates areas of agreement between multiple shortest trees. Consensus techniques can also be applied to information from different sources, e.g. combining trees generated from molecular and morphological data sets. All consensus trees are obtained by combining comparable groups, components, from all the original trees but techniques differ in the way in which the components are combined. The most commonly used consensus techniques, strict, semi-strict, also known as combinable components, and majority rule, are discussed.

Strict consensus is the most stringent consensus technique in that it only accepts components which are present in every tree. Components present in a strict consensus are unambiguous because they are present in all the original trees. However, any ambiguity will produce a polytomy where the relationship between the components is not resolved and less conservative approaches can reveal possible relationships obscured by the strict consensus.

Semi-strict consensus includes all those components which are not contradicted in any of the trees. Components therefore do not need to be present in all of the trees. Semi-strict consensus can reveal groups obscured in the strict consensus and if trees are combined from different data sets components only present in one set may remain in the semi-strict consensus which would not be accepted by the strict consensus.

Majority rule generates a tree which includes all the components which occur in a certain percentage of the original trees. The cut-off is arbitrary, although it is often set at 50%, but there is no reason to prefer to accept components that occur in the majority of cladograms over even those that only occur in one cladogram. For this reason majority rule is not generally accepted as a consensus technique but it is a useful means of searching for patterns when large numbers of trees are generated.

Tree statistics

There are a number of statistics which can be used to describe aspects of trees and the relationship between the trees and the data set from which they were generated.

Tree length is the statistic which cladistic analyses aim to optimise for a particular data set. It is the trees of minimal length which maximise information about relationships based on

the principle of parsimony. Tree length is a measure of the number of steps of character state changes, or transformations, which are required to account for the distribution of data on the tree. It depends on the number of characters and the number of character states and for each tree is derived by adding the number of lengths of the individual characters.

The **consistency index** (Kluge & Farris 1969), *c.i.*, for a character has been used to measure the extent of homoplasy of that character. It is represented by *m/s* where m is the minimum amount of change possible for a character (*m* is equal to the number of states of the character minus one) and *s* is the actual number of changes in the character observed on the tree. If *s* exceeds *m* the extra steps observed are attributable to homoplasy and the consistency index has been used as a measure of homoplasy.

The **ensemble consistency index** can also be derived from $CI = M/S$, where *M* and *S* are equivalent to *m* and *s* summed for all the characters in the data set. If no homoplasy is present CI = one, decreasing as homoplasy increases. *CI* has been found to be negatively correlated with the number of terminal taxa and the number of characters. This reduces its value in comparing data sets with different numbers of taxa or characters although Goloboff (1991) considers it an appropriate measure of homoplasy. The *CI* is sensitive to uninformative characters, symplesiomorphies and autapomorphies, which inflate it without adding support for groupings.

Consistency index expressed as *m/s* can never be zero and the **rescaled consistency index** was devised to allow a character which fits the tree as poorly as possible to be expressed as zero (Farris 1989). It is obtained by linear rescaling of *CI* from zero to one.

The **retention index**, $RI = g - s / g - m$, where *g* is the greatest amount of change that a character could require on the tree (Farris 1989). *RI* is high when character state changes occur on internal nodes and low when concentrated on branches leading to terminal taxa. It has the advantage that it is not sensitive to uninformative characters.

Confidence assessment of trees

Tree description is extremely complex and poorly understood but it is often considered desirable to be able to assess the amount of faith we can put in trees. True statistical tests of confidence have not been devised but three alternative approaches have been proposed.

Bootstrap and jacknife approaches have been applied to trees (Felsenstein 1985). These techniques are based on resampling of the data to assess how frequently components found in the original trees are repeated. They involve random sampling of the rows or columns to give a data set the same size as the original which is then analysed to give a tree or trees. The process is repeated and the strict consensus after 100 replicates can be compared with the original tree or trees. The percentage occurrence of a particular component can be considered an index of support for that component. Felsenstein (1985) considers 95% to represent support of the component by at least three characters. However there is no way to assess the evolutionary significance of any level of bootstrap (or jacknife) support and no reason why components which are supported by three or more characters are evolutionarily more significant than any other.

Randomisation is an alternative means of resampling the data and is based on randomly reassigning character states within the data matrix (Faith & Cranston 1990). Significant cladistic covariance is said to exist if 95% or more trees produced from a randomly resampled data matrix are as long as or longer than the original tree. This test has been criticised as a weak means of corroborating a hypothesis (Carpenter 1992) and again there is no evolutionary reason to favour any level of randomness over any other.

Decay analysis is a commonly used method of assessing tree stability. It assesses branch support by searching for the number of extra steps required to lose a branch in the consensus of near-most parsimonious trees (Bremer 1994). It is a simple test which does not involve perturbing the data set. However, the original tree, or trees, were favoured because they were the most parsimonious and searching for longer trees defeats the purpose of the original search.

None of the above measures of confidence were applied in this study. Cladistic methods are used to generate trees which are hypotheses of evolutionary relationships based on explicit interpretation of data. Parsimony was used as the criterion for finding the tree (or trees) which best explains the distribution of characters. It is therefore inappropriate to apply statistical tests which eliminate parsimony.

Chapter 5: Morphological analysis

A. Introduction

Before a data matrix for a cladistic analysis could be constructed careful consideration was needed to ensure characters were homologous and that appropriate taxa were included in the analysis. These two subjects are discussed in this chapter. The resultant matrix is presented in appendix III. The morphological data matrix was then analysed and the results are presented in the final part of the chapter.

B. Character selection and coding

The selection and interpretation of characters is probably the most important step in a cladistic analysis (Pimentel & Riggins 1987). Despite, or perhaps because of, this importance there is considerable discussion and disagreement in the literature about the methods best employed. Much of this discussion centres around the best method of coding characters and the true nature of the variation being described (Stevens 1991).

i) Character coding

Characters can be treated as independent variables which have alternative character states, i.e. each character in a data matrix is represented by a single column and the symbol within the column indicates the state assigned to the character in a particular taxon; or the states can be treated as individual columns, the states within which are either present or absent. Using the latter method to score multi-state characters effectively treats each state as a separate character with only the possibility of presence or absence. For example if the corolla exists as red, blue or white, coding the states separately introduces three characters, red corolla present or absent, blue corolla present or absent, and white corolla present or absent. This introduces two more characters than if they are coded as a single character, corolla colour, with three possible alternative states. This method has been advocated for multi-state characters (Wiley 1980) and has also been reiterated for two-state characters (Pleijl 1995) whereby two separate presence/absence characters are required to describe the variation found in a single character. These approaches introduce considerable redundancy and overweight the value attached to them since two or more characters are required to describe a single evolutionary transformation (Pimentel & Riggins 1987).

ii) Quantitative versus qualitative characters

Stevens (1991) in his classic paper discusses the nature of many characters used in cladistic analyses. His discussion revolves around the distinction between quantitative and qualitative characters and the difficulty of defining quantitative character states satisfactorily. Stevens (1991) points out that many so-called discrete, qualitative states are in fact continuous, quantitative characters and are only coded in discrete states by the use of botanical definitions which do not reflect the reality of the variation. Gift and Stevens (1997) tested the value of quantitative characters in *Kalmia* by giving a range of measurements representing continuous variation in several characters to a number of different people and asking them to divide the results into states. There was almost no agreement on the character states chosen, highlighting the extreme degree of subjectivity such characters are subject to. However, Thiele (1993), whilst accepting that many quantitative characters are difficult to divide into exclusive states, advocates their use since they can impart important information to a cladistic analysis. There can be considerable information in quantitative characters and eliminating them may unnecessarily reduce resolution and frequently result in the loss of the majority of characters! When quantitative characters are used the structure they impose should be treated with some caution and the extent of the variation present in the character should be made as explicit as possible.

iii) Ordering and weighting characters

It is sometimes considered desirable to constrain the order of transformation between different character states, e.g. it may be considered that red flowers can transform to white flowers but the reverse transformation is not possible. In such case it is possible to constain the analysis in accordance with hypothesised character transformations by ordering characters. However, ordering characters requires assumptions which rarely have verifiable foundation.

In addition, it is also possible to weight characters which are considered to have high evolutionary significance. Weighting may be applied prior to the analysis, *a priori* but there is rarely, if ever, any justification for applying extra value to characters in this way. *A posteriori* weighting can also be applied using successive weighting. This technique selects characters which show low homoplasy in initial analyses and then weights them in successive searches. The theoretical basis of this approach is controversial (Carpenter 1994) and in practice also presents difficulties since there are a number of indexes which can be

used to determine homoplasy of characters and no criterion against which to judge them. Additionally there is no means of determining whether best, worst or mean fit of the character to the tree is most desirable (Swofford & Begle 1991). Because of these difficulties all characters in the *Hypenia* analysis were treated as unordered and unweighted.

iv) Characters in the Hyptidinae

Characters used in my cladistic analysis are based on the morphological observations outlined in chapter 2. As far as possible I have tried to use morphological characters which have well-defined discontinuities. Explicitly quantitative characters are minimised, but corolla length has been maintained as there is some discontinuity in the data (figure 5-3). Most of the characters rely on distinctions in morphology derived from differential changes in dimension and so are quantitative in the sense of Stevens (1991). All characters were unordered and unweighted. The following is an exposition of the characters used in the analysis with discussion of the nature of the variation that each one represents.

Stem

1. virgate stems - absent (0) / present (1).

Coded as a simple presence / absence character. The presence of virgate stems applies to those taxa with long, unbranched stems devoid of leaves.

2. upper internodes - with indumentum (0) / glabrous (1). This character has been coded in discrete states but shows some degree of continuous variability. Presence or absence of indumentum on the upper internodes applies to the inflorescence branches and to the stem between the lower inflorescence branch and the upper leaves.

3. Wax on internodes - absent (0) / present (1).

A simple presence / absence character.

4. Upper internodes - not swollen (0) / with globose swellings (1) / swollen along length (2).

This character is relatively easily coded into discrete states. Globose swellings occur at intervals along an internode compared to state 2 which consists of an enlargement of the entire internode. State 2 shows some gradation with state 0 but represents a distinct discontinuity and is easily coded.

Leaf

Leaf morphology and anatomy is extremely variable in the Hyptidinae but it is difficult to discern characters which have value above the species level (Rudall 1979 & 1980b). Most characters are autapomorphies or are present in widely divergent taxa and are apparently very homoplasious. The character selected is quantitative but was the only leaf character that

could be divided with an obvious discontinuity. Because of their extreme variability anatomical characters have not been included.

5. Petiole - sessile (< 2mm) (0) / petiolate (> = 2mm) (1).

Trichomes

There are many trichome types in the Hyptidinae (see chapter 2) and their distribution on the plant is very variable. It is possible to find many characters from the distribution and density of indumentum but they are very difficult to code in discrete states. The presence or absence of a particular type of trichome is a more satisfactory character but this limits the number of trichome characters.

6. Long setose trichomes on basal parts - absent (0) / present (1).

Setose trichomes are associated with the 'greasy pole' syndrome but are often found in the absence of other syndrome characters.

7. Branched trichomes - absent (0) / present (1).

Branched trichomes are limited in their distribution in the Hyptidinae and provide potentially valuable phylogenetic information.

Inflorescence

The inflorescence is an important source of characters for the classification of the Hyptidinae and help to resolve relationships both within *Hypenia* and in *Hyptis* and its satellite genera. The inflorescence can be broken down into a relatively large number of more or less easily scored characters which are outlined below.

Indeterminate axes

8. Indeterminate axis between cymose sub-units- expanded and visible (0) / contracted and obscured (1).

The indeterminate axis is variably developed and is most readily characterised by the extent of development between the cymes. This character is continuously variable but can be coded according to whether it can be easily seen between the cymes or if it is obscured by them. This is partially dependent on the size and structure of the cymes themselves but is easily observed and makes it possible to code the character into discrete states .

9. phyllomes - undifferentiated from vegetative leaves (0) / subulate or caducous (1).

The phyllomes are almost always reduced in size compared to the vegetative leaves in all species of the Hyptidinae. However, the character as it coded here is associated with a

change in morphology of the phyllomes. In this sense this is a qualitative character with discrete states.

Cymose subunits

Most of the characters in the cymose subunits are potentially continuously variable but in practise they display little intermediacy. Characters are associated with the extent of development of the axes.

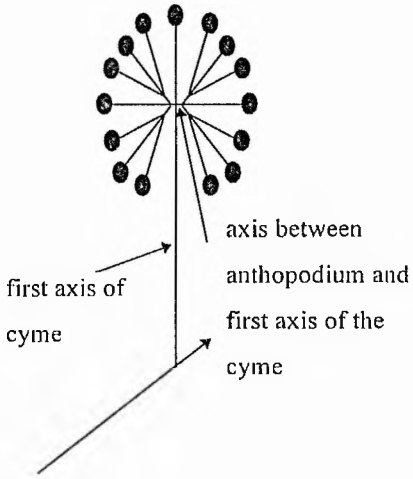
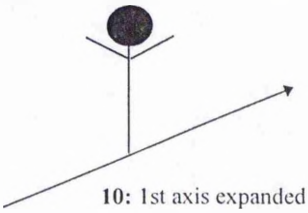


Figure 5-1: Parts of the cyme

10. 1st axis - reduced (0) / expanded (1).

Cymes are either sessile or borne on a well-developed first axis which is clearly visible and not obscured by the cyme.The length of the first axis is a continuous character but is coded in discrete states according to the relative position of the flowers. Introducing this discontinuity reflects the distinction between sessile cymes and pedunculate cymes.

1. *Hypenia*

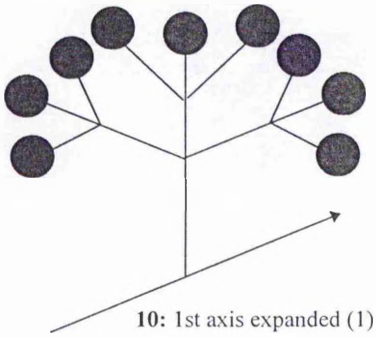


10: 1st axis expanded (2)

14: intermediate axes absent (0)

13: anthopodium reduced (1)

2. *Hyptidendron vepretorum*

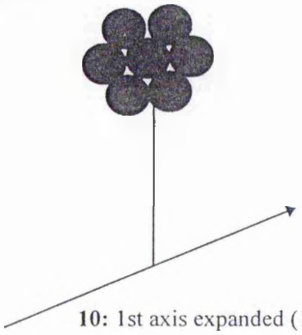


10: 1st axis expanded (1)

14: intermediate axes expanded (1)

13: anthopodium expanded (0)

3. *Hyptis rugosa*



10: 1st axis expanded (1)

14: intermediate axes reduced (1)

13: anthopodium reduced (1)

Figure 5-2: Examples of character coding applied to axes within cymes

See characters 10, 13 and 14 for further explanation

11. Cincinnate divisions - absent (0) / present (1).

This is a clearly defined discrete, qualitative character which is relatively easy to score.

12. Bracteoles - absent (0) / present (1).

The presence or absence of bracteoles is a discrete, qualitative character but as discussed in chapter 2 they can be variable within individual plants and are absent from some flowers of *Hypenia brachystachys* and *Hypenia densiflora*. However, they are scored as present if most flowers have bracteoles and they are absent only from terminal flowers in the cyme.

13. Anthopodium - expanded (0) / reduced (1).

Since the anthopodium is defined as the ultimate axis below the flower it must be present if flowers are present, hence it is not possible to code its absence. This is a potentially continuous character and is rather difficult to code with a convincing discontinuity. The character as it is applied here refers to the development of the anthopodium within the cyme

and is coded as expanded when it is clearly visible and reduced when it is obscured by the flowers.

14. Intermediate axes between 1st axis and anthopodium - absent (0) / expanded (1) / reduced (2).

This character is effectively coded in two parts, once as a presence / absence character and then with two potential states if the axis is present. There are no axes between the first axis of the cyme and the anthopodium if there is only a single flower, e.g. in *Eriope* or many species of *Hypenia,* which have bracteoles. In this case the first axis of the cyme corresponds to the propodium. If there are single-flowered cymes and no bracteoles, e.g. *Hyptis elegans,* then all the axes below the anthopodium are coded as absent.

Strictly speaking the transition between states one and two is continuously variable but it is relatively easy to code into discrete states which have a profound impact on cyme morpholgy. In species with capitulate cymes these axes are reduced, leading to the formation of the 'capitulum'. Expanded intermediate axes give rise to open cymes as found in *Hyptis tafallae* and *Hyptidendron* section *Umbellaria.* The number of axes between successive divisions varies, frequently between cymes on the same individual, eg. *H. brachystachys* and because of the difficulty of observation and its variability, has not been coded.

Flower

15. Orientation - flower not resupinate (0) / resupinate (1).

This is a simple, qualitative character. It can be difficult to ascertain the position of the flower when it is pressed on a herbarium sheet but those species coded in as resupinate have been observed in the field.

Calyx tube

16. Flowering calyx tube shape - cylindrical (0) / campanulate (1).

Although coded as a qualitative character with two discrete states there is potential overlap since the character is defined by botanical terminology. However, the two different terms refer to a combination of different quantitative variables which in combination can be readily coded as distinct shapes.

17. Flowering calyx tube - straight (0) / sigmoid (1).

A discrete, binary character.

18. Conspicuous hairs in mature calyx throat - absent (0) / present (1).

Conspicuous hairs are coded as present if they obscure the nutlets in the mature calyx and absent if the interior surface of the calyx is glabrous or sparsely hairy but not obscuring the nutlets. Hair density in the calyx is a continuous character and there is a degree of arbitrariness in the difference between the two states. However, the presence of dense hairs in the calyx throat has been used as an important character in *Eriope* and is relatively easy to score according to a more or less defined discontinuity.

19. Lobe length - < 1 mm long (0) / > 1 mm long (1).

This is an explicitly quantitative character but it can be scored unambiguously for all taxa included.

Fruiting calyx lobes

20. Lobe shape - deltoid (0) / aristulate (1).

The deltoid state refers to the development of triangular membrane from the medial nerve of the calyx lobe to the calyx tube. The aristulate condition refers to calyx lobes which lack this membrane development, although membrane is present. It is potentially continuous because the membrane is still present but problematic intermediates were not found in the taxa sampled.

21. Posterior lobe connation - three posterior lobes free at base (0) / three posterior lobes connate at base (1).

Although coded in two discrete states there is some continuous variation between the states. Most species in the Hyptidinae have a slightly assymetrical, gibbous calyx. In this case the three posterior lobes are arranged over the two anterior but there is little or no connation of the anterior lobes. Those species coded as lobes connate have the three anterior lobes connate for at least one quarter of their length.

22. Posterior lobe size - single posterior lobe same size as lateral posterior lobes and anterior lobes (0) / single posterior lobe expanded relative to other lobes (1).

Some species of *Eriope,* particularly in section *Eriope,* have a conspicuously expanded posterior lobe of the calyx and this character is included to provide resolution in *Eriope.*

23. Position of posterior lobes - straight (0) / reflexed (1).

This character is coded with two discrete states according to the position of the three posterior lobes relative to the calyx tube. In *Eriope* the posterior lobes show independent behaviour to that of the two anterior lobes, hence their coding as separate characters in this analysis.There is a degree of continuous variation between the states but assigning either state to individuals rarely presents a problem.

24. Position of anterior lobes - straight (0) / reflexed (1).

Coded in the same way as character 23.

Corolla

There are few truly qualitatively discrete characters to be found in the corolla, most variation is associated with relatively subtle changes in the relative size and shape of various, particularly the tube. Nevertheless there are some important characters with states which can be defined with convincing discontinuities.

25. Tube constriction over nutlets - absent (0) / present (1).

Most species of Hyptidinae have corolla tubes which are not constricted over the nutlets but either flare from the base or remain more or less the same diameter along their length. The corolla tube in some members of the Hyptidinae has a distinct constriction over the nutlets and in these taxa is coded as tube constriction present.

26. Tube - cylindrical (0) / campanulate (1).

The two states refer to the width of the corolla tube at its mouth. If the corolla tube is a more or less uniform cylinder along its length it is coded as cylindrical, if the corolla is very broad from just above the nutlets to its mouth it is coded as campanulate. There is a considerable discontinuity between the corolla shape of *Eriope* and *Hyptis* resulting from this change in the shape of the tube, some intermediacy is seen in the *Hyptidendron* in which some species (e.g. *H. canum*) have rather broad corolla mouths but which are basically cylindrical in shape and have been coded as such.

27. Colour - white (0) / pink, lilac, blue (1) / red or yellow (including shades of pink and orange)(2).

Corolla colour is very variable in the Hyptidinae and coding all the colours under one character produces a single multi-state character. The delimitation of states was problematic and probably remains somewhat arbitrary. However, many taxa show colour combinations in the corolla and the states delimited are a reflection of the commonest combinations observed. White may be regarded as an absence since it denotes lack of pigment but it was frequently found in combination, especially with lilac, pink or blue, in which case the colour was coded as uncertain. Red and yellow were scored as a single colour since within section *Hypenia* subsection *Ellipticae,* where yellow is commonly present, many species have both yellow and red pigments in the corolla, the proportion determining the exact shade. Many individuals have flowers of different colours, from red to yellow and shades in-between and the corolla frequently changes colour through its lifetime. It is therefore impossible to discern a discontinuity between the two colour states.

28. Tube length - <= 7mm (0) / 7.5 - 18 mm (1) / > =18.5 mm (2).

An explicitly quantitative character but there is considerable information in corolla tube length within *Hypenia* and the states used have been selected as far as possible to eliminate intra-taxon variation within this group. The corolla tube length was measured on 50 specimens of *Hypenia* from all species and plotted in figure 5-3. It is possible to discern two discontinuities in the data, marked on the figure, although the seven mm cut-off is somewhat arbitrary and may disappear if further individuals are included. There is a clear discontinuity between 18 and 23 mm with no ambiguity present in defining this state. Outside of *Hypenia* there is virtually no information in the character delimited by these states since more or less all species have corollas less than 7 mm long.

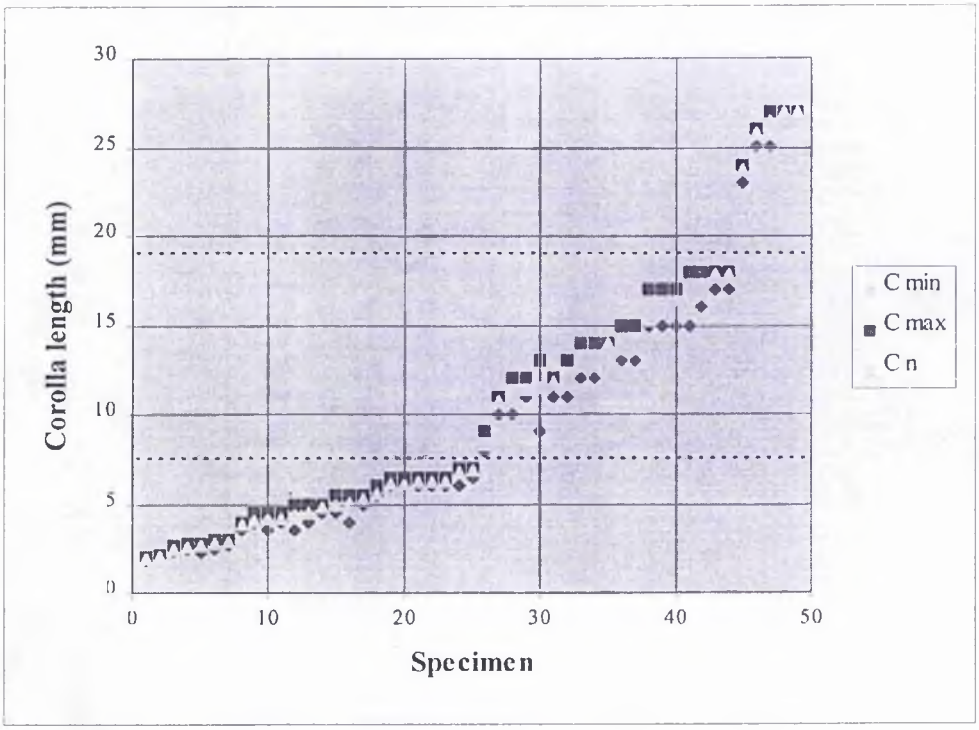


Figure 5-3: Corolla length in *Hypenia* specimens

See text for explanation

29. Internal projections in corolla tube - absent (0) / present (1).

This refers to the internal projections which effectively divide the corolla along its length and are a conspicuous feature of *Hypenia* section *Hypenia.* It is easily coded as a discrete presence / absence character.

Androecium

30. Stamen filaments - glabrous (0) / hairy (1).

Most species of Hyptidinae and *Isodon* have hairy stamen filaments and this is a useful character to define the ingroup and provide some resolution in the outgroup.

Gynoecium

31. stylopodium - absent (0) / present (1).

Harley (1976, 1988a) treats the stylopodium as a qualitative character and recognises two variants if the stylopodium is present, stylopodium long and stylopodium short. Rudall (1981) points out that the abscission zone represented by the stylopodium is present in all Labiatae and only varies in its position. Hence, it is a continuous character and is defined simply by the presence of the abscission zone in relation to the nutlets, i.e. if it is present above the top of the nutlets the stylopodium is present and if it present below the level of the nutlets it is absent. There is an obvious discontinuity in the position of the abscission zone in relation to the nutlets and the stylopodium is scored in this analysis as a simple presence/absence character.

C. Selection of taxa to include in the analysis

The taxonomic problems outlined in chapter 1 provided the focus of the analysis and were used as the basis for the selection of taxa to include in the analysis. The problems were:-

1. The relationship between *Hypenia* and the other genera of the Hyptidinae.
2. Infra-generic classification of *Hypenia.*
3. Species delimitation in *Hypenia,* particularly in the 'macrantha complex'.

The first two problems were concerned with constructing the most appropriate classification for groups of species. To solve these problems clades need to be indentified to be used as the basis of a classification. The third problem is concerned with defininig taxa and is about searching for differences between species. It is therefore not suitable for cladistic analysis.

Sampling of taxa was based on selecting species from all the currently recognised sections (see Harley, appendix I) in *Hypenia* and *Eriope* in an attempt to answer the first two questions. It was not considered appropriate to include all species within the 'macrantha complex' because differentiation between many of them was poor. In this case the relationships may still be partially tokogenetic and not appropriate for cladistic analysis. In addition the characters which distinguished closely related species were autapomorphies and provided no information about groupings. *H. subrosea* was included because its white

flowers are anomalous in the 'macrantha complex'. All other species of *Hypenia* outside of the 'macrantha complex' were sampled except for *H. concinna* which is only represented by one herbarium specimen and is possibly conspecific with *H. brachystachys*.

The outgroup was selected from *Hyptis* and *Hyptidendron*. *Hyptis hagei* and *Hyptis asperifolia* were considered by Harley (pers. comm.) to be putatively basal in *Hyptis*, and possibly the Hyptidinae as a whole and were included in the analysis. In addition *Hyptidendron* and *H. vepretorum* were included since *Hyptidendron* was considered to be closely related to *Hypenia* and *Eriope* (Harley 1976, 1988a).

Table 5-1: Taxa included in the morphological analysis

Genus	**Section**	**Species**
Hypenia		*H. vitifolia*
		H. salzmannii
		H. micrantha
		H. irregularis
		H. gracilis
	Densiflora	*H. densiflora*
		H. brachystachys
	Hypenia subsection *Hypenia*	*H. aristulata*
		H. reticulata
		H. subrosea
	Hypenia subsection *Ellipticae*	*H. macrantha*
		H. crispata
Eriope	*Eriope*	*E. hypenioides*
		E. glandulosa
	Nudicalyx	*E. salviifolia*
		E. sincorana
Outgroup		
Hyptidendron	*Hyptidendron*	*H. canum*
	Umbellaria	*H. vepretorum*
Hyptis	*Subumbellaria*	*H. asperifolia*
		H. hagei
	Cyanocephalus	*H. rugosa*

i) Morphological data matrix

The data matrix of character distributions of the selected taxa is shown in appendix III.

D. Results of the morphological analyses

The analysis was conducted in PAUP version 3.1.1 (Swofford 1993) using the heuristic search with default options, i.e. saving trees of minimal length only; collapse zero length branches; stepwise addition to find starting trees and swap on minimal trees only; random addition sequence; and branch swapping using Tree Bisection and Reconnection (TBR). 348 trees were generated, 64 steps long, CI = 0.516, RI = 0.735, RC = 0.379.

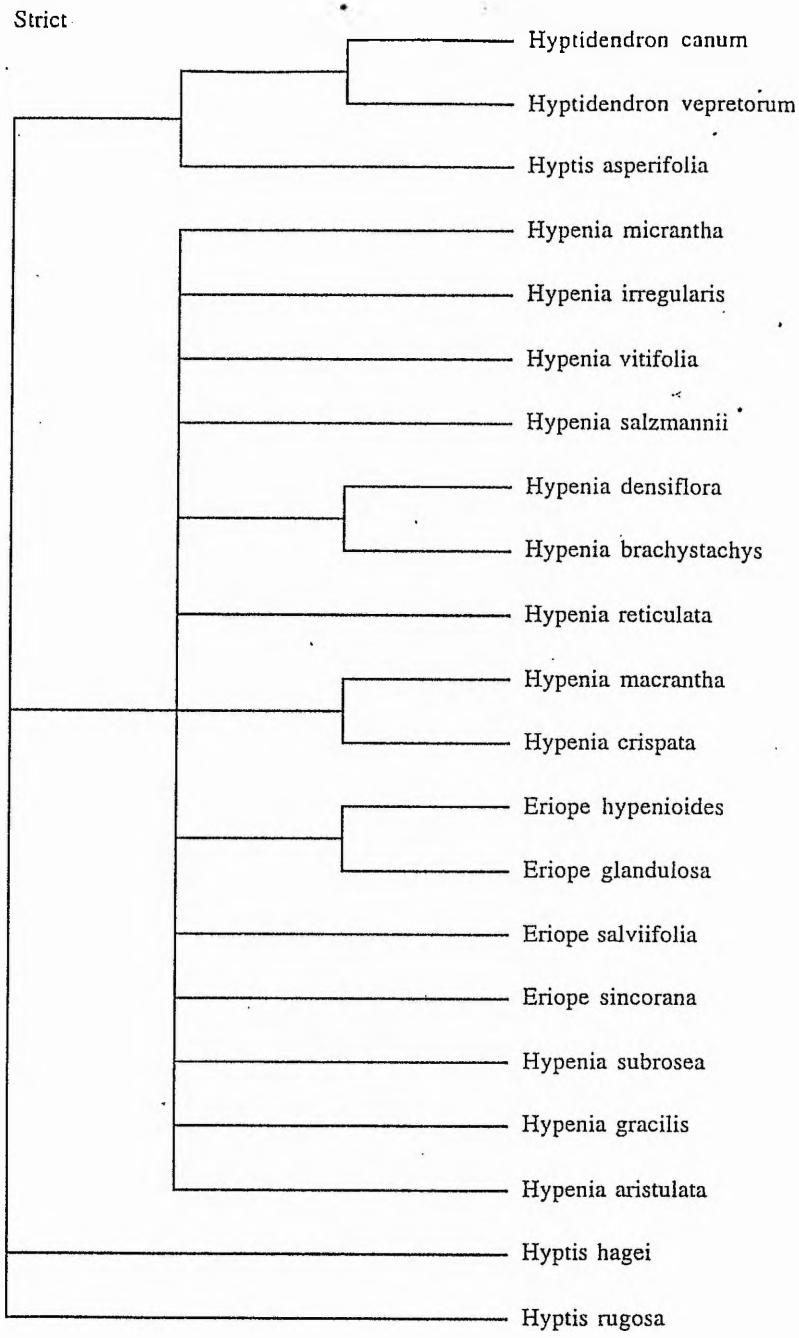


Figure 5-4: Strict consensus of 348 trees produced by the morphological cladistic analysis

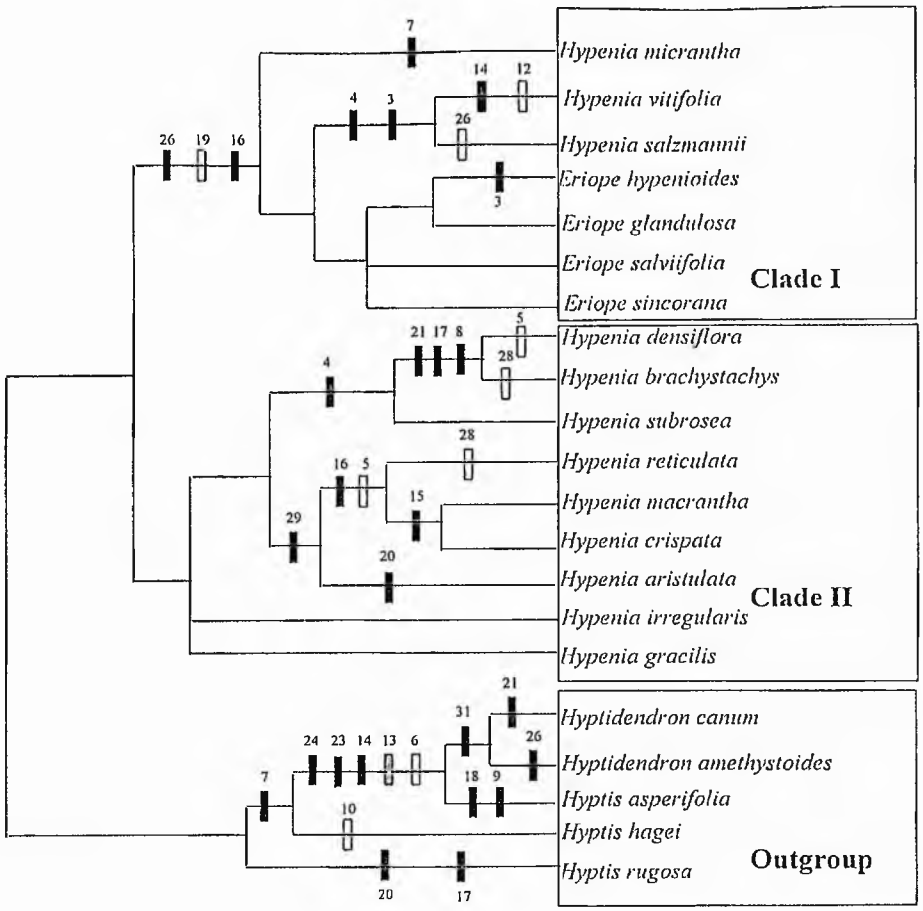


Figure 5-5: Tree A: morphological cladistic analysis

Transformations are coded as follws:

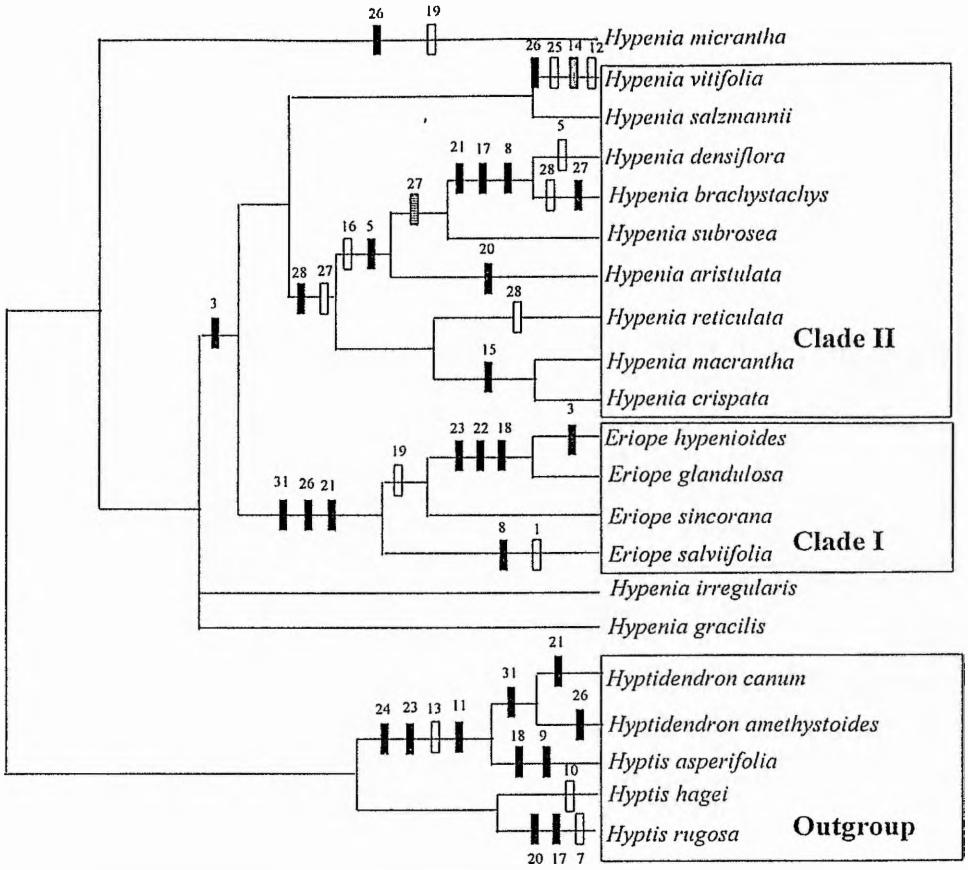


Figure 5-6: Tree B: morphological cladistic analysis

Transformations are coded as follows: 0 > 1; 1 > 0; 0 > 2; 1 > 2; 2 > 0; 2 > 1; 1>2

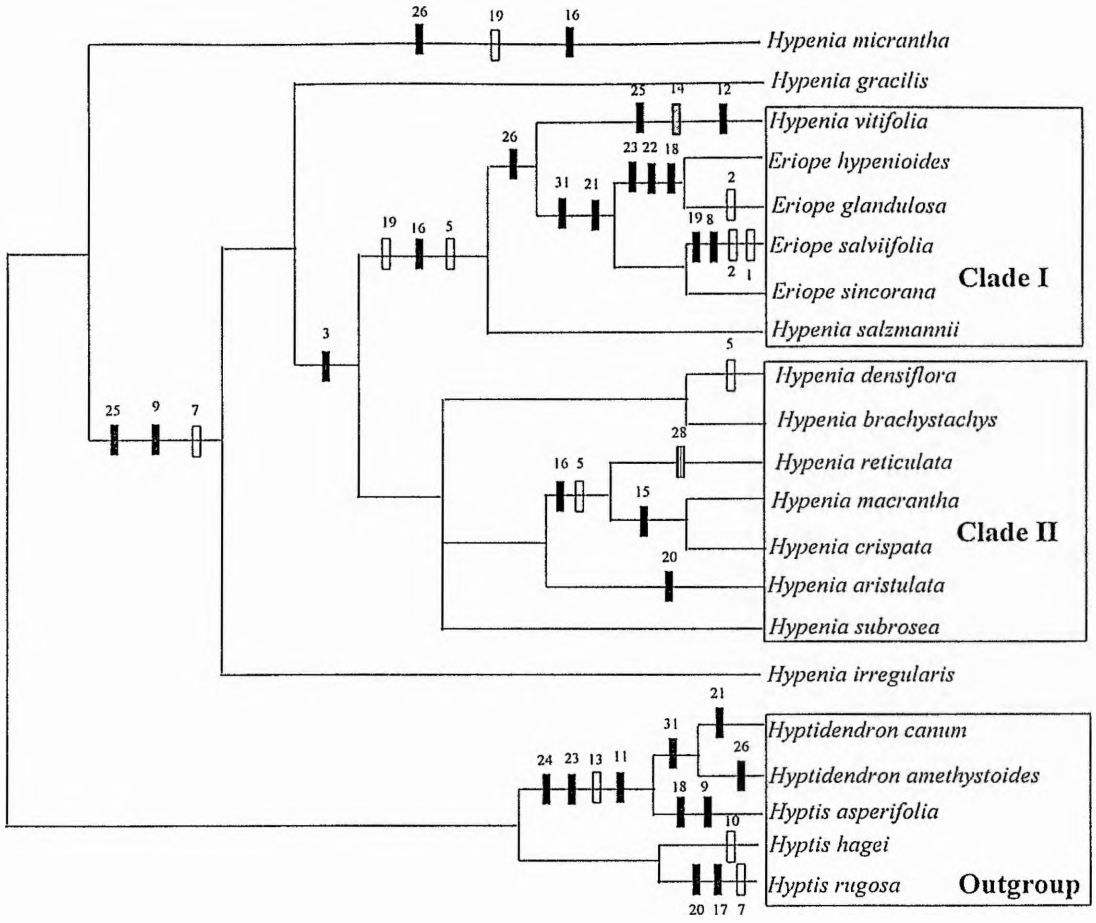


Figure 5-7: Tree C: morphological cladistic analysis

Transformations are as follows:

0 > 1	2 > 0
1 > 0	2 > 1
0 > 2	1>2
1 > 2	

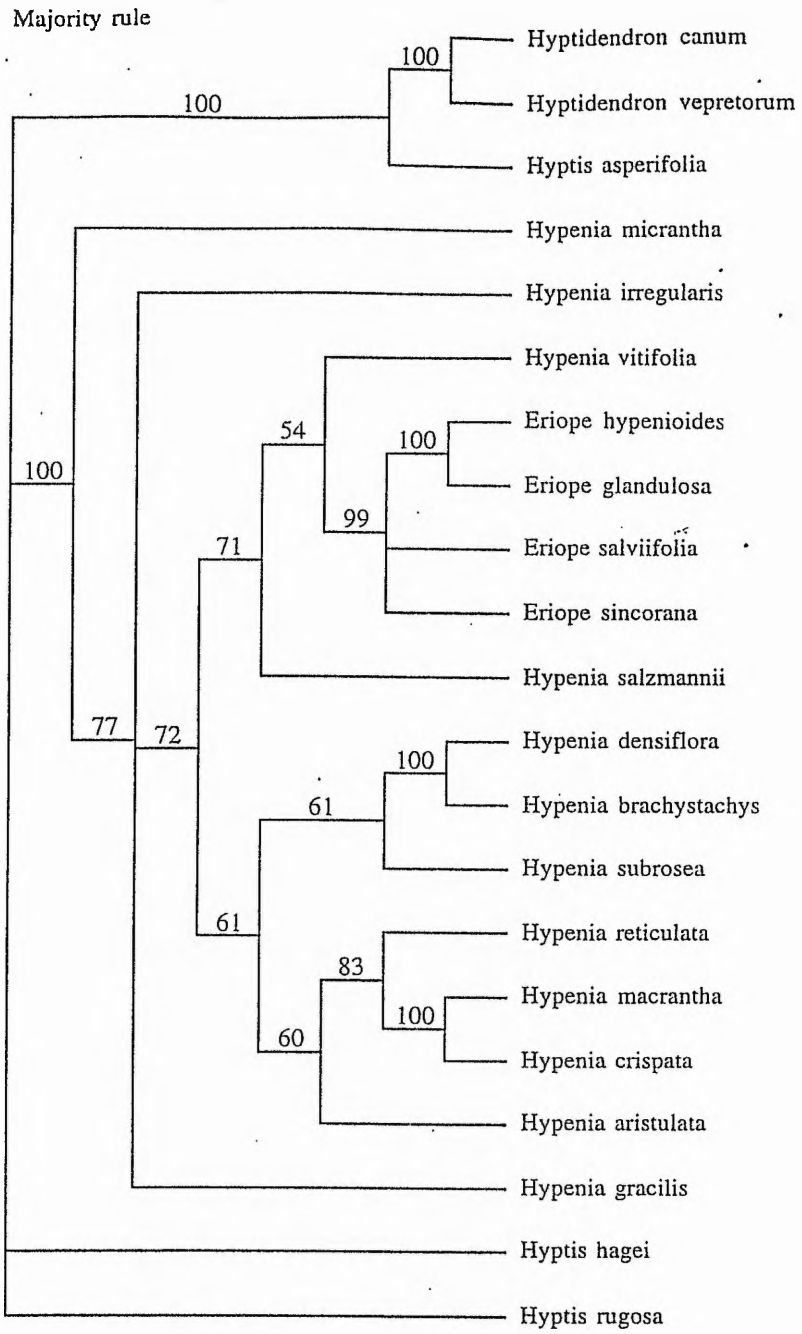


Figure 5-8: Majority rule consensus of the morphological analysis

Numbers on the branches indicated the percentage of trees in which the component is present.

E. Discussion of the morphological analysis

The strict consensus is shown in figure 5-4 (the semistrict was identical). The outgroup showed some resolution with *Hyptis asperifolia* always in the same clade as the two

Hyptidendron species. In the ingroup there were three species pairs: *Hypenia densiflora* and *H.brachystachys*; *H. macrantha* and *H. crispata,* the two resupinate-flowered members of the 'macrantha complex'; and *Eriope hypenioides* and *E. glandulosa.* No other resolution was present.

The cladograms shown in figures 5-5, (tree A) 5-6 (tree B) and 5-7 (tree C) were selected to show variable positions of the taxa which contributed most to the lack of resolution in the strict consensus. The basic pattern of the three cladograms is similar. There are two main clades, one composed of *Hypenia* species (clade II) and the other containing all four species of *Eriope sensu stricto* (clade I). However, there are significant differences in the position of several taxa between the three cladograms. The cladograms illustrated indicate that the lack of resolution in the strict consensus is mostly associated with the position of six species: *H. micrantha, H. irregularis, H. gracilis, H. salzmannii, H. vitifolia* and *H. subrosea. H. micrantha* moves from being sister to the whole ingroup (trees B and C) to sister to a clade composed of *H. vitifolia, H. salzmannii* and the *Eriope* species (clade 1, tree A). Similarly, *H. irregularis* and *H. gracilis* move from sister positions outside the two main clades (trees B and C) into the *Hypenia* clade (clade II, tree A). *H. vitifolia* and *H. salzmannii* move between the two main clades and either occur with *Eriope* species (trees A and C) or with *Hypenia* (tree B). The two species also shift position in relation to each other. In trees A and B they occur as sister groups on the same branch but in tree C they are successive sister species to *Eriope.* In the *Hypenia* clade *H. subrosea* is either sister to all the 'macrantha complex' species and *H. densiflora* and *H. brachystachys* or is embedded in a clade comprised of the latter two species and *H. aristulata.*

The characters which change unambiguously on each branch are shown on all the cladograms and the character changes associated with the most variably positioned taxa are discussed.

i) Tree A

In tree A, clade I contains the four *Eriope* species, *H. micrantha, H. vitifolia* and *H. salzmannii* and is supported by funnel-shaped corolla, campanulate calyx and short calyx lobes. *H. micrantha* is separated from the rest of the clade by its branched trichomes and *H. vitifolia* and *H. salzmannii* are supported together by the presence of wax and globose swellings on the upper internodes. The presence of wax is homoplasious in the clade however, since this character state is also present in *E. hypenioides.*

Clade II is not supported by any unambiguous characters and the position of *H. irregularis* and *H. gracilis* is unresolved in relation to the rest of the clade. The remainder of the clade splits into two. *H. densiflora* and *H. brachystachys* are sister to *H. subrosea* in one clade supported by globose swellings on the internodes. The other clade consists of the rest of the 'macrantha complex' which is supported by the presence of internal projections in the corolla. Within this clade the *H. macrantha* and *H. reticulata* are supported together by the resupination of the corolla.

The outgroup is fully resolved with *Hyptis rugosa* as sister to the remaining four species and *Hyptis hagei* between *H. rugosa* and a clade composed of *Hyptis asperifolia* and the two *Hyptidendron* species. This latter clade is supported by five characters, deflexed anterior and posterior calyx lobes, presence of cincinnate divisions in the cymes, expanded anthopodia and internal cyme axes reduced.

ii) Tree B

In tree B, *H. micrantha, H. irregularis* and *H. gracilis* are placed as sister species to the rest of the ingroup. Clade I is composed of the *Eriope* species only and is supported by connate calyx lobes, campanulate calyx tube and presence of the stylopodium. Clade II consists of *Hypenia* species and is supported by the presence of wax. Within clade II *H. vitifolia* and *H. salzmannii* are sister species on a branch supported by small (<1mm) flowering calyx lobes. The rest of *Hypenia* is supported by the red or yellow corolla and between 7.5 and 18 mm long. *H. densiflora* and *H. brachystachys* are sister species nested within the 'macrantha complex' with *H. subrosea.* The three species are supported together by the presence of a white corolla. *H. densiflora* and *H. brachystachys* are supported by connate calyx lobes, sigmoid flowering calyx and contracted indeterminate axes.

The outgroup is fully resolved with *Hyptis rugosa* and *Hyptis hagei* on a branch which is not supported by any characters. The other three taxa occur in the same position as tree A.

iii) Tree C

In tree C *H. micrantha* is in the same position as in tree B and *H. irregularis* and *H. gracilis* are in unresolved positions outside of the two main clades. Clade I includes *Eriope* plus *H. salzmannii* and *H. vitifolia* and is supported by petiolate leaves, campanulate calyx and short flowering calyx lobes. In this cladogram the *Eriope* clade within clade I is supported by connate calyx lobes and presence of the stylopodium.

The position of *H. densiflora* and *H. brachystachys* as well as *H. subrosea* is unresolved in relation to the 'macrantha complex. As in all the other trees *H. macrantha* and *H. crispata* are sister species supported together by their resupinate corolla. The outgroup is identical to tree B.

F. Implications of the morphological analysis

As discussed above, the detailed evolutionary implications of the morphological analysis are ambiguous. The lack of resolution in the strict consensus indicates the variability in the trees generated and the lack of agreement between them. However, there are some important features of the morphological analysis which have important implications on taxonomy The monophyly of the ingroup, i.e. *Hypenia* and *Eriope* is supported but there are no characters which unambiguously support the entire clade. In addition *Eriope* is nested within *Hypenia* in all the cladograms which makes *Hypenia sensu* Harley (1988a) paraphyletic. *Eriope* appears to be a well supported group characterised by the presence of stylopodium but its exact position within *Hypenia* is still unclear. However, *H. salzmannii* and *H. vitifolia* frequently occur in the same clade (figure 5-8). The relationship between *H. salzmannii* and *H. vitifolia* themselves is unresolved although in all the trees illustrated they occur either on the same branch or as sister species. *H. irregularis* and *H. gracilis* are relatively isolated and they are not placed on the same branch in any of the trees.

In some of the trees the position of *H. subrosea* as sister to *H. densiflora* and *H. brachystachys* makes the rest of the 'macrantha complex' (see chapter 1) paraphyletic. *H. densiflora* and *H. brachystachys* are sister species in all the analyses and are supported by several characters including their contracted indeterminate axes and connate calyx lobes. The resupination of the corolla in *H. macrantha* and *H. crispata* appears to confer considerable support for the relationship between these two species. The position of the two *Hyptidendron* species as sister to *Hyptis asperifolia* is consistent in all the trees and is supported by the position of the calyx lobes, branched hairs and cincinnate divisions in the cymes.

Lack of resolution in the strict consensus was a result of homoplasy in a number of characters. The size of the calyx lobes, presence of wax, calyx lobe connation and petiolate leaves are all characters which show reversal or parallelism in different parts of the trees. Non-homoplasious characters include the presence of the stylopodium and the resupinate corolla.

Homoplasious characters are a major problem in morphological analyses, largely I feel because of the difficulties of delimiting the characters outlined in the introduction to this chapter. In many cases morphological characters are a complex expression of genetic and phenotypic variation which defy accurate representation. For this reason molecular characters which can more or less be reduced to simple changes in bases at specified locations offers a major advance in phylogenetic techniques. A molecular analysis of *Hypenia* is presented in the following chapter.

Chapter 6: Molecular analysis

A. Introduction to the chapter

Molecular methods have recently become one of the most widely-used and powerful tools available to systematists. Sequencing of the Internal Transcribed Spacer (ITS) region of nuclear ribosomal DNA was used in this study to provide an additional source of characters to investigate the phylogenetic relationships of the supra-specific taxonomy of *Hypenia.* The theoretical background to molecular techniques is presented followed by the methods used. Results are presented and discussed in the final part of the chapter.

B. Introduction to molecular methods in systematics

i) Why use molecular genetic markers ?

It is fundamental to systematics that only genetically inherited traits can inform studies of phylogeny. Most characters of the organism, morphological or behavioural, are genetically controlled and can be used to construct a phylogeny. However, direct analysis of the genetic material has a considerable advantage over phenotypic expressions of the genotype in that the genetic basis of variation and modes of transmission can be explicitly stated. The genetic basis of most morphological characters is rarely known in sufficient detail to allow for confident assessment of their information content and classic studies have highlighted the phenotypic plasticity of genetically identical plants raised in different environments (Clausen et al. 1940). Phenotypic variation of this sort can be misleading if characters affected by it are applied to phylogeny reconstruction.

Before molecular methods were introduced genetic investigation in plants was limited to a restricted number of genes in a few cultivated species which were easily crossed, have short life cycles and well known mode of inheritance. Since the introduction of molecular techniques virtually any organism and a wide range of genes can be subject to levels of investigation which were previously unknown. This has enabled the entire range of biological diversity to be opened up to phylogenetic assessment based on direct analysis of a potentially enormous source of variation within the genome (Avise 1994).

With the increasing ease of applying molecular techniques to a wide range of organisms and the enormous amount of potential variation that can be discovered, coupled with an

increasing understanding of the nature of evolution of the genome, molecular methods are becoming a standard part of the systematist's repertoire. Developments have been rapid and the range of techniques available is now very wide. However one method, DNA sequencing, now dominates molecular phylogenetic studies. The reasons for this, and the reasons for its adoption in my study, are outlined below.

ii) Non-sequencing molecular methods

Enzyme electrophoresis was a commonly used method to assess molecular variation and is still employed in the field of population genetics to study infraspecific genetic structure (Hamrick & Murawski 1990, Hamrick et al. 1992). Interpretation of isozyme bands for systematic purposes is problematic above the species level and direct analysis of the genotype, i.e. the DNA itself, rather than the phenotype, e.g. the enzyme products of DNA, has been increasingly used by systematists.

A commonly used non-sequencing method of DNA analysis was restriction site mapping of the chloroplast genome. Restriction site mapping involves the use of restriction enzymes to give variably-sized fragments of DNA which can be separated by gel electrophoresis and visualised. Variation among individuals can result from base substitutions within cleavage sites, additions or deletions of DNA, or sequence rearrangements. Each source of variation produces characteristic banding changes. These fragments or restriction fragment length polymorphisms (RFLPs) can be interpreted as discrete characters and have been commonly used in phylogenetic studies of interspecific variation (see Palmer et al. 1988). In a number of families, notably the Asteraceae, the detection by hybridisation of restriction fragments can reveal structural features of the chloroplast genome which are a useful source of characters for generic level analyses (Downie & Palmer 1992).

iii) Sequencing

Direct sequencing of selected parts of the genome has rapidly become the most popular tool available to molecular systematists. This popularity is due to a combination of factors: nucleotides are the most basic unit of information in organisms; the data generated by nucleotide sequences is relatively easy to analyse and model; and the potential amount of information is enormous (Hillis et al. 1996). The predominance of sequencing in recent molecular systematic studies has been facilitated by a number of technological advances, in particular the polymerase chain reaction which is now used as a standard reaction to amplify the required DNA sequence.

iv) Choosing an appropriate part of the genome for phylogenetic analysis

Levels of diversity are not consistent across the genome since not all parts are equally variable (Wolfe et al. 1987). Some areas, which may code for essential proteins, change only very slowly as most changes will adversely affect protein production and will not be maintained in populations. By contrast, changes in other areas will have a less dramatic effect and are more likely to survive in a population and be passed on to later generations. Different parts of the genome will therefore provide information at different levels and selecting an area which displays variation at the appropriate level in the taxonomic hierarchy is essential for systematic studies.

Chloroplast DNA has been frequently used in plants for systematic studies. Chloroplast genes evolve in much the same way as nuclear genes but with a considerably reduced mutation rate (Clegg & Zurawski 1992). One of the most commonly used chloroplast sequences is the gene that codes for the large sub-unit of rubisco (*rbcL*) which has ca. 1470 base pairs and thus has the potential to generate very large amounts of phylogenetic information. In practice genes like *rbcL* have highly conservative regions associated with the essential role that the gene plays in photosynthesis. Most changes in the protein-coding region of such a gene will be lethal and most of the variation in *rbcL* occurs in non-coding areas. Hence the actual number of characters for most genes is considerably lower than the total number of nucleotides. *RbcL* shows low rates of change between families or even between taxa above the order level and can be compared across all flowering plants. It has been widely exploited for high-level systematic studies (Chase et al. 1993).

Other parts of the genome, especially outside of the chloroplast, show less conservative rates of change. For example the internal transcribed spacers of the nuclear ribosomal region (ITS) is more useful than *rbc*L in providing information on relationships at lower levels in the taxonomic hierarchy. This is associated with its structural rather than direct coding function (Baldwin et al. 1995).

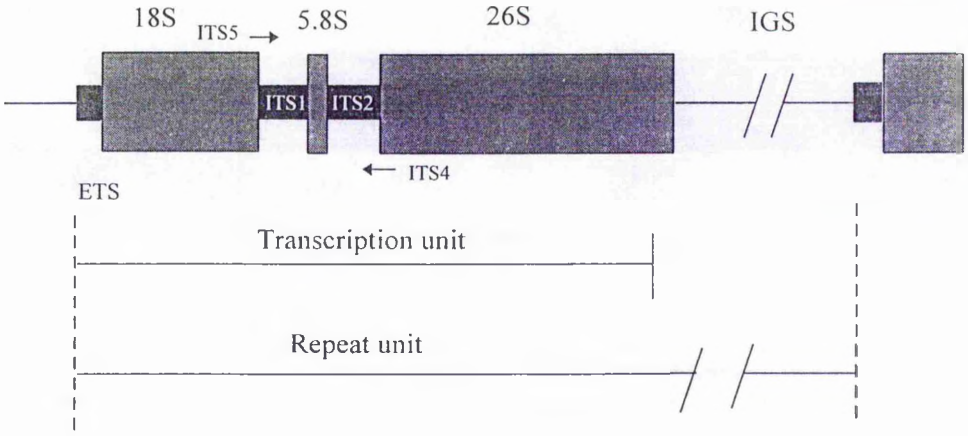
The nuclear genome has several advantages in comparison to chloroplast DNA (cpDNA). Chloroplast DNA is cytoplasmically inherited from one parent and can be effected by hybridisation and introgression or lineage sorting. Incongruencies between phylogenies generated by cpDNA and other parts of the genome or morphology can often result from these factors (Doyle 1992). These problems can be avoided with the use of nuclear DNA and

its use in phylogenetic studies can enable a more accurate reconstruction of the evolutionary history of plant taxa.

v) Internal transcribed spacers of nuclear ribosomal DNA

The internally transcribed spacer (ITS) region of ribosomal nuclear DNA has been identified as a part of the nuclear genome that can be applied to phylogenetic studies and has been used in plants for phylogenetic studies at the species level (Baldwin 1992, Baldwin et al.1995). ITS was chosen for this study as a region likely to generate the appropriate amount of variation to elucidate relationships within *Hypenia* and its close relatives.

The genes that code for ribosomal RNA (rRNA) are repeated many times in nuclear DNA and can account for a large proportion of the genome. Each rDNA unit consists of a transcribed region separated from the next repeat unit by an intergenic spacer (IGS). The transcribed region consists of an external transcribed spacer (ETS), the 18S gene, an internal transcribed spacer (ITS-1), the 5.8S gene, a second ITS (ITS-2) and the 26S gene (see figure 6-1).



Structure of typical plant rDNA, after Hamby & Zimmer 1992.

Figure 6-1: Diagramatic representation of the ITS region

Two primers (see PCR reaction below), ITS4 and ITS5, are used to identify the entire region prior to amplification by PCR. ITS4 is complimentary to the part of the sequence flanking the ITS-2 region on the end of the 26s subunit and ITS5 is located on the opposite strand on

the 18s subunit next to the ITS-1 region. Using these two primers both ITS regions and the intervening 5.8S gene are amplified.

Practical advantages and limitations of sequencing the ITS region are becoming apparent as it is applied in an increasing number of studies (Sang et al. 1994, Savard et al. 1993) and have been comprehensively reviewed by Baldwin et al. (1995). One important feature of the ITS region is the presence of multiple copies in the genome. This increases the chances of amplification of complete sequences if the DNA is degraded and minimises the risks of amplification of contaminating sequences. However, associated with multiple copies is the risk of different copies from different chromosomes having divergent sequences. Nevertheless there is usually a high degree of sequence agreement between multiple copies from the same individual and from the same taxon (Baldwin et al. 1995).

The entire ITS region is relatively short (about 700 nucleotides) and ITS-1 and ITS-2 are both less than 300 base pairs long (ITS-1: 187 to 298 bp and ITS-2: 187 to 252 bp) and the 5.8S subunit is almost invariably 163 or 164 bp long (Baldwin et al. 1995). The small size of the region increases the chance of complete sequences being present in degraded DNA and enables the possibility of amplification of the ITS region from herbarium specimens.

ITS-1 and ITS-2 show similar levels of divergence but in studies in the Compositae ITS-1 was found to include more potentially informative characters than ITS-2 (Baldwin 1992, Baldwin et al. 1995). Trees generated from combined information from both ITS-1 and ITS-2 show more complete and robust resolution than trees generated from each region individually however and Baldwin et al. (1995) recommend the use of both regions together. The 5.8S region adds little to the analysis because of its invariability.

Alignment of ITS sequences can be complicated by insertion or deltion (indel) mutations which require the insertion of gaps to assure the homology of nucleotides at each position along the sequence. Gaps can be treated either as missing data, as a fifth character state or as an additional presence/absence character with sequence gaps treated as missing data. However, the approach adopted makes little difference to the resultant ITS tree topologies (Baldwin et al. 1995).

The level of divergence of ITS is variable but has been found to be too high to allow for reliable comparison at the family level in the Compositae (Baldwin 1992, Kim & Jansen

1994) and several other families including Rosaceae, Saxifragaceae and Polemoniaceae (see Balwin et al. 1995).In these groups ITS has been found most appropriate for resolving relationships within genera and among closely related genera. There seems to be some correlation with the rate of ITS evolution and life form, ancient woody groups showing much lower rates of change relative to herbaceous taxa of recent origin (Baldwin et al. 1995).

In the Asteraceae, ITS provided strong evidence for the relationship between the Hawaiian silverswords and a group of California tarweeds (Baldwin 1992). Also in the silversword alliance, ITS data supports relationships which conflict with the cpDNA phylogeny but correlate well with evolutionary hypotheses generated from chromosomal evidence (Baldwin 1992). ITS has also been used to improve the resolution of relationships in species groups by helping to identify cryptic clades in *Astragalus* (Fabaceae) (Wojciechowski et al. 1993) and has been used in *Amelanchier* (Rosaceae) to identify the parents of a hybrid taxon (Campbell et al. 1993). So far there have been few studies using ITS in the Labiatae but work in *Lavandula* shows good resolution between species within the genus and support for a reappraisal of infrageneric boundaries (Upson 1997, unpublished PhD thesis).

ITS was a good candidate for sequencing in order to improve resolution or confirm relationships within *Hypenia* and its relatives because of its established use for these purposes in other groups. Studies in the Compositae have found appropriate levels of variation in herbaceous genera (Baldwin 1992). *Hypenia* and its relatives are a similarly mostly herbaceous group which would be expected to show similar levels of divergence.

C. Technical aspects of DNA sequencing

There is a series of standard technical procedures required to undertake sequencing projects which are outlined below.

i) Polymerase Chain Reaction (PCR)

Until the development of the polymerase chain reaction (PCR) in the late 1980s sequencing was limited by the necessity for access to relatively large amounts of fresh material and complex cloning procedures. These difficulties can now be overcome with PCR in a single reaction and using very small amounts of material. The range of potential source material is remarkable and herbarium sheets or even fossils can be used, although silica-dried or fresh plant tissue is preferred.

The PCR reaction is summarised in 6-2 and described below. The reaction is based on repeated cycles which start by heating double-stranded template DNA and denaturing the molecule into single strands. The reaction mix contains a DNA polymerase, commonly *Taq*, an enzyme isolated from a bacterium (*Thermus aquaticus*) which is able to function at very high temperatures. Polymerases like *Taq* catalyse the construction of DNA from deoxynucleotide triphosphates (dNTPs) using single-stranded DNA as a template. The desired piece of DNA to be amplified is identified by a short segment of DNA, the primer, which is compatible with a known segment on the target DNA. After denaturing the DNA by heating the reaction cools and the primer sequences anneal to the complementary part of the single-stranded DNA.

The construction of DNA by polymerases is unidirectional. Sequences are characterised by a 3' end from which nucleotides can be added until a 5' end terminates the sequence. The polymerase builds the second strand from the 3' end of the primer using the dNTPs in the reaction mix. For each sequence to be amplified there are two primers which anneal on opposite strands and at each end of the target DNA. The polymerase then catalyses the construction of lengths of DNA which start with the complementary primer sequence. As the reaction proceeds most of the copies produced are of the length of the DNA between the primers. Repeated cycling of the polymerase chain reaction produces a high concentration of target DNA which is suitable for sequencing.

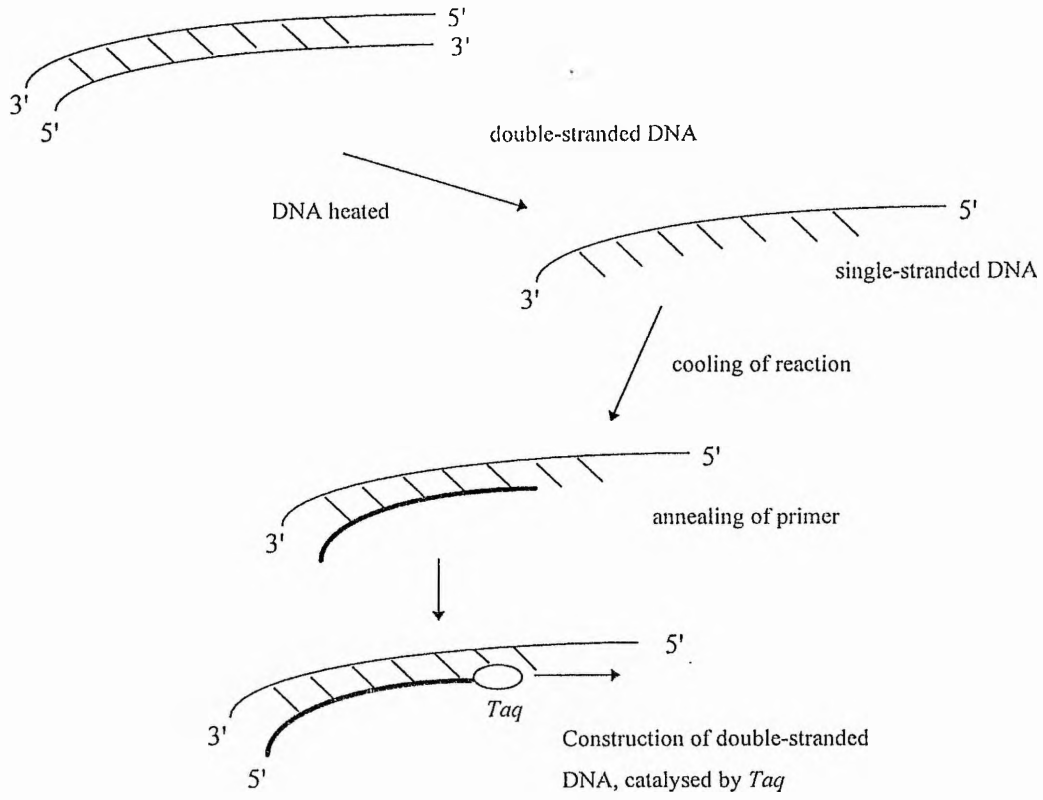


Figure 6-2: Schematic representation of the PCR reaction

See text for explanation

Mg^{2+} is a necessary cofactor for *Taq* function and MgCl is always part of the PCR mixture. Substances such as bovine serum albumin (BSA) are thought to stabilise the enzyme and are frequently added to enhance the efficiency of the reaction.

An important consideration when amplifying DNA by the polymerase chain reaction is the possibility of contamination. In theory the target sequence can be amplified from a single piece of DNA and if the primer binds to a piece of DNA from a source other than the required one it could amplify sufficiently to dominate the sample. For this reason the target DNA needs to be in the reaction mixture at sufficient concentration to reduce the random possibility of initial amplification of foreign material.

ii) Automated sequencing

Sequencing methods have developed rapidly over the last 10 years and the technology required to apply them has become more accessible, contributing to the massive increase in data now available. The procedure which is now becoming more or less standard, and the one used in this project, is automated sequencing.

Sequence products are generated during a cycle sequencing reaction based on the Sanger Dideoxy method (Sanger et al. 1977). The concentrated target DNA is produced by PCR and denatured by heating to produce single-stranded DNA. The primer (the same one as used in the initial PCR reaction) anneals to the single-stranded target sample and DNA synthesis is catalysed by a thermostable DNA polymerase (e.g. *Taq*). From the 3' end of the primer *Taq* adds dNTPs in the same way as described for the PCR, using the target DNA as a template. Cycle sequencing reactions differ from PCR in having four dideoxynucleotides, ddNTPs (ddATP, ddCTP, ddGTP, ddTTP), each of which is labelled with a different coloured fluorescent dye, present in the reaction mixture. On some strands in the reaction a given ddNTP will be incorporated into the growing strand, which because it lacks a 3' end, stops extension of the sequence and labels the last nucleotide in the strand. Successive cycles of denaturation, annealing and synthesis result in amplification of labelled fragments of DNA varying in length from one nucleotide to the maximum number of nucleotides in the required sequence.

In an automated sequencer the labelled fragments are separated by electrophoresis and detected as they pass a stationary laser. Because the fragments are labelled with different colours and can be separately visualised, the entire sequence can be run in one lane, allowing for up to 36 complete sequences to be run on one gel. The sequence is recorded directly into a computer and can then be interpreted using the appropriate software. Two sequences are generated for each DNA segment investigated, using primers from each end of the segment and running in opposite directions. This minimises errors generated as the sequence quality degenerates towards the end of the run and becomes difficult to interpret. The two sequences are assembled by computer and edited manually to minimise ambiguities.

Once the data has been downloaded onto the computer the two sequences, one from each primer, need to be edited and assembled. The complete sequence can then be transferred into

a matrix where it needs to be aligned with the other sequences so that homologous bases are present at each position.

D. Methods used for generating ITS sequences

The methods used for generating ITS sequences from the Hyptidinae in this project are outlined below, detailed protocols are given in appendix IV.

i) Source material for DNA extraction

DNA was extracted from living material grown in the Jodrell glasshouses at the Royal Botanic Gardens, Kew or from leaves dried in silica gel collected in the field. Attempts were also made to extract DNA from herbarium specimens.

Fresh material from living plants is the ideal medium from which to extract DNA but this limits the number of taxa which can be analysed to those in cultivation. Frozen material also provides a good quality material but freezing and shipping frozen material is generally impracticable. The use of silica gel to dry plant material, usually leaves, although flowers and other non-woody parts can be used, is now standard when collecting plant parts for DNA analysis. Rapid desiccation of the material causes minimal damage to the cells and restricts the activity of enzymes which would otherwise degrade the DNA. The leaves can also be stored in the silica gel, ensuring that they are kept in a constantly dry environment. Only small quantities of material are needed for sequencing, usually less than one gramme and this can easily be collected at the same time as herbarium specimens are being made as vouchers and for traditional taxonomic studies. However, lack of silica-dried material or fresh material was a considerable limitation and caused some taxa whose phylogenetic position was uncertain to be omitted from the molecular analysis (see chapter 7).

The success of extraction of analysable DNA from herbarium material depends on factors outwith the control of the investigator. The quality of the results is dependent on how quickly the material has been dried, how it has been stored and on the structure of the plant itself. Because only very small quantities need to be taken from the herbarium sheet the affect on the specimen is small but because of the uncertainty of getting reliable results extractions from herbarium specimens were restricted.

The origin of the source material, voucher specimens and Chase code number associated with all DNA extracted in the Jodrell laboratory are indicated in table 6-1. The names *H. glauca,*

H. recoria, Paraguay 1 and Paraguay 2 all refers to specimens now placed in *H. reticulata.* These names are retained in the table and the cladograms to indicate the different accessions of this species. PCD numbers refer to specimens collected for the Projeto Chapada Diamantina floristic study of central Bahia, Brazil. The two *Hyptis hagei* sequences were from the same locality and have the same voucher. Vouchers are held in the herbarium at K unless otherwise indicated. Some living collections have no voucher information cited on the database at K in which case voucher were made using the accession number given.

Table 6-1: Herbarium vouchers for extracted DNA

Genus	species	voucher	origin	MC no.
Eriope	*glandulosa*	Atkinson et al. PCD 2264	Si	3218
Eriope	*hypenioides*	Atkinson et al. PCD 2554	Si	3026
Eriope	*salvifolia*	Harley et al. 19486	cult.	1402
Eriope	*sincorana*	Giulietti et al. PCD 1551	Si	1579
Hypenia	*brachystachys*	Irwin et al. 32886	herb	1200
Hypenia	*crispata*	Harley et al. 28144	Si	3253
Hypenia	*glauca*	Harley s.n.	Si	3373
Hypenia	*macrantha*	Atkinson & Giorgio 152	cult.	3030
Hypenia	Paraguay 1	Harley et al. 28034	Si	1167
Hypenia	Paraguay 2	Harley et al. 28084	Si	1199
Hypenia	*recoria*	Atkinson & Giorgio 154	cult.	3031
Hypenia	*salzmannii*	Atkinson et al. PCD 2501	Si	3023
Hypenia	*vitifolia aff.*	Atkinson et al. PCD 2376	Si	3025
Hypenia	*densiflora*	Heringer 477	herb	1201
Hyptidendron	*canum*	Bridgewater et al. 350 (E)	Si	3225
Hyptidendron	*vepretorum*	Forzza et al. 107	Si	3249
Hyptis	*hagei* 1	Ferreira et al. PCD 1804	Si	1582
Hyptis	*hagei* 2	Ferreira et al. PCD 1804	Si	3371
Hyptis	*rugosa*	Atkinson & da Silva 148	Si	3252
Hyptis	*asperifolia*	Harley s.n.	Si	3376

ii) Extraction

DNA was extracted using a modified procedure to extract total DNA (Doyle & Doyle 1987) as outlined in appendix IV. Initial extractions from material of the Hyptidinae were disappointing. Secondary compounds are a prominent feature of the Labiatae, particularly the

Nepetoidae and may have caused problems with extraction. Using double volumes of CTAB to increase the relative amounts of buffer to impurities produced reasonably reliable results although DNA concentration was never high. Little difference was found between precipitation in isoproponol or ethanol and both were used.

iii) Caesium chloride gradient

Caesium chloride solutions were used to produce a density gradient with DNA precipitating, according to its density, as a band which can be removed from the solution leaving RNA and other impurities with low densities at the bottom of the tube. Details of the procedure are outlined in appenix IV.

Caesium chloride extraction is expensive and time-consuming but it produces very clean DNA which has maximum stability for long term storage. Baldwin et al. (1995) recommend the use of caesium chloride gradients when extracting DNA for ITS sequences since very clean DNA is a significant factor in enhancing the success rate when sequencing this region.

QIAGEN columns used for PCR product clean-ups can also be used to extract DNA or clean impure samples. They were not used to extract DNA from source material but samples extracted and cleaned with caesium chloride were further cleaned with QIAGEN columns if initial PCR amplification proved difficult.

If more than 1.5 ml of liquid was extracted from the CsCl gradient the sample was concentrated with sucrose.Concentration of DNA by osmosis through the dialysis tubing also concentrates impurities which can interfere with PCR reactions so this step was avoided if possible. All extracted total DNA was stored in the Jodrell laboratory, Kew at -80°C.

iv) PCR

Master mixes which included enough reagent for all the samples run in each reaction were used to enable increased accuracy in aliquoting very small volumes. 5µl of total DNA were included in each reaction with dNTPS, magnesium free buffer, magnesium, *Taq*, BSA, primers ITS4 and ITS5 and sterile double distilled water to a total volume of 100 µl. Volumes of the reagents used in the mix are indicated in appendix IV. For every PCR run a positive control to check the viability of the reagents and a negative control to check for the presence of contaminant DNA were included. Template DNA, which had been used successfully before, was used as the positive control and the negative control was sterile

double distilled water. Samples were subject to an initial denaturing temperature of 94°C for one minute, followed by thirty cyles of 94°C for one minute to denature the DNA, 48°C for one minute to anneal denatured strands and 72°C for three minutes to enable sequence extension followed by rapid cooling to 0°C before the start of the next cycle. At the end of the PCR the samples were held at 4°C before cleaning.

After PCR the samples were cleaned using the protocol outlined in appendix IV and were then ready for cycle sequencing reaction. The QIAquick protocol can also be used for cleaning DNA prior to PCR if tDNA samples produce unreliable results.

v) Cycle sequencing reaction

Cycle sequencing is a relatively straightforward reaction and there was little need to experiment with the variables. The total volume for each cycle sequencing reaction was 5 µl, making master mixes essential for accurate aliquoting of such small volumes. The relative volume of cleaned PCR product to water could be manipulated according to the intensity of the band recorded after PCR. Most of my PCR products did not produce intense bands on the gel and maximal volumes of DNA were added to the sequencing reaction. Reaction quantities and the procedure for cycle sequencing is specified in appendix IV.

Each PCR product was sequenced twice using one primer in each reaction. The primers ITS 4 and ITS 5 were used at 20 x the dilution used for PCR. Having two sequencing reactions from the same PCR product, both running in opposite directions enables ambiguities to be eliminated, or at least considerably reduced, when interpreting the entire sequence.

Cycle sequencing reactions were cleaned according to the protocol in appendix IV and were then stored in the freezer at -20°C until space became available on the automated sequencer. The samples were prepared by the sequencing technician and loaded onto the sequencing gel on the Applied Biosciences Inc. (ABI) Prism 377 DNA sequencer. The results were then downloaded directly onto the computer ready for assembly, editing and alignment.

Once the sequences had been transferred from the automated sequencer they were edited in Sequence Navigator and then assembled in Autoassembler. Original datafiles are held on the Medusa hard disk in the Jodrell laboratory, Kew under M.W. Chase file numbers. Assembled sequences were first aligned using Clustal followed by manual editing in PAUP. The first 40

characters and the last 50 were eliminated from the analyses because of their extreme variability and difficulty of alignment.

vi) Sequence editing

Several programs have been developed to visualise and edit sequences generated from automated sequencers, including Sequence Navigator (Applied Biosystems) which was used in my project. Sequence Navigator shows the sequence as a series of coloured peaks, each peak corresponding to a single base, colour-coded according to which base is present. The program assigns a base to each peak and ambiguities are indicated by the program at each doubtful site by **n** (=any).

Ambiguities are caused by a number of factors. One common source of problems with sequences from my project was low intensity signal, particularly with increasing distance from the primer end. Sequence Navigator responds to the intensity of the dye at each locality and the most intense colour is represented by a peak. If the signal is poor the program exaggerates the x axis, sometimes pulling up spurious peaks from the 'background noise'. If this was a problem along the length of the sequence it was rejected and run again but many sequences were interpretable along most of their length, only becoming too ambiguous for reliable interpretation at more than two thirds the distance from the primer. This length of fragment could then be assembled with the complementary sequence which had been reliably sequenced from the opposite end, thus allowing for the interpretation of the entire sequence.

vii) Sequence assembly

Once the two sequences were edited they were transferred into Autoassembler (version 1.4, Applied Biosystems). Autoassembler aligns each strand against the other until it finds the best match of base against base. The two strands were viewed with the base calls indicated above each one and standard ambiguity codes assigned to uncertainties (see chapter ? for the codes). The assembled sequence was edited. If the base was still ambiguous the code assigned by Autoassembler was kept in the sequence.

The original data files in Sequence Navigator and Autoassembler were stored on the computers in the Jodrell laboratories, Kew.

viii) Sequence alignment

After editing and assembly the entire sequence was transferred into a matrix where the rows corresponded to taxa (actually individual sequences) and the columns corresponded to base positions, i.e. characters. States at each locality, or character were either A, C, G, T, ?, ambiguity code or –. Spaces (–) were interpreted as phylogenetically uninformative but because ITS sequences vary in length spaces need to be inserted to align the bases at each locality so that they are homologous.

If enough gaps are inserted into a matrix any sequence can be aligned so that they do not differ at any position. This is clearly absurd. When aligning a matrix manually there is a tendency to insert more spaces than are appropriate to the data. This underestimates the extent of phylogenetic signal. Manual alignment of each sequence against those already present in the matrix can also introduce bias into the data depending on the order in which the sequences are added into the matrix . The Clustal alignment algorithm is available in Sequence Navigator and constructs alignment based on similarity. It tends to reduce the number of deletions/insertions and interpret changes at each locality as transformations.

Different parts of the ITS gene show different rates of change. The coding part of the gene is highly conserved and there is little variation in this part of the sequence. Much of the variation found in this region has little or no impact on the analysis as it consists mostly of autapomorphies which are phylogenetically uninfomative. The flanking region at either end of the gene is much more variable and although alignment can present problems in these regions they are much more phylogenetically valuable. The very variable regions at each extreme of the sequence were often best eliminated from the analysis due to the high error rate associated with difficulties of interpretation under dye blips or with weak signal at each end of the sequence.

ix) Analysis

The matrix was analysed in PAUP 3.1.1 (Swofford 1993) and PAUP * (Swofford 1996), using heuristic search to look for the most parsimonious tree. The initial tree was generated by stepwise addition with a random addition sequence (1000 replicates). The tree-bisection-reconnection (TBR) branch swapping algorithm was used and no topological constraints were imposed. All shortest trees were kept. All characters were unordered and of equal weight.

E. Results of ITS sequences

i) Sequences generated

20 sequences were generated from 16 taxa in the Hyptidinae. In the ingroup four sequences were obtained from *Eriope* and ten from *Hypenia*. In the outgroup two sequences were generated from *Hyptidendron* and four from *Hyptis*.

ii) Results of cladistic analysis

21 trees were generated after 1000 replicates with tree length of 450 steps. The CI = 0.742, RI = 0.735 and RC = 0.546. The strict consensus of 21 trees is shown in figure 6-3. The semistrict consensus was identical to the strict consensus. One tree is shown in figure 6-4 as a phylogram, i.e. with all branch lengths indicated.

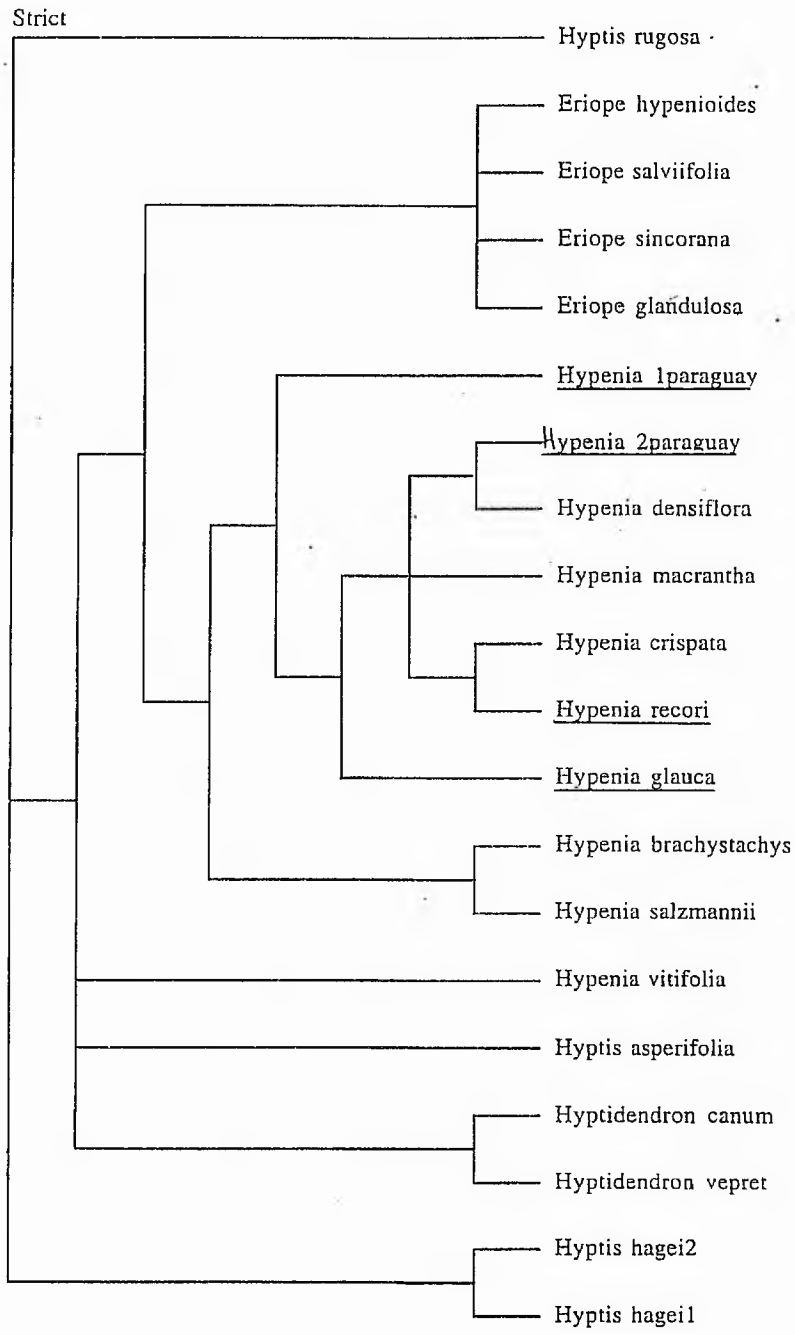


Figure 6-3: Strict consensus of the ITS analysis

Names underlined are accessions of *H. reticulata*.

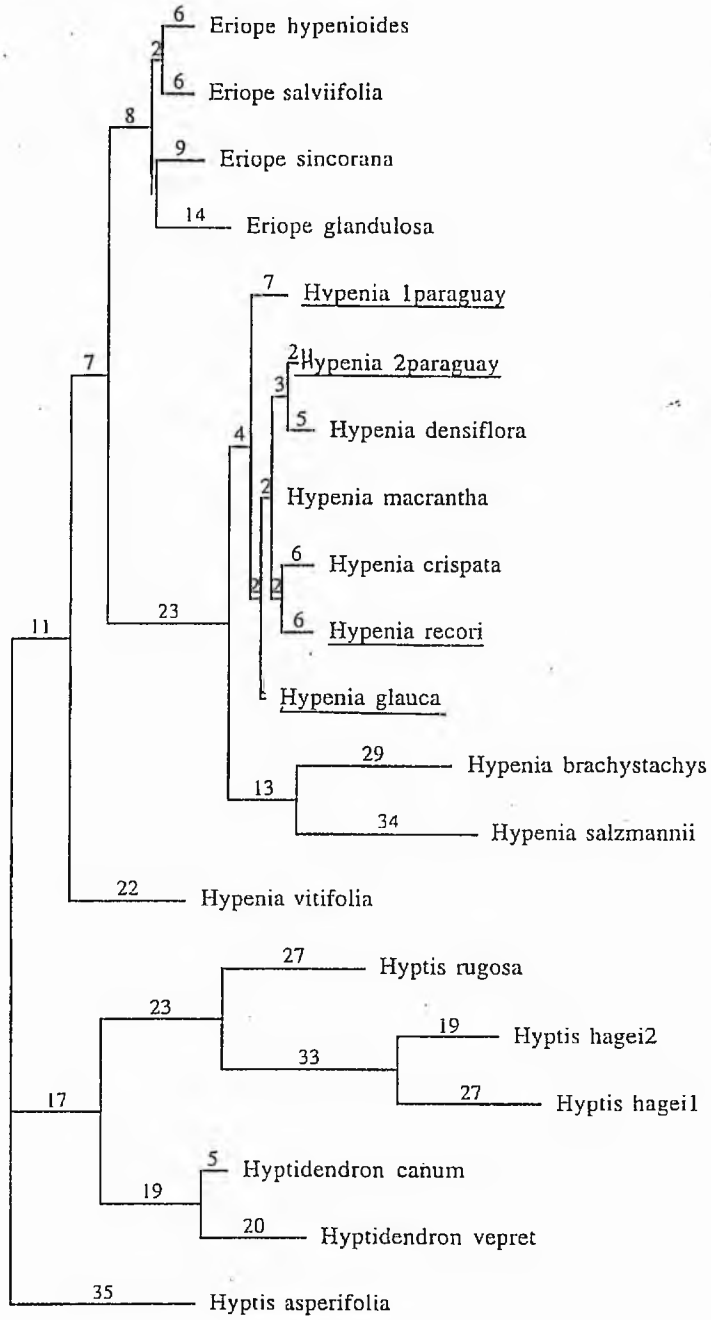


Figure 6-4: Tree 1 of 21 generated from the ITS analysis with branch lengths indicated

Names underlined are accessions of *H. reticulata*.

F. Discussion of the ITS analysis

The strict consensus (figure 6-3) shows that *Hypenia,* except for *H. vitifolia,* and *Eriope* form a monophyletic group. The position of *H. vitifolia* is not resolved and it shifts position with the two species of *Hyptidendron* and *Hyptis asperifolia* as sister to the rest of *Hypenia* and *Eriope.* Within the *Hypenia / Eriope* clade the four species of *Eriope* form a monophyletic group sister to the rest of *Hypenia. Hypenia* itself consists of two clades, one including *H. salzmannii* and *H. brachystachys* and the other including all the 'macrantha complex' species and *H. densiflora.* This latter group indicates that *H. reticulata* is a paraphyletic taxon since the four accessions of this species are scattered througout the clade. Interestingly, *H. densiflora* is placed as sister to one of the *H. reticulata* accessions from Paraguay. Branch lengths in this clade are very short (figure 6-4) and relationships indicated within this group should be treated with some caution. Branch lengths of two or three base changes may be caused by errors in alignment or reading the sequence. Nevertheless differentiation between ITS sequences of taxa in the 'macrantha complex' is poor. Although morpholgical differentiation is also lacking in parts of the 'macrantha complex' (see chapter 9) the molecular and morphological evidence are not congruent in detail in this group. Thus *H. macrantha* is a morphologically well defined taxon which is not reflected by the lack of characters supporting its position in the molecular evidence (figure 6-4).

The placement of *H. salzmannii* as sister to *H. brachystachys* is also unexpected on morphological grounds. However, branch lengths in this group indicate considerable divergence between the two species (figure 6-4).

The outgroup is not resolved which suggests the possibility of paraphyly in *Hyptis. Hyptis hagei* was represented by two accessions which are placed together but the branch lengths below each accession are very long. Both species of *Hyptidendron* are placed together in all the trees. The position of *Hyptis asperifolia* is not resolved in the tree shown and its position indicates a potentially close relationship with the *Hypenia / Eriope* clade.

G. Additional sequences in the Hyptidinae

In addition to the analysis of *Hypenia* and its close relatives other taxa were included from a wider sample in the Hyptidinae. This enabled a preliminary investigation of relationships in the Hyptidinae using examples from *Plectranthus, Ocimum* and *Isodon* as outgroups. The additional taxa included in this analysis are listed in table 6-2. The full matrix is shown in appendix V. The analysis was run in the same way as the previous molecular analysis.

Table 6-2: Herbarium vouchers for additional DNA sampled in the Hyptidinae

Genus	species	voucher	origin	MC no.
Hyptis	*eriocephala*	Hart. et al. 1713 (Arnold)	cult.	1588
Hyptis	*leptostachys* ssp. *caatingensis*	Harley et al. 19957	cult.	1589
Hyptis	*mutabilis*	Harley et al. 4126	cult.	1406
Hyptis	*petraea ?*	Forzza et al. 108	Si	3223
Hyptis	*verticillata*	Harley s.n.	Si	3375
Peltodon	*pusillus*	Harley 28117	Si	3248
Peltodon	*longipes*	Harley s.n.	cult.	3247
Raphiodon	*echinus*	1993 1074	cult.	1403
Outgroup				
Isodon	*pharicus*	Stainton, Sykes & Williams 3521(BM)	cult.	3032
Isodon	*rugosus*	00 69 19330	cult.	3033
Ocimum	*gratissimum*	Harvey et al. s.n.	cult.	3226
Ocimum	*selloi*	Solomon 9897	cult.	1583
Plectranthus	*oertendahlii*	1969 5189	cult.	3372

i) Results including additional sequences in the Hyptidinae

The analysis including additional sequences from the Hyptidinae generated 150 trees, length 1132, CI = 0.543, RI = 0.628 and RC = 0.341.

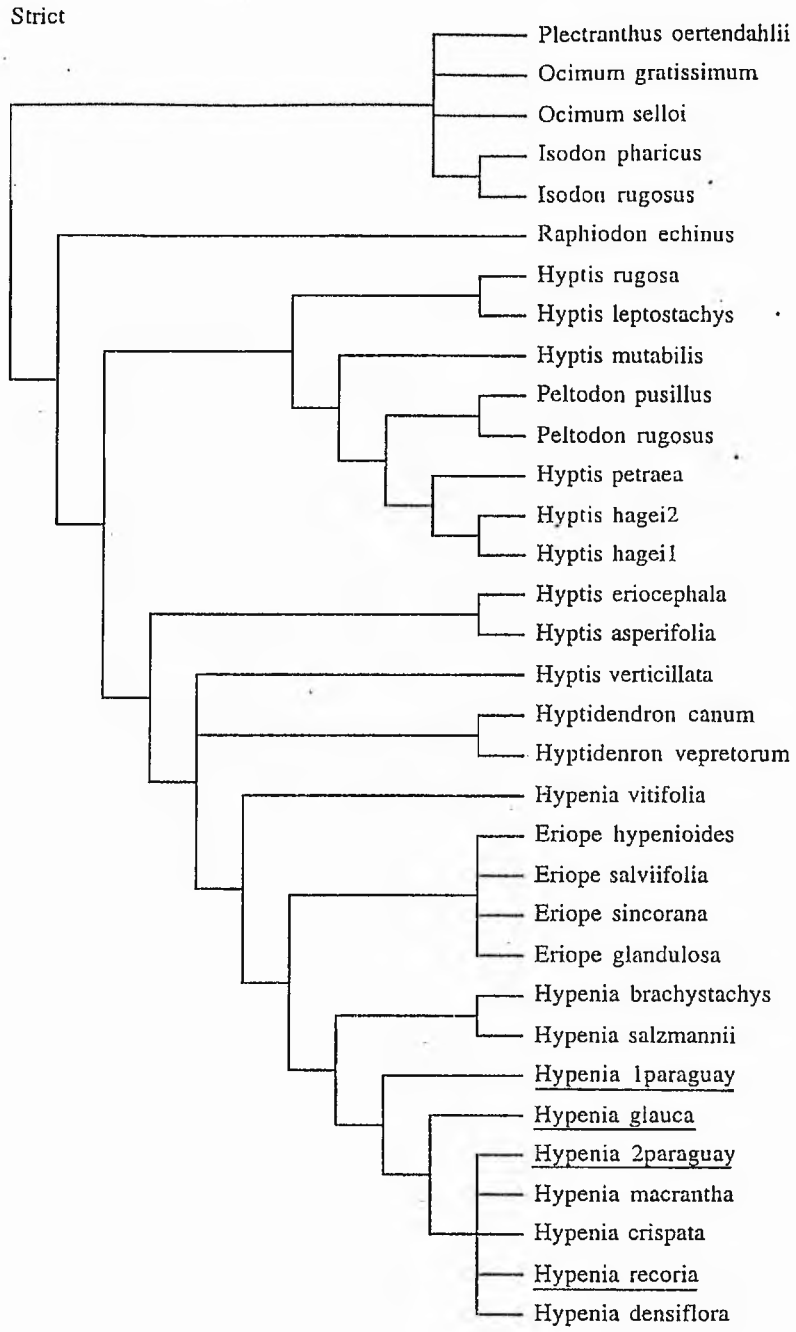


Figure 6-5: Strict consensus of 150 trees from the ITS analysis of the Hyptidinae

Names underlined are accessions of *H. reticulata*

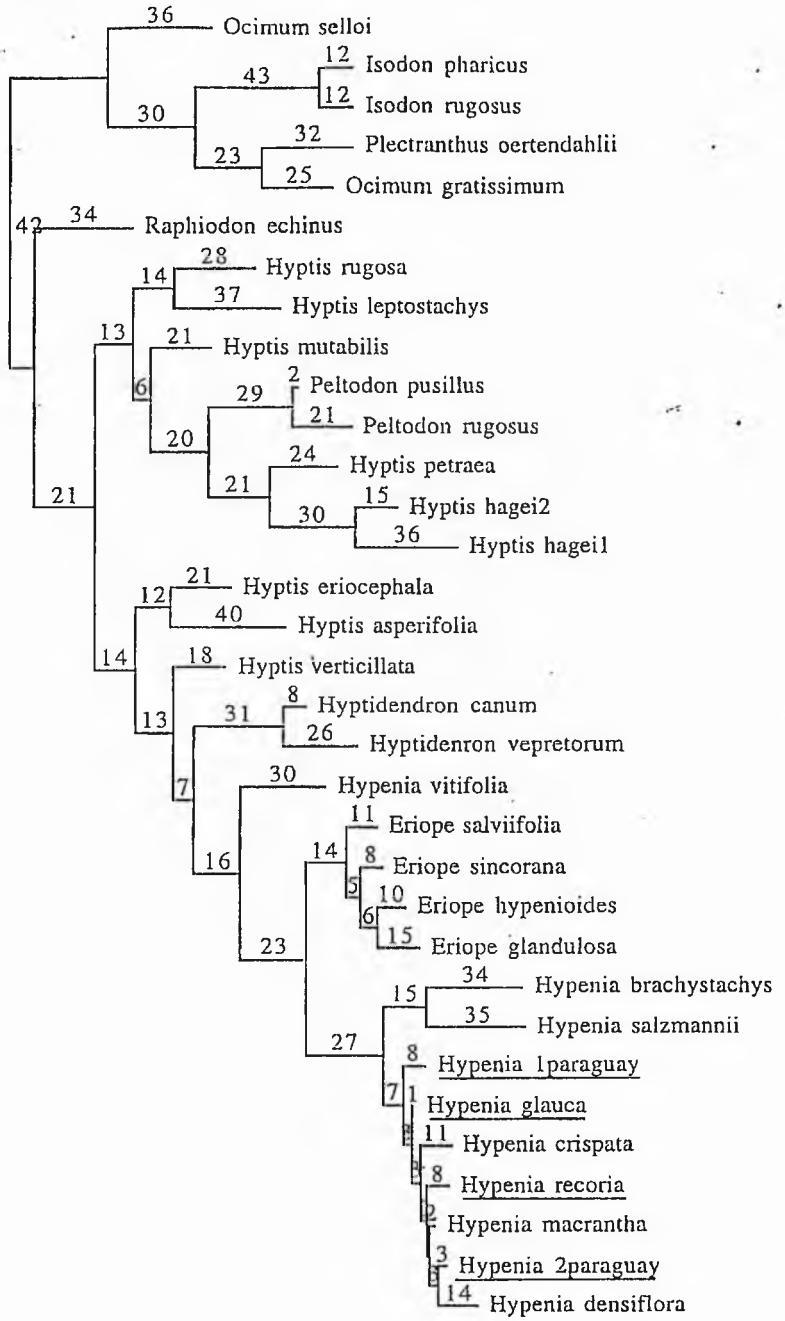


Figure 6-6:Tree 1 of 150 from the ITS analysis of the Hyptidinae

Names underlined are accessions of *H. reticulata.*

ii) Discussion of the additional Hyptidinae analysis

The strict consensus of the analysis including additional species from the Hyptidinae is shown in figure 6-5 (the semistrict was identical to the strict). The principal differences between the results of this analysis and the previous one is in the increased resolution of the taxa outside of the *Hypenia / Eriope* clade. The relatively large number of trees are a product of the lack of resolution in the 'macrantha complex' and *Eriope* and movement of *Hyptis verticillata.* There are no differences in the position of taxa between the two ITS analyses within the *Hypenia / Eriope* clade.This analysis indicates that the two species of *Hyptidendron* are sister to the *Hypenia / Eriope* clade. It also strongly suggests that *Hyptis* is paraphyletic and *Hypenia, Eriope, Hyptidendron* and *Peltodon* are all derived from within *Hyptis* but that *Raphiodon echinus* is sister to the remainder of the Hyptidinae sampled. *Hyptis rugosa* is placed with *Hyptis leptostachys* which together are sister to a clade including the two accessions of *H. hagei.* Two species of *Peltodon* are nested in the middle of this clade. *Hyptis asperifolia* and *H. eriocephala* were sister to all the *Hyptidendron, Hypenia* and *Eriope* sampled. This latter clade also include *Hyptis verticillata* which was sister to *Hyptidendron vepretorum* and *H. canum* in a monophyletic group sister to the rest of *Hypenia* and *Eriope.*

The tree shown in figure 6-6 differs little from the strict consensus (figure 6-5). The main difference outside of the *Hypenia / Eriope* clade is in the resolution of the position of *Hyptis verticillata* as sister to *Hypenia, Eriope* and *Hyptidendron* species. Within *Hypenia / Eriope* the tree is fully resolved but shows little difference from the reduced ITS analysis.

The long branch lengths in the outgroup and in the Hyptidinae outside the *Hypenia / Eriope* clade suggest that more sampling is necessary to fill in the gaps between the taxa sampled. However, the resolution between these taxa in the strict consensus suggests that the overall structure of the tree is robust.

Chapter 7: Combined analysis

A. Introduction to the chapter

The philosophical background to combining morphological and molecular is outlined in this chapter. This is followed by a comparison of the results of the morphological and molecular analyses. The results and discussion of the combined analysis are then presented. The chapter concludes with a consideration of the taxonomic implications of the cladistic analyses presented in this study.

B. Combining molecular and morphological data

The principal problem in the morphological analysis was lack of resolution caused by homoplasious characters. High homoplasy in morphological data may be the result of poor character delimitation caused by the frequently continuous nature of the variation (see chapter 5). It is this subjective nature of morphological character delimitation which seems to me to be the major problem with morphological cladistic analyses. Perhaps the strongest reason for using molecular data is that it is less subject to problems of character delimitation. However, morphological characters display a much wider range of variation than molecular characters within a single gene. If morphological and molecular data sets are available it is possible to combine the evidence from both. The arguments presented for and against this approach are discussed below.

Molecular data sets have been used to construct phylogenies based on the molecular evidence alone (e.g. Baldwin et al. 1995, Chase et al. 1993) and have also been used in direct combination with morphology to generate a phylogeny based on all the available evidence (e.g. Bruneau et al. 1995, Pennington 1996). There has been some debate on the degree to which combined analyses should be undertaken which has led to three alternative approaches to handling morphological and molecular data: total evidence uses the combination of all data in a single cladistic analysis (Kluge & Wolf 1993, Bruneau et al. 1995); consensus techniques look for areas of consensus between morphological and molecular data sets (Miyamoto & Fitch 1995); and the third approach is to only combine data matrices that do not strongly support conflicting trees (de Queiroz 1993).

Arguments against combining data sets were presented by Miyamoto and Fitch (1995) who were concerned that separate data sets from independent sources should be maintained in

order to corroborate the analyses. Bull et al. (1993) advocate statistical testing of different data sets to assess the degree of homogeneity between them. If the test indicates that data sets are homogenous or, if heterogenous the source of heterogeneity can be identified, then they accept the value of combining data sets. However, if the data sets are heterogenous and the reason for divergence cannot be discerned they recommend that combining data sets should be avoided since the results are likely to be worse than those from the individual data sets. This approach is similar to that of de Queiroz (1993) since the presence of conflicting trees from different data sets indicates heterogeneity between them.

An additional concern with combining molecular sequence data and morphology is that the molecular data may swamp the morphology. However several studies have found that this is not automatically a problem and the contribution made by the morphology is not overwhelmed by the molecular data (Smith & Sytsma 1994, Bruneau et al. 1995).

Bruneau et al. (1995) summarise the arguments for combining data and conclude that a combined approach, derived from multiple data sources makes fewer assumptions about dependence of data. In addition combining data from different sources can enhance weak signal in each source which may be obscured by noise in the data. In his study of the neotropical woody legume genus *Andira* A. L. Jussieu, Pennington (1996) found no resolution and generated a large number of trees from morphological data. Consensus techniques provided little extra resolution but using the total evidence approach and combining his morphological and molecular data he found considerably enhanced resolution and was able to identify clades and to trace the phylogeny of fruit dispersal syndromes in the genus.

Consideration also needs to be given to the nature of morphological characters which are a summary of a potentially vast amount of data derived from many different DNA loci in conjunction with the environment. This means that morphological analyses are, in the sense of molecular analyses, a combined approach and it is rarely possible, or desirable, to separate out different data sets. In addition, different data sets, even from more apparently divergent sources such as DNA or phytochemistry, are usually a practical consequence of the way in which data is gathered and do not represent independent data. Following this reasoning in this study the total evidence approach was used and molecular and morphological data sets were combined and directly analysed together.

C. Comparison of morphological and molecular analyses

Although the molecular analysis lacked a number of representative taxa which were included in the morphological analysis an overall comparison can be made of the results of the two analyses. The trees generated by the molecular and morphological analyses differ in several respects. The strict consensus of the morphological analysis shows very little resolution whereas that of the molecular, ITS analysis is much more resolved and there are fewer trees. However, much of the variation in the morphological analysis was associated with the position of several taxa which were not included in the molecular due to lack of silica-dried material, notably *H. micrantha, H. irregularis* and *H. subrosea.*

One of the most important differences between the two analyses was in the position of *H. vitifolia.* In the molecular analysis it was consistently sister to the rest of the *Hypenia / Eriope* clade but in the morphological analysis *H. vitifolia* was nested within *Hypenia* as sister species to *H. salzmannii.*

H. densiflora also shows a significantly different position in the molecular analysis. The molecular data places *H. densiflora* in the middle of the 'macrantha complex' clade. The branch lengths are very short in this group (figure 6-4) and there is little resolution between species. However, the four accessions of *H. reticulata* do not occur together and *H. densiflora* occurs as sister to one of the accessions from Paraguay. The morphological analysis places *H. densiflora* as sister to *H. brachystachys* and corresponds to previous taxonomic interpretations of relationships. This is contradicted by the molecular evidence which indicates considerable divergence of *H. brachystachys* from *H. densiflora* and indeed, places it closer to *H. salzmannii.* However, the branch lengths in the *H. salzmannii / H. brachystachys* clade are very long and show extensive differentiation between the two taxa. Some of the morphological trees support the position of *H. densiflora* within the 'macrantha complex' and sister to *H. subrosea.*

i) Taxa included in the combined analysis

The taxa included in the combined analysis were the same as those in the morpholgical analysis (table 6-1). Taxa not sequenced were coded as missing data (i.e. ? in PAUP) in the ITS matrix.

ii) Incorporating the morphological data into the molecular

The morphological characters were incorporated into the molecular data set by using position to indicate the character and transposing the states into bases. The sequences were cut at the 700th base and bases 701 to 732 were then scored for each morphological character with A = character state 0; C = character state 1; G = character state 2; and T = character state 3. Standard DNA ambiguity codes were employed to indicate uncertainty where M = A/C (0/1), R = A/G (0/2), W = A/T (0/3), S = C/G (1/2), Y = C/T (1/3) and K = G/T (2/3).

D. Methods of analysis

The combined analysis was conducted in the same way as the molecular analysis using PAUP 3.1.1 PAUP* (Swofford 1996), the latest version of the program, was also used for some replicates using the same parameters.

E. Results of the combined analysis

114 trees were generated 598 steps long, CI = 0.699, RI = 0.705 and RC = 0.493. The strict consensus is shown in figure 7-1 and one phylogram with branch lengths indicated is shown in figure 7-2.

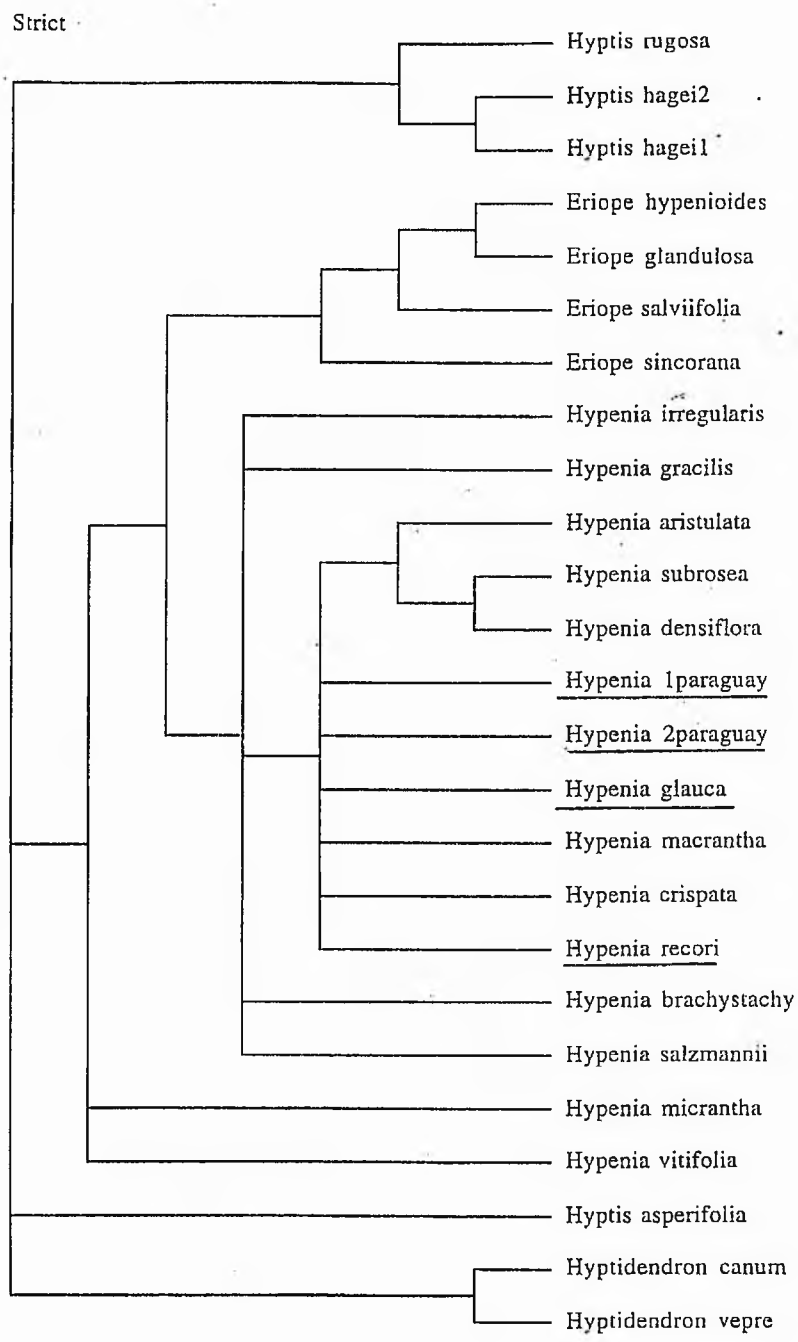


Figure 7-1: Strict consensus of the combined analysis

Names underlined are accessions of *H. reticulata*.

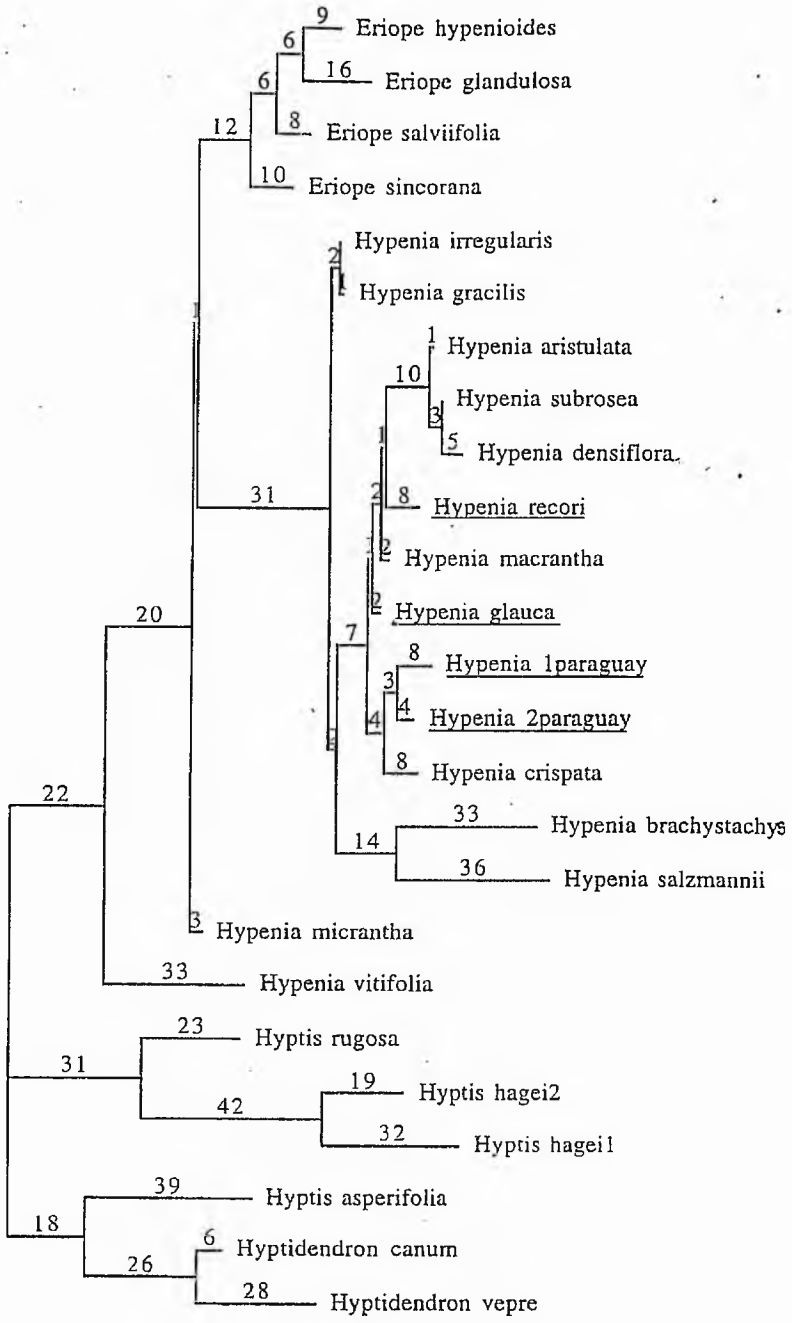


Figure 7-2: One tree of 114 generated by the combined analysis with branch lengths indicated

Names underlined are accessions of *H. reticulata*.

F. Discussion of the combined analysis

The combined analysis is similar to the molecular analysis although the strict consensus shows less resolution presumably because of the influence of the morphological analysis. Otherwise the main differences between the strict consensus of the combined and molecular analyses are concerned with the position of taxa not included in the molecular analysis. Taxa not included in the molecular analysis were: *H. irregularis, H. gracilis, H. subrosea, H. aristulata* and *H. micrantha. H. micrantha, H. irregularis* and *H. gracilis* all fall more or less in the same place as they did in the morphological trees B and C (figures 5-6 and 5-7). The other two taxa, *H. subrosea* and *H. aristulata* both occur in the same position as tree B, i.e. embedded in the 'macrantha complex' with *H. densiflora* nested in the same clade. A notable difference between the combined and the morphological analysis is the position of *H. brachystachys.* In the strict consensus the branch supporting *H. brachystachys* and *H. salzmannii* together collapses and the two taxa occupy unresolved positions with *H. irregularis, H. gracilis* and the 'macrantha complex' (figure 7-1). In the combined tree (figure 7-2) *H. brachystachys* is in the same position as the molecular analysis, i.e on the same branch as *H. salzmannii* sister to the 'macrantha complex' (including *H. densiflora*). Although *Hypenia* is less resolved in the combined analysis compared to the molecular analysis, the addition of morphological characters confers full resolution on *Eriope* in the combined analysis.

The outgroup shows some differences between all three analyses although the two species of *Hyptidendron* are sister species in all three. However, the relationship of *Hyptidendron* to *Hyptis asperifolia* is not resolved in the strict consensus of the molecular analysis or the combined analysis but in the morphological analysis it is in the same clade as *Hyptidendron.* Nevertheless the combined tree illustrated (figure 7-2) also supports this relationship.

Species delimitation is not a problem that can be tackled by cladistic methods. However, the molecular analysis strongly suggests that *H. reticulata* is a paraphyletic species. A pragmatic approach was taken to the delimitation of species in the 'macrantha complex' which is discussed in more detail in chapter 9. Classification is discussed in the following chapter.

Chapter 8: Classification of *Hypenia*

A. Introduction

This chapter is concerned with generating a workable classification of *Hypenia.* The chapter is divided into two parts: the first part is concerned with the theory of classification and nomenclature of supraspecific taxa and involves a comparison of the traditional Linnaean system with recent developments in phylogenetic taxonomy; the second part applies the theoretical considerations to the practical problem of devising a generic and infra-generic classification of *Hypenia.*

B. Classification and nomenclature

It has become increasingly accepted that classification based on phylogeny has the highest intrinsic value (Stevens 1986). However, the rules of nomenclature are derived from those first established by Linnaeus (1737, 1751), over 100 years before Darwin expounded the theory of evolution by natural selection (Darwin 1859). Calls have recently been made to change the traditional, Linnaean, system because of perceived inadequacies in the way it represents the phylogenetic history of groups of organisms (de Queiroz & Gauthier 1990, 1992, 1994, de Queiroz 1997). The first part of this chapter is concerned with the conflict which arises between the traditional system and the recent developments in phylogenetic systematics.

The section starts by defining terms applied to the processes of naming organisms. This is followed by a theoretical discussion on the function of classifications. An exploration of the development of the current rules of nomenclature in the context of Linnean classification is then presented.

i) Definition of terms

Taxonomy can be defined as the study of all aspects of variation of organisms conducted in conjunction with the development of a system of classification. In this sense it is synonymous with the term **systematics** (Stace 1989). **Classification** is the process of assigning organisms to classes in a way which allows for reference to their constituent organisms. **Nomenclature** follows from classification and is the process of assigning names to classes according to established regulations. Classification and nomenclature are therefore

the two major processes required to construct a taxonomy (or system) which is based on a synthesis of knowledge derived from systematic (or taxonomic) research.

ii) The philosophical basis of classification

Systematics may be considered to consist of two parts starting with the investigation of evolutionary history followed by recognition of named taxa by shared characters. Stevens (1984) discussed how concepts associated with the investigation of evolution have changed without a corresponding change in the methods of naming taxa. In pre-evolutionary systematics taxa were based on shared attributes and were arranged in a *scala naturae* where the ladder represented increasing complexity with distinct gaps between the rungs. As more of the diversity of life was investigated this was later replaced by the concept of a web of reticulating relationships held together by threads of intermediate individuals. With the publication of the *Origin of Species* (Darwin 1859) concepts about what classification should represent changed so that the web became a tree with a third, historical, dimension.

Pre-evolutionary taxonomy was concerned with the identification of groups consisting of similar individuals with no concept of why they might be similar. Systematics at this time was concerned mostly with the functional aspects of arranging diversity into a system which would ease identification. With the onset of evolutionary thinking an explanation for the similarity became available and systematics became concerned with representing evolutionary history. However, the rules of nomenclature were established in the pre-evolutionary phase of systematics and although the meaning of names has changed fundamentally the methods of naming have remained essentially the same.

iii) Functional aspects of a nomenclatural system

A nomenclatural system is essential to communicate ideas about relationships between taxa. There are a number of features which are essential to include in a system of nomenclature and which are discussed further below.

Stability

Stability is one of the most often cited factors which needs to be maximised (e.g. Silva 1996). An element of stability is essential because continually changing names are difficult to keep track of and will never be accepted outside of the taxonomic community, especially as the reasons for name changes are little understood. This resistance to name changes is often an important factor in making taxonomic decisions.

Consistency

Consistency refers to the necessity for nomenclature to be consistent in order to predict the position of a taxon in a classification. A name should denote that all members of a taxon are equivalent and can all be defined by possession of the same characters.

Communication of information

Information content in a stable, reliable system will be high as a consequence of its very stability and reliability but the rules used to apply names to the system need to ensure that the names themselves supply a key to the system without the need for detailed specialist knowledge.

iv) The Linnaean system

Linnaeus (1737, 1751) devised a system of naming based on a nested hierarchy in which species belong to genera and the genera in turn belong to families and so on in ever more inclusive categories. This system was later used by de Jussieu (1789) and has become the basis for current nomenclatural rules. Species are the basic unit of the system and are described by a binomial name: the first name denotes the genus, a group of similar species; the second part of the binomial is the specific epithet and is unique to one species in the genus. The Linnaean system requires that every taxon, i.e. a group of organisms designated by a taxonomic name, should belong to all the categories which occur above it in the hierarchy. This means that all species must belong to genera, all genera must belong to families and all families must belong to orders and so on.

The Linnaean system was developed as a practical means of communication and users of it have often made decisions which enhance effectiveness of communication at the expense of biological accuracy. The concern to maximise information content, in particular to enhance the mnemonic value of the classification, has often played a role in past classifications, for example de Jussieu (1789) recognised only 100 families in his system. Stevens (1997) discussed the philosophy that Bentham, the first monographer of the Hyptidinae, applied to all his classifications, both from an investigation of the implicit beliefs expressed in his work and from explicit comments made by Bentham. From this it is clear that classification was as much a practical activity as a philosophical one. Bentham thought that taxa, particularly genera and families have an ideal size but he was less explicit about how taxa should be defined. The widely different sizes of the genera in the Hyptidinae may be explained because Bentham gave greater weight to some floral characters, e.g. the cochleariform calyx lobes of *Peltodon,* than to others.

The Linnaean system has been subject to considerable debate and modification and is represented in its modern form in the various codes of nomenclature produced for plants, animals, viruses and bacteria. All the codes, including The International Code on Botanical Nomenclature (Greuter et al. 1994), lay down strict laws on the ways names are formed. The rules are aimed at standardising the way taxonomic information is presented and are an attempt to ensure that nomenclature is consistent. For example, standard suffixes are applied at each level in the hierarchy above the genus level, for plants, -ales is used for orders, -aceae denotes a family name and -oideae is used for subfamily, etc. This is a useful key to the assigned rank of the taxon under question and is a way of conveying information about relationships which is readily understood.

Definitions of taxa in the Linnaean system

In the Linnaean system, definitions of taxa are based on the distribution of characters. For example, species A and B are differentiated by their different corolla colours but are united into genus C because they share a number of characters including corolla shape. Definitions based on the possession of shared characters are used at all levels in the hierarchy. However, there is no intrinsic way of assigning rank by the possession of a particular character. The definition of rank is only concerned with position in relation to the rest of the hierarchy. Thus, a genus is a group between the species and family levels and a section is a group of species within a genus.

Typification

Typification provides a concrete means of fixing an abstract name. There are two ways of typifying names under the Linnaean system. One is to assign a specimen, i.e. an organism, as a type and is used at the species-level. The other is to specify a name as a type and is used above the species-level but ultimately refers to the type specimen of the species which is nominated type. Types perform the function of providing a reference point and are the object to which names, the formal product of taxonomy, refer. Thus the Asteraceae is defined as the family which contains the genus *Aster,* the type genus of the family. Types have no evolutionary significance and in the preceding example there is no biological meaning associated with the designation of *Aster* as the type genus of the Asteraceae. This means that there is no objective measure to determine which specimen or organism should be designated as the type of a particular taxon.

Representation of phylogeny

The principle of grouping by descent is not central to the Linnaean system (Stevens 1984) but one of the main reasons for its continuing acceptance is the compatibility between the

Linnaean hierarchy and the hierarchy which has arisen as a consequence of the evolution of life on earth. Thus, the Linnaean system is logically compatible with the representation of nested sets of species, monophyletic groups and characters (Dominguez & Wheeler 1997). Hierarchical systems are also very efficient in the way they represent information. Once the taxon name in a hierarchical system is known considerably more information can then be inferred. Thus, for a species represented by a binomial name the generic part of the name provides information on which other species are considered to be taxonomically allied to it at the same level in the hierarchy.

v) Phylogenetic taxonomy

Recent discussion in the cladistic literature has started to address the problems which arise when existing rules of nomenclature are used to convey new ideas about the phylogenetic position of taxa. Attempts have been made to devise a nomenclatural system which is compatible with phylogeny. This system has been called phylogenetic taxonomy (de Quieroz & Gauthier 1990) and is discussed in more detail below.

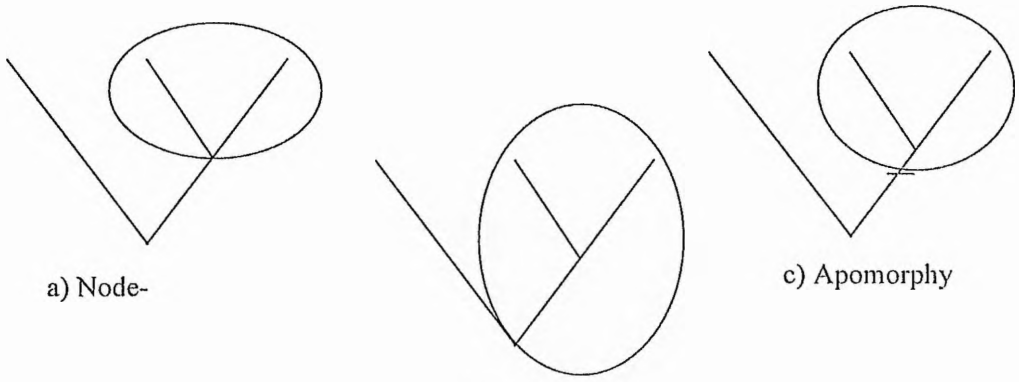
Definitions of taxa in phylogenetic taxonomy

Since historical processes produced the character distributions we see, it is more logical to give primacy to history when constructing classifications (Stevens 1986). Ideally, this means that classifications should use shared history rather than shared characters to define taxa. Conflicts between phylogeny and the Linnaean system arise because definitions of taxa within the Linnaean system are based on characters. If a defining character is lost in a taxon which belongs to a larger group of organisms descended from a common ancestor the descendant taxon will be assigned separate status on the basis of this character despite its historical place within the larger group. This is a frequent occurrence and is the cause of the large number of paraphyletic groups given taxon status (see below for a further discussion of paraphyletic groups in the Hyptidinae).

Cladistic theory requires all groupings in a taxonomic system to be monophyletic, i.e. to share common ancestry (Hennig 1966). Three definitions of monophyly have been recognised by de Queiroz & Gauthier (1990). Node-based definitions are used to define the name of a clade stemming from the most recent common ancestor (example 1a); stem-based definitions define the name of a clade of all species sharing a more recent common ancestor with one specified organism, species or clade than with another (example 1b); apomorphy-

based definitions define the name of a clade stemming from the first ancestor to evolve a specified character (example 1c).

Example 1



Stem-based definitions of monophyly are subject to instability if the specified taxon (or organism) to which the clade is compared moves position with different hypotheses. In this case, the clade defined under one hypothesis ceases to exist under another, even if the constituents of the clade under question remain the same. Apomorphy-based definitions are subject to even more uncertainty if the chosen apomorphy subsequently proves to be homoplastic and therefore defines more than one clade. Node-based definitions are the most stable means of defining monophyletic taxa are used as the basis of taxon definition by de Queiroz and Gauthier (1994).

Typification

Using the node-based definition of monophyly outlined above de Queiroz and Gauthier (1994) propose a change in definitions of taxa from those based on arbitrarily chosen type organisms or taxa to definitions based on type organisms selected in a way which best represents the position of the ancestor of the clade.

Representation of phylogeny

Phylogenetic taxonomy in the sense of de Queiroz and Gauthier (1990, 1992, 1994) attempts to use nomenclature to represent phylogeny by presenting the names of taxa in a particular order. The order used starts with the most inclusive taxon and then employs indentation to indicate increasingly exclusive groups within it. See example 2 for a phylogenetic representation of the Hyptidinae in the Labiatae (only inclusive taxa which are relevant to the Hyptidinae are represented). Name endings are not specific to any rank and the only means

of assessing position in the phylogeny is from the entire classification, preferably in conjunction with a cladogram. Thus, if the Hyptidinae were to be accorded the same status as the Ocimeae the ending -inae would be retained to indicate that the constituents of the taxon remain the same. However, there would be no indication from the name alone that the Hyptidinae are now considered to be equivalent to the Ocimeae, this information is only available from the indented classification (see example 3). Previous attempts to make nomenclature compatible with the representation of phylogeny have used similar approaches (Farris 1976) but as Wiley (1979) pointed out they present considerable practical difficulties, particularly if the classification extends over more than one page. An alternative convention was suggested by Nelson (1972, 1973) whereby taxa were listed, with each taxon being the sister group of the one after it in the sequence.

Example 2: Phylogenetic taxonomy of the Hyptidinae within the Labiatae: current status

Labiatae
- Nepetoideae
  - Ocimeae
    - Ociminae
    - Plectranthinae
    - Hyptidinae

Example 3: Phylogenetic taxonomy of the Hyptidinae: revised status

Labiatae
- Nepetoideae
  - Ocimeae
    - Ociminae
    - Plectranthinae
  - Hyptidinae

vi) Maximising the functional value of a classification

The functional value of a system of classification can be assessed in terms of the three criteria, identified above, i.e. stability, consistency and information content. These three criteria are discussed below for the Linnaean and phylogenetic systems.

Stability

Synonymy, caused by the allocation of different names to the same taxon, is a symptom of instability in a system of classification but it is also a product of changing understanding of relationships. Rules are required to decide which name of those already in existence should be used. The Linnaean system assigns priority to the earliest validly published name. De Queiroz and Gauthier (1990) propose that the way of deciding which retains priority should be changed to that of the taxon which is closest to the ancestral node. This change would not reduce nomeclatural instability since in many cases the position of the ancestor is not resolved. Even if it were, the number of names allocated to synonymy would be the same using either a phylogenetic taxonomy or a Linnaean one. However in a phylogenetic taxonomy the spelling of the name which has priority would not change even if the position of the taxon in the hierarchy has shifted. In examples 2 and 3 above the meaning of Hyptidinae changed but the spelling of the name remained the same. In the Linnaean system if the Hyptidinae were to be considered equivalent to the Ocimeae it would become a tribe and would have to be allocated a different ending, -eae, so that the name would change from Hyptidinae to Hyptideae. Thus the Linnaean system requires name changes where the phylogenetic system does not. However, if the name does not change the taxonomic change is obscured (Dominguez & Wheeler 1997).

Consistency

One of the consequences of the nested hierarchy of ranks in the Linnaean system can be the introduction of a series of names for the same taxon merely to denote equivalence in the hierarchy. In the case of a species which is identified as forming a monophyletic clade sister to an order consisting of several thousand species arranged in a number of families and many genera the single sister species must be assigned generic, family and order rank. This introduces a series of different names ascending the hierarchy which all apply to the same entity. In this case the names applied in the Linnaean system do not have consistent meaning. One set of taxon names is used to describe increasingly inclusive groups of species united by common characters whilst the other set of names describes a single species and their sole purpose is to confer the appropriate rank.

An additional problem with introducing equivalence throughout the hierarchy when trying to represent cladistic relationships, is the problem of running out of ranks. Translating a phylogeny into a nested hierarchy may require many additional ranks which are not commonly used in nomenclature (Crane & Kenrick 1996).

Cantino et al. (1997) and Kron (1997) have applied phylogenetic taxonomy to the Labiatae and Ericales respectively. Both concurred with the recommendation to abandon exhaustive subsidiary taxa as a means of eliminating large numbers of 'empty' names. To convey the full classification they recommended that the phylogeny should be presented with the nomenclature. In this case names would only be applied to meaningful groupings (i.e. clades) which can be identified by characters. The full classification, including un-named clades, which have no defining characters, could then be presented using an accompanying phylogeny in specialist publications.

Information content

Many features of the Linnaean system are used to indicate the position of a taxon in the nomenclatural hierarchy, e.g. standardised suffixes of higher taxa. The information conveyed in this way does not reflect an intrinsic, biological, property of the named taxon but does indicate its position in relation to other taxa in the hierarchy. The phylogenetic system proposes the abandonment of name changes according to position in the phylogeny (hierarchy) and therefore no information is available from the name itself as to the relative position of the taxon.

Some aspects of Linnaean taxonomy were not discussed in detail by de Queiroz and Gauthier but were relevant to my taxonomic studies. In particular, binomial name changes, which are probably the commonest cause of nomenclatural instability, were not seriously tackled by current discussion on phylogenetic taxonomy. It is therefore impossible to compare the Linnaean or phylogenetic systems on this issue. Assignment of generic rank was one of the biggest problems in the classification of *Hypenia* and needed to be considered in more detail.

vii) Generic rank

Within the Linnaean system the particular rank applied to a group above the species level has little value in itself, what matters is the arrangement of taxa within the hierarchy. However, the generic rank has particular importance for two reasons. The first is a consequence of the rules of nomenclature in the Linnaean system since to acquire a binomial all species have to be assigned to genera, making the genus a mandatory rank in the Linnaean system. The second reason is a result of the way species are recognised and grouped together by the human mind. Most people appear to be able to recognise certain groups of species as being related and these groups have frequently been given names. Many of the conspicuous genera

in the European flora possessed names prior to Linnaeus and continue to bear common names which correspond closely to formally recognised genera. The common names rose, dock, oak and buttercup are all applied in a way which corresponds closely to formal generic delimitation. Thus for identification purposes genera should be easily defined and, although in theory the genus has no primacy in classification, its place in identification schemes is, if only the result of long-standing convention, an important one.

Since all species must be assigned a generic name, changes to generic names have more impact than any other change within the current rules of nomenclature. This can introduce a possible source of inertia in changes to generic names since nomenclatural stability is most reduced by name changes at this rank.

Binomial names were not considered in detail by de Queiroz and Gauthier (1992, 1994) but for the purposes of this study they are perhaps the most important category to be considered. Genera are subject to all the problems discussed by de Queiroz and Gauthier (1992, 1994), but their part in binomials makes them essential unless binomial names are to be abandoned or they change their meaning completely. Species could be designated by uninomials (Schander & Thollesson 1995) but the suggestions made to how this could be done are clumsy or impracticable, requiring either very long names or use of the existing species name with a date. Even with the addition of the date there are many specific epithets which are extremely common and are unlikely to be unique even in one year of publication. In view of the practical value of generic names in conveying information they are particularly important and should be maintained. However, care must be taken in defining generic boundaries because of the destabilising effect of changes at this rank.

Paraphyletic genera

Cladistic theory does not accept the recognition of paraphyletic taxa but in practice they are a common taxonomic feature. This is the result of defining taxa based on shared characters and retaining the same rank for taxa which are derived from within another taxon with common ancestry. Such taxa at the generic rank have been referred to as 'convex genera' (Brummitt 1996). This is a particularly relevant problem in this study since the recognition of *Eriope* as a separate genus from *Hypenia* makes *Hypenia* a paraphyletic genus. Paraphyletic genera have been defended by Brummitt (1996) on pragmatic grounds. If a group can be easily recognised then it should be. However, this causes a problem of defining the group from

which it is separated since the resultant 'convex genus' will have no unique defining character.

viii) Philosophical basis of the classification used in this study

Biological diversity is incredibly complex and any system used to classify it will be imperfect. Therefore taxonomists need to make decisions about what their classifications aim to acheive.

In my view communication of information is the prime function of taxonomy and the Linnaean system provides a far more efficient means of fulfilling this function than the system proposed by de Queiroz and Gauthier. The Linnaean hierarchy was originally devised as an artificial system but it is logically consistent with the representation of evolution. Although names should be consistent with a knowledge of evolutionary patterns and processes they should be possible to apply without detailed knowledge of history. Thus, character-based definitions are essential. However, the inconsistent meaning of taxon names does seem to be a genuine problem although they are essential in the current system. A classification which cannot be understood unless it is presented in its entirety in a complex format and which requires a cladogram for clarification seems to be an inadequate response to the problem of representing phylogeny through nomeclature. Although phylogenetic taxonomy *sensu* de Queiroz and Gauthier presents a valid criticism of the Linnaean approach to nomenclature and can increase stability by reducing orthographic changes, their system seems inadequate for the need of taxonomy to communicate understanding of biological diversity. In addition, it potentially obscures information because it lacks a coherent structure which fails to make the artificial nature of the system explicit.

To my mind the aim of classification is not the same as phylogenetic reconstruction. Classification is primarily concerned with communicating information about similarities between groups of organisms. Phylogenetic reconstruction attempts to discover historical relationships. Ideally, classification should represent phylogeny but this may not always be practical. In particular, relationships indicated by molecular characters but not supported by morphology are difficult to communicate in practice. Other problems, particularly paraphyly, are apparently endemic in systematics and it is not always desirable to eliminate it from classification.

C. Classification of *Hypenia*

With these problems in mind the classification of *Hypenia* was constructed using the combined analysis as a theoretical base from which to work (figures 7-1 and 8-1). However, the combined phylogeny is not completely compatible with the classification. The classification derived from the combined phylogeny and the points at which they diverge are discussed below. Paraphyly in the Hyptidinae is also discussed and the generic boundaries and infra-generic classification of *Hypenia* is outlined.

i) Generic classification

The analyses presented do not present an unequivocal picture of relationships in *Hypenia* which can be translated directly into a classification of the genus. Nevertheless, there are some features of the cladistic analyses which have important implications on the taxonomy of *Hypenia.* Perhaps the most striking feature of all the cladistic analyses is the position of *Eriope* nested within *Hypenia,* thus making *Hypenia* paraphyletic (figures 5-4, 6-3, 7-1) . The paraphyly of *Hyptis* was also indicated in the larger molecular analysis of the Hyptidinae (figure 6-6). As currently circumscribed *Hypenia* is a paraphyletic group and the generic and infra-generic limits of *Hypenia, Eriope* and *Hyptis* need to be reconsidered.

There are a number of options available to solve the problem of paraphyly in *Hypenia* but they produce groups with widely diverging membership and diagnostic characters. The various ways in which these taxa can be named in a way which is consistent with the phylogeny are discussed below. These taxonomic schemes do not eliminate paraphyly and the arguments for and against paraphyletic groups at different taxonomic ranks are considered in *Hypenia.*

Paraphyly at the generic level can be reduced by a number of means which are discussed below. These options are also summarised on the strict consensus of the combined analysis in figure 8-1.

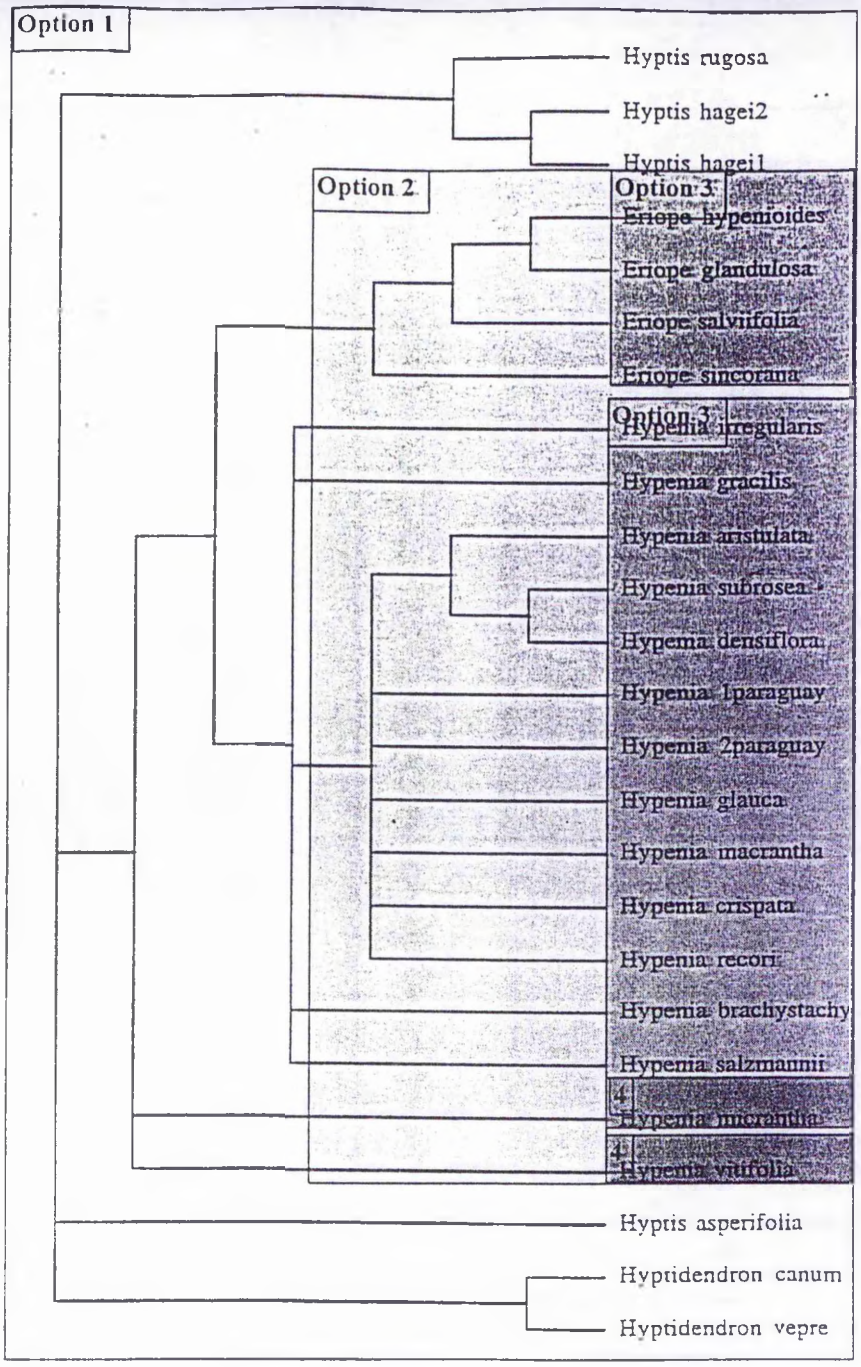


Figure 8-1: Strict consensus of the combined analysis showing the options for generic classification

The shaded areas show generic boundaries under each option. Only the two new genera are shown for option 4, the generic boundaries between the remainder of *Hypenia* and *Eriope* are the same as in option 3.

1. Sink all of *Hypenia, Eriope* and *Eriopidion* species into *Hyptis.* To eliminate paraphyly completely at the generic level it may also be necessary to sink all the other genera of the Hyptidinae into *Hyptis.* In this case *Hyptis* would be defined by the morphology of the anterior corolla lobe. *Asterohyptis* lacks the defining character of the genus and if it proves to be derived from within *Hyptis* generic definitions would need to be reconsidered.

Although *Hyptis* would be easy to define in this way I do not feel that at this stage my phylogeny was complete enough to justify such wholesale changes. Sinking *Hypenia* and *Eriope* into *Hyptis* obscures the distinctiveness of these species groups and hence diminishes the information content of the taxonomy. In addition this option involves many name changes and hence would contribute to taxonomic instability.

2. Combine *Eriope, Eriopidion* and *Hypenia* in one genus and maintain the remaining genera of the Hyptidinae. *Eriope sensu lato* would then be defined by the presence of bracteoles (apart from *H. vitifolia*). Characters associated with the 'greasy-pole syndrome' could also be used to help define the genus. These characters are not universally distributed in *Eriope sensu lato* but they are uniquely confined to it.

This arrangement emphasises the distinctiveness of *Eriope* and *Hypenia* and, at the same time, highlights the close relationship between them. Bracteoles, the defining character of the genus, are not universally distributed but they are only absent from *H. vitifolia* (and its close relative, *H. longicaulis*), a species with very well-developed 'greasy-pole syndrome' characters. It is thus a relatively easily defined genus. Instability caused by changing names is relatively high with this option, especially since *Hypenia* has only recently been changed from *Hyptis.* The status of the remaining genera of the Hyptidinae may need to be reconsidered but this should be left until a more detailed phylogeny is available for them.

3. Maintain current generic limits. *Eriope sensu stricto* would be defined by unique corolla and calyx characters in combination with the presence of the stylopodium and *Eriopidion* would be separated from *Eriope* by the lack of the stylopodium and the presence of several autapomorphies. However, this arrangement causes difficulties in the definition of *Hypenia* since it has no unique, universally distibuted, defining characters and can only be separated from *Eriope* by the absence of the stylopodium.

The main appeal of this option is that it has minimal impact on name changes. In addition *Eriope sensu stricto* is a distinctive taxon which is easily identified. However, *Hypenia* is not so easy to identify and would remain a rather disparate group.

4. Maintain the current circumscription of *Hyptis* and *Eriope* and eliminate paraphyly from the *Hypenia / Eriope* clade by recognising *H. vitifolia* (together with the closely related *H. longicaulis*) as a genus defined by the presence of multi-flowered cymes lacking bracteoles and *H. micrantha* as another monotypic genus defined by the presence of leaf-like phyllomes and branched hairs.

This option has the advantage of minimising name changes whilst at the same time maintaining consistency with the combined phylogeny. However, it over-emphasises the distinctiveness of *H. vitifolia* and *H. micrantha* and obscures their morphological similarity to *Eriope* and *Hypenia.* In addition, *H. micrantha* is poorly known and there is no molecular evidence to support its position in the combined analysis.

Generic status applied to *Hypenia*

In the taxonomic accounts in chapter 10 I have used *Eriope* as the generic epithet for *Hypenia* species, i.e. I have chosen to adopt option two for the generic treatment. This is because it allows for consistent definition using well-defined morphological characters which are widely distributed in the genus.

ii) Infra-generic ranks

The strict consensus of the phylogeny presented in the combined analysis does not show consistent clades which can be recognised at the infra-generic level. However, morphological variation in *Hypenia* and *Eriope* is sufficiently high to justify the recognition of infra-generic ranks. I have not attempted to eliminate paraphyly, nor have I attempted to make the infra-generic classification of *Eriope sensu lato* completely consistent with phylogeny. Instead my infra-generic classification has largely been constructed to allow for maximum ease of identification and has been based on the distribution of morphological characters in the genus. As a result the distribution of morphological characters has been used as the basis of my infra-generic classification.

In other respects the molecular and morphological analyses show conflicting positions of a number of taxa and the combined analysis (figure 7-1) largely reflects the molecular results

(figure 6-3). The morphological analysis included several important taxa which were not included in the molecular analysis. Their position in the combined analysis largely followed morphology and the following discussion refers to the results of the morphological analysis for these species. The groups discussed below and recognised in my classification are imposed onto the strict consensus of the combined analysis in figure 8-2.

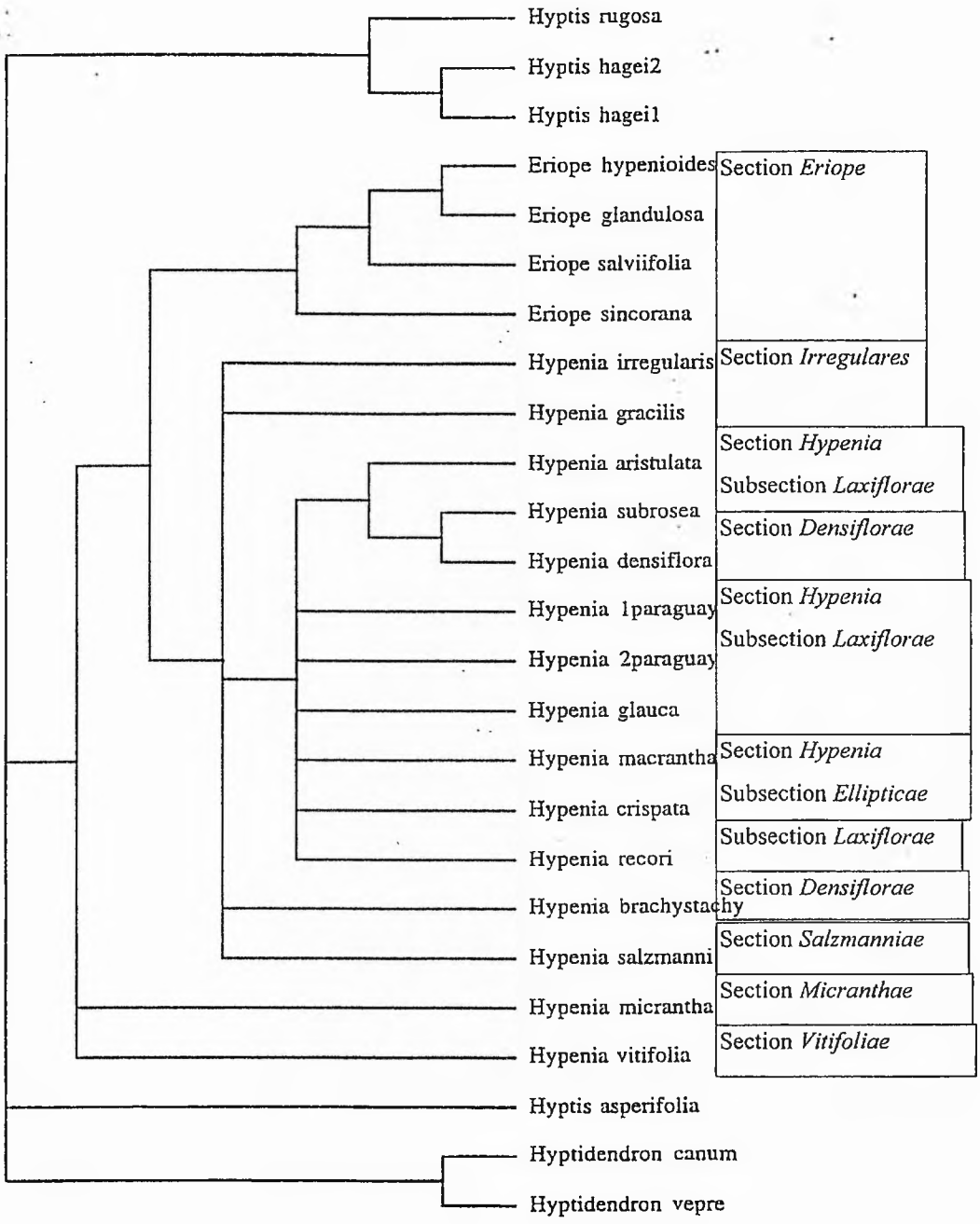


Figure 8-2: Strict consensus of the combined analysis showing the classification of infra-generic ranks

In all the analyses the 'macrantha complex' occurs consistently as one clade. In my classification the 'macrantha complex' has been recognised as one taxon defined by the large flowers and open inflorescence. In some of the morphological trees (figures 5-5 and 5-6) *H. densiflora* and *H. brachystachys* nest in the middle of the 'macrantha complex' as the sister group to *H. subrosea.* This indicates that the 'macrantha complex' may be paraphyletic and the place of *H. densiflora* in the molecular analysis also suggests this (figures 6-4 and 6-5). Certainly *H. subrosea*, with its white flowers and globose swellings on the stems, shows a strong resemblance to *H. densiflora* and *H. brachystachys.* However, its inflorescence structure suggests a close relationship with the 'macrantha complex'. Inflorescence characters have been traditionally emphasised in *Hypenia* (cf. *Laxiflorae* and *Densiflorae* of Bentham (1848) and Epling 1936, 1949) and they have high practical value in immediate recognition of taxa. For this reason *H. subrosea* has been maintained in the 'macrantha complex' (section *Hypenia,* see below).

The resupinate-flowered group nests within the 'macrantha complex' and recognition of it as a separate taxon makes the remainder of the 'macrantha complex' parapyletic. However, resupination of the flower is a conspicuous character which is correlated with flower size and habit characters which, although less well-defined, support the recognition of this group as a taxon in its own right.

The relationship with *H. subrosea* discussed above and the evidence of the molecular analysis indicates that *H. densiflora* at least, is nested within section *Hypenia* (the 'macrantha complex').However, *H. densiflora* and *H. brachystachys* are strongly supported together in the morphological analysis and have been placed in the same taxon, despite the contradictory evidence of the molecular and combined analyses. They are well defined by the crowded cymes in the inflorescence, connate calyx lobes, sigmoid flowering calyx and the presence of waxy stems with globose or elongate swellings. The molecular evidence is interesting and the relationship with *H. salzmannii* requires further investigation. However, the morphological characters are extremely well-defined and separating *H. densiflora* and *H. brachystachys* in a practical taxonomy seems unjustified.

The relationship of *H. gracilis* and *H. irregularis* to each other is not clearly indicated in the morphological analysis but they have been placed together in the classification because they have very similar small flowers with equal calyx lobes in one-flowered cymes and lack waxy, swollen stems. No molecular evidence was available for these uncommon species.

The morphological analysis suggests a close relationship between *H. salzmannii* and *H. vitifolia* (figures 5-5 and 5-6) but this is contradicted in the molecular and combined analyses which place *H. vitifolia* in an isolated position in relation to the rest of *Hypenia* (figures 6-5 and 7-1). Morphological characters, especially the lack of bracteoles, cyme structure and corolla shape suggest that *H. vitifolia* is also morphologically isolated in *Hypenia,* despite its position in the morphological analysis. As a result I have recognised *H. vitifolia* (together with *H. longicaulis* which is morphologically very similar) as one taxon and *H. salzmannii* as another. *H. salzmannii* has small, blue flowers which are unique in *Hypenia,* although the tubular corolla and inflorescence structure is common in *Hypenia.*

The morphological analysis strongly indicates the isolated position of *H. micrantha* and it has been recognised in my classification as a monotypic taxon defined by presence of branched hairs, campanulate corolla and leaf-like phyllomes.

The rank of the groups identified above are as follows. *Eriope sensu stricto* and the 'macrantha complex' have been recognised as sections and a number of monotypic sections or sections consisting of two or three species have been constructed to accomodate the remaining species. These are: section *Vitifoliae,* including *H. vitifolia* and *H. longicaulis*; section *Irregulares* including *H. irregularis* and *H. gracilis*; section *Salzmanniae* with *H. salzmannii*; and section *Micranthae* with *H. micrantha.* In addition, *H. brachystachys* and *H. densiflora* have been placed together in section *Densiflorae* along with *H. concinna,* which is morphologically very similar to *H. brachystachys.*

Section *Hypenia* (the 'macrantha complex') has been subdivided into two subsections, subsection *Ellipticae* includes the resupinate-flowered species and subsection *Hypenia* contains specie in the section. The taxonomy of section *Eriope* is outwith the scope of this study and it has not been subdivided in this account. *Eriopidion* is included in section *Eriope* for the purposes of this study although no formal decision has been made about the status of this taxon. The full classification and formal taxonomy of *Hypenia* species is presented in chapter 10.

Chapter 9: Species delimitation

A. Introduction to the chapter

This chapter is concerned with species delimitation in *Hypenia.* The chapter starts with an outline of species concepts and an exposition of the species concept used in this study. The biggest taxonomic problem at the species-level was concerned with species delimitation in the 'macrantha complex' and a discussion is presented here for this group. Species delimitation in most other cases was less problematic and notes on their delimitation are presented with the species descriptions in chapter 10.

B. Species concepts

The basic unit of classification is the species, but finding a definition has proved controversial and as yet there is no comprehensively accepted species concept. Nevertheless, an explicit species concept is a necessary part of any monographic investigation, although it is frequently missing from many botanical monographs (Luckow 1995).

In a study of the taxonomy of the relatively well-known British flora, it was found that most species were uncontroversially defined, despite the failure of most taxonomists to adopt explicit or consistent species concepts (McDade 1995). Problematic species were associated with processes such as hybridisation or asexual reproduction, problems which were identifiable and could throw considerable light on evolutionary processes. The species concept adopted by taxonomists, consciously or otherwise, therefore appears to be fairly robust and applicable to describing patterns in nature despite considerable disagreement in the definition of species or even in many cases, with no explicit definition at all. However, the scientific method demands definitions and many different species concepts have been proposed. It is possible to classify them according to their theoretical base.

i) Taxonomic species concept

Taxonomic species concepts are rarely described as such but there seems to be general agreement in the application of the species category in floras and monographs (see above). According to Davis and Heywood (1963) a definition of a taxonomic species is based on 'assemblages of individuals with morphological features in common and separable from other such assemblages by correlated morphological discontinuites in a number of features.'

In other words, a species under this definition is a group of samples which are similar enough to be considered the same thing, both by the taxonomist studying them and by others. An essential part of this definition is the ability to describe the species to other people using characters which are consistent and observable. Taxonomic species concepts in this sense are based on phenetic evidence. In addition, they are highly subjective and cannot be defined with any degree of precision. As a result considerable effort, and much literature, has been devoted to devising a precisely defined species concept which is consistent with current studies of evolutionary processes. However, taxonomic species concepts are an 'effective first step' in the task of describing biodiversity and as such are employed by most taxonomists.

ii) Biological species concept

The biological species concept as expounded by Mayr (1969b) defines species by the reproductive potential of individuals to interbreed. This criterion has been strongly criticised, and is now largely rejected, as the basis for defining species. Cladistic methodology rejects it because the capacity to interbreed is plesiomorphic and therefore cannot be used as a criterion for assessing relatedness (Rosen 1979). If individuals belonging to A are all capable of interbreeding then the reproductive isolation that occurs when B arises is an apomorphy and the reproductive capacity of A becomes a plesiomorphy. In addition, mechanistic definitions have been rejected because they are based on contemporary micro-evolutionary processes which are not compatible with the assessment of hierarchical historical events in a phylogenetic system (Luckow 1995, Donoghue 1985). Objections to the biological species concepts based on events observable in nature arise from the many examples of distinguishable entities capable of interbreeding or of apparently indistinguishable entities which cannot interbreed. For these reasons there has been considerable activity directed at finding a species definition to replace the biological species concept.

iii) Phylogenetic species concepts

Compatibility with cladistic methodology is an important requisite for species concepts and a number of different definitions have been proposed under the heading of phylogenetic species concepts. Even within this theoretical framework there are considerable disagreements which reflect divergent approaches to systematics. Two classes of species concept can be identified which have developed in the framework of phylogenetics: character-based species concepts and history-based concepts (Baum & Donoghue 1995).

iv) Character-based species concepts

Character-based concepts rely on the identification of characters for species recognition. Nelson and Platnick (1981) defined species as 'the smallest diagnosable cluster of self-perpetuating organisms that have unique sets of characters'. This was developed under various names, including the phylogenetic species concept by Cracraft (1989) and Nixon and Wheeler (1990, 1992). However, a unique character can be either an apomorphy or a plesiomorphy and groups defined by plesiomorphies, at whatever level, are not acceptable to some (Nelson 1989). Another objection was raised by Baum and Donoghue (1995) who point out that fixation of characters in populations may occur some time after the splitting event which gave rise to a new species. Characters which remain variable after splitting of two species cannot be used to indicate speciation, even if such an event has occurred. They contend that species concepts based on character delimitation alone cannot be used in an evolutionary context and that historical processes must be recognised in a truly phylogenetic species concept (Baum & Donoghue 1995).

v) History-based species concepts

History-based approaches are concerned with the identification of lineages of organisms connected by reticulate (tokogenetic) relationships which are confined within the group (Frost & Kluge 1995). These 'internodal species' therefore exist as lineages and speciation occurs when the lineages split. Under this concept a lineage can be defined as exclusive if all its members are more closely related to each other than to any member of another lineage (Donoghue 1985). However, this definition of exclusivity can be reduced to absurd levels since any lineage can be shown to be non-exclusive if looked at closely enough, even body cells (Frost & Kluge 1995). Exclusive species were therefore defined by Baum and Shaw (1995) as 'a basal group of organisms all of whose genes coalesce more recently with each other than with those of others outside the group'.

The problems with history-based approaches have considerable bearing on practical aspects of taxonomy. History can only be inferred indirectly from character distributions. Rejecting character-based species concepts introduces the considerable difficulty of how you recognise species as such. A further problem with history-based approaches is concerned with future events. If two indistinguishable but isolated populations are investigated there is no way of knowing whether they will remain isolated and give rise to new species, come together again

and resume tokeogenetic relationships or become extinct. As a result internodal species cannot be distinguished until they are separated by identifiable characters or become extinct.

vi) The composite species concept

An attempt to reconcile history and character-based species concepts has been made with the development of the 'composite species concept' (Kornet 1993). Kornet (1993) pointed out that every isolated population is a potential new lineage and the internodal species concept would treat them as new species. This leads to the problem of recognising more or less every isolated population as a species and moreover, that the fate of these populations cannot be determined. Kornet (1993) overcomes this problem by defining composite species as lineages of internodons which begin with the fixation of a new character in an ancestral internodon and end by extinction or with another fixation in a descendant internodon. The concept is composed of internodal lineages which are also taxa since they are defined by novel characters (autapomorphies) which can be investigated. Introducing the fixation of characters allows for the identification of the beginnings and endings of internodons and of the limits of tokogenetic relationships in exclusive lineages. Composite species defined in this way are paraphyletic groups of internodons, only becoming monophyletic when they become extinct.

vii) Species concept used in this study

My attempts at species delimitation were based on a theoretical acceptance of evolutionary processes and the need to incorporate them into a consistent species concept but at the same time recognising the practical basis of taxonomy. My method of delimiting species was basically to follow the traditional approach of delimitation by morphological similarity and then to search for consistent discontinuities in character state distribution. In the absence of detailed information about micro-evolutionary and macro-evolutionary processes in the species studied the only meaningful species concept which could be applied to *Hypenia* was the taxonomic species concept.

viii) Paraphyletic species

If speciation events occur when lineages split then, by definition, species which are paraphyletic cannot exist. However, the possibility of paraphyletic species has been discussed in the literature. For example Baum and Shaw (1995) discuss the concept of the 'metaspecies'. If two groups of lineages of differing sizes co-exist the smaller will coalesce earlier than the larger and so the smaller group of lineages will be a species for some time

before the larger 'metaspecies'. A similar concept has also been discussed using the terms 'paraspecies' and 'cladospecies' which are derived from within the paraspecies (Ackery & Vane Wright 1984). Olmstead (1995) using the words 'plesiospecies' and 'apospecies' to respectively describe species which are only defined by plesiomorphies or apomorphies. Crisp and Chandler (1996) investigated the concept of metaspecies and paraspecies with examples from the Australian flora and concluded that investigating and accomodating paraphyletic species in taxonomy is necessary to an increased understanding of evolutionary patterns.These concepts appear to be appropriate to *Hypenia,* for example *H. reticulata* may well be described as a plesiospecies and other non-resupinate members of the 'macrantha complex' as apospecies.

ix) Infra-specific taxa

Taxonomists often recognise the need for infra-specific categories to accomodate patterns of variation found within species. The most commonly used infra-specific ranks are subspecies and variety. General principles have been formulated to decide which rank is most appropriate. Davis and Heywood (1963) define a supspecies as "a considerable segment of a species with a distinct area and a distinct morphology" and variety as " a local *facies* of a species ..., morphologically distinct and occupying a restricted geographical area". Thus the distinction between subspecies and variety, and even between subspecies and species, is not clearly made. A survey of the use of infra-specific taxa in published monographs found no consistency in the method of their application (Hamilton & Reichard 1992). There was consistency in the use of infra-specific ranks by individuals but there was little or no consistency between them. However, there was a tendency to reverse the method of application of subspecies and variety between American and European authors. Because of the lack of consistent usage for infra-specific ranks and because their usage implies a level of knowledge about relationships which I did not feel was justified, I have not used infra-specific categories in the taxonomy of *Hypenia.*

C. Section *Hypenia* : the 'macrantha complex'

Species delimitation in section *Hypenia,* or the 'macrantha complex', presented the biggest difficulty in *Hypenia* and the taxonomy of the group is discussed below.

The 'macrantha complex' is widely distributed in central Brazilian savannas (figures 10-10, 10-11 and 10-12) but from the middle of the nineteenth century until the middle of this century the flora of central Brazil remained one of the least explored in the world. There was

a relative flurry of activity in the first half of the nineteenth century which produced the collections available to Bentham. In the latter half of the century, Glaziou was the only major collector and unfortunately the locality information of his collections can be unreliable (Wurdack 1970). As a result Epling, working mostly in the first half of this century, had only a little more information available to him than Bentham. Epling (1949) recognised more or less the same taxa as Bentham and described several new ones including *H. perplexa, H. aristulata* and *H. pauliana*, all in the 'macrantha complex'. There were still very few specimen citations for each species.

After Epling's last major publication on *Hyptis* (Epling 1949), more collections were made in central Brazil and it became apparent that there was a great deal of variation in the 'macrantha complex' particularly in Epling's key characters of indumentum and leaf size and shape. These characters are very variable and display few discontinuities and all the names previously published in *Hypenia* subsection *Laxiflorae* were later synonymised under *Hyptis macrantha* (Epling & Mathias 1957). Thus the 'macrantha complex' has been subject to extremes of splitting and lumping, even by the same author.

Resupination of the flower was a previously overlooked character which proved to be important in classification in the section. The six species with resupinate flowers, i.e. subsection *Ellipticae,* were relatively easy to define and are all narrow endemics with restricted geographical ranges. They are defined by distinct autapomorphies, e.g. the broad phyllomes and bracteoles of *H. calycina,* the yellow, clustered flowers of *H. crispata* and the long, exserted corollas and dentate leaves of *H. niquelandiensis.*

In subsection *Laxiflorae*, i.e. species with non-resupinate flowers, I have recognised eight species, two with a wide geographical range, *H. reticulata* in eastern and southern savannas and *H. macrosiphon* in western and northern savannas, plus six geographically restricted endemics. The six geographically restricted taxa have well-defined autapomorphies, e.g. *H. aristulata* with aristulate calyx teeth and long hairs on the upper stem and *H. caiaponiensis* with its distinctive inflorescence and minute glandular hairs in the inflorescence. *H. recoria* is defined by its broad bracteoles and the taxon can be correlated with ecological preferences as it is apparently confined to the savanna margins of gallery forest. However, once the well-defined taxa are extracted from the non-resupinate group a large number of specimens from a wide geographical area are left over. It has proved very difficult to find convincing ways to split them although they represent a bigger range of morphological diversity than other

recognised species. I have split this diversity into two taxa, *H. reticulata* and *H. macrosiphon*, on the basis of leaf and inflorescence characters but taxon delimitation remains problematic. This is discussed further below.

i) Variation in *H. reticulata* and *H. macrosiphon*

The two taxa, *H. reticulata* and *H. macrosiphon* were identified in the same way as the other species in the 'macrantha complex' by using the traditional taxonomic approach, i.e. placing specimens which look most alike into piles and then trying to search for characters which can be used to reliably identify all the members of each pile. Within *H. reticulata* in particular, the initial stages of this process produced many piles because the specimens displayed considerable morphological diversity. Each pile could often be correlated with geography and the locality of many specimens could be identified from their morphology. Equally however, many specimens could not be placed in any particular pile and some localities produced specimens of widely diverging appearance. Defining characters also proved elusive as none of the most variable, and easily described, characters were consistently correlated with each other or with geography.

H. macrosiphon proved to be less variable than *H. reticulata* and was separated by a combination of a number of characters. The type of *H. macrosiphon*, Kuntze s.n. has distinctive leaves which are broad and partially lobed, have a cordate base and conspicuously dentate margin. The inflorescence has short, glandular indumentum throughout and the single-flowered cymes are borne on short peduncles (< 10mm). The Kuntze collection is from Mato Grosso, in the west of Brazil and most collections from the same region have similar leaf and inflorescence characters. *H. macrosiphon* is defined in this revision by the leaves, which are variable in shape but tend to be broad with a truncate or cordate base and always have a coarsely dentate margin, and by the indumentum on at least the most terminal branches of the inflorescence. Collections from the area of Cristalina in central Goiás have sparsely hairy leaves and conspicuous globose fistulae, characters which are also seen in collections from Paraguay and Mato Grosso do Sul. However, the leaf shape and indumentum is typical for *H. macrosiphon* and the lack of geographical coherence of these specimens lead me to treat them as part of *H. macrosiphon*.

H. reticulata is the most variable species and is very widespread in southern and eastern Brazil where it occurs in intensively collected but fragmented areas of savanna. The species is characterised by its ovate to lanceolate leaves with serrate or crenate margins and glabrous

inflorescence branches with widely-spaced cymes. These characters are all found in other species of section *Hypenia* and in effect the principal definition of *H. reticulata* is derived from its lack of defining characters. In other words, *H. reticulata* is what is left over when other species with defining characters in subsection *Hypenia* have been separated.

Individual populations can be recognised on the basis of variation in a number of characters. The indumentum of the lower leaf varies considerably, some collections are sparsely pubescent whereas others are tomentose. Leaf indumentum is mostly constant at each geographical locality, many collections from Minas Gerais have tomentose indumentum (this character contributed to the recognition of *H. perplexa* by Epling). However, collections from the same locality can vary from tomentose to sparsely pubescent, this can be seen at Mogi-Guaçu in the state of São Paulo. The shape of the leaf at this locality also varies, the type of *H. pauliana,* Burchell 5522, has cordate leaves with dentate margins and tomentose lower surface but collections from nearby Mogi-Mirim have larger, ovate leaves with serrate margins and sparsely pubescent lower surface.

Most collections have dense hairs in the calyx throat but those from Paraná are usually sparsely hairy or glabrous. The type of *H. glauca,* St Hilaire 1447, closely resembles Paraná collections, although it was collected in southern Minas Gerais. Some São Paulo specimens are also very similar to the form represented by St Hilaire 1447. Another recognisably distinct form occurs in at Serra do Cipó in Minas Gerais. This population has glandular pubescent peduncles and upper inflorescence branches and individuals often have relativly short peduncles. Collections from this locality are identifiable using these characters. However, there are collections from Serra do Cipó with long, sparsely pubescent peduncles which are scarcely distinguishable from collections made at other localities in Minas Gerais.

Collections from Distrito Federal and surrounding parts of Goiás are relatively geographically isolated and this is reflected in the vegetative morphology of collections from this area. The leaves are distinctly lanceolate but inflorescence and floral morphology is scarcely distinguishable from more southerly and easterly localities.

I have found it impossible to find consistent, easily described characters to maintain boundaries between previously described species or to justify describing new ones. Hence I have taken a very broad view of *H. reticulata,* although I recognise that as circumscribed here it is perhaps a different entity to the other species in section *Hypenia* (see discussion

under paraphyletic species above). The pattern of variation in *H. reticulata* also precluded the recognition of infra-specific taxa and no subspecies or varieties have been recognised. The type of *H. reticulata* is the type for *Hypenia* and for this reason this name is retained over others described at the same time by Bentham in 1832.

Chapter 10: Formal taxonomy

A. Introduction

This chapter presents the formal taxonomic accounts for the species formerly included in *Hypenia.* It is preceded by an introduction to their previous classification followed by my arrangement of *Hypenia* species as they now are in *Eriope.* All *Hypenia* species and sections of *Eriope* are included in a key. This is followed by the systematic description of the species of *Hypenia.* A taxonomic index is presented. Distribution maps for all the species described are included at the end of the chapter.

i) Previous sectional and subsectional classification of *Hypenia*

In Bentham's original work on *Hyptis* section *Hypenia* (Bentham 1833) no infra-sectional classification was included. Bentham later revised section *Hypenia* in De Candolle's Prodromus (1848) by adding several species and dividing the section into subsections *Laxiflorae* and *Densiflorae.* The subsections were distinguished by the open paniculate inflorescence of *Laxiflorae* compared to the crowded inflorescence of *Densiflorae.*

The first flora account of what amounts to *Hyptis* section *Hypenia* was in *Flora Brasiliensis* (Schmidt 1858). Schmidt's classification was similar to that of Bentham (1848) but he called the section *Paniculatae* with subsections *Confertiflorae* and *Laxiflorae*. Subsection *Laxiflorae* corresponded closely to Bentham's subsection of the same name but *Confertiflorae* included all Bentham's *Densiflorae* with the addition of a number of species now classified elsewhere in *Hyptis, Eriope* or *Hyptidendron.*

Briquet (1896) also published a classification of *Hyptis* section *Hypenia.* He included seven subsections. Subsection *Densiflorae* was more or less equivalent to Bentham's (1848) *Densiflorae* although Briquet separated *Hyptis irregularis* and *H. concinna* into subsection *Irregulares*; subsection *Paniculatae* included three species placed in *Hyptis* section *Minthidium* by all other authors; and subsections *Longiflorae, Ellipticae, Coarctatae* and *Laxiflorae* were all divisions of Bentham's subsection *Laxiflorae.* Briquet did not assign types to any of his subsections and these have all been lectotypified in this account apart from *Laxiflorae* sensu Briquet. This taxon contained three species, all of which were included in subsection *Laxiflorae* sensu Bentham, but excluded the type. The use of the name *Laxiflorae* by Briquet is confusing and is not considered further in the taxonomic accounts.

The most significant monographic work on *Hypenia* after Bentham was by Epling, first published in 1936 as part of his *Synopsis of South American Labiatae*. This was used as the basis of a monograph of *Hyptis*, published in Spanish (Epling 1949) which remains the most recent species-level revision of *Hypenia*. In both works Bentham's (1848) infra-sectional classification of *Hypenia* was retained although Epling (1936) transferred species from Bentham's *Hyptis* section *Siagonnarhen* into *Hypenia*. These additional species have all been transferred out of *Hypenia* into *Eriope* (Harley 1976, 1988a). Harley also described two new species (Harley 1974) and elevated *Hypenia* from sectional to generic rank (Harley 1988a).

Major taxonomic works relevant to *Hypenia* (Bentham 1848, Briquet 1896, Epling 1949) are summarised in table 10-1 with all the species which have been included in *Hypenia* sensu Harley (1976, 1988a) listed and their past classification indicated.

Table 10-1: Summary of past classifications of *Hypenia*

Species	Bentham (1848)	Briquet (1896)	Epling (1949)	Harley (1976 & 1988a)
aristulata Epling			*Laxiflorae*	***Hypenia***
blanchetii Benth.	*Densiflorae*	*Densiflorae*	*Densiflorae*	***Eriope***
brachystachys Pohl ex Benth.	*Densiflorae*	*Densiflorae*	*Densiflorae*	***Hypenia***
calophylla St.Hil.ex Benth.	*Laxiflorae*	*Laxiflorae*	syn. of *vitifolia*	
calycina Pohl ex Benth.	*Laxiflorae*	*Ellipticae*	*Laxiflorae*	***Hypenia***
campanulata Benth.	*Laxiflorae*	*Laxiflorae*	syn. of *simplex*	
coccinea Mart.ex Benth.	*Laxiflorae*	*Longiflorae*	syn. of *gardneriana*	
concinna Benth.	*Densiflorae*	*Irregulares*	*Densiflorae*	***Hypenia***
crispata Pohl ex Benth.	*Laxiflorae*	*Coarctatae*	*Laxiflorae*	***Hypenia***
densiflora Pohl ex Benth.	*Densiflorae*	*Densiflorae*	*Densiflorae*	***Hypenia***
durifolia Epling			*Laxiflorae*	***Hypenia***
effusa S. Moore			*Laxiflorae*	***= micrantha***
elegans Briq.ex Mich.		*Pubescentes*	§ *Minthidium*	
floribunda Briq.ex Mich.		*Pubescentes*	§ *Minthidium*	
gardneriana Epling			*Laxiflorae*	***Hypenia***
glauca var. *gardneriana* Benth.	*Laxiflorae*		syn. of *gardneriana*	
glauca St.Hil.ex Benth.	*Laxiflorae*	*Longiflorae*	*Laxiflorae*	***Hypenia***
heterantha Benth.	§ *Siagonarrhen*	§ *Siagonarrhen*	*Densiflorae*	***Eriope***
		§§ *Nudiflorae*		
hypoleuca Benth.	§ *Siagonarrhen*	§ *Siagonarrhen*	*Densiflorae*	***Eriope***
		§§ *Nudiflorae*		

irregularis Benth.	*Densiflorae*	*Irregulares*	*Densiflorae*	***Hypenia***
latifolia Mart.ex Benth.	§ *Siagonarrhen*	§ *Siagonarrhen* §§ *Nudiflorae*	*Densiflorae*	***Eriope***
laxiflora Mart.ex Benth.	*Laxiflorae*	*Laxiflorae*	syn. of *reticulata*	
lindmaniana Briq.			syn. of *macrosiphon*	
longiflora Pohl ex Benth.	*Laxiflorae*	*Longiflorae*	syn. of *reticulata*	
macrantha St. Hil. ex Benth.	*Laxiflorae*	*Ellipticae*	*Laxiflorae*	***Hypenia***
marifolia Benth. in DC.	*Densiflorae*	*Densiflorae*	*Densiflorae*	***Hypenia***
melochioides St. Hil. ex Benth.	*Densiflorae*	*Densiflorae*	syn. of *densiflora*	
micrantha Benth.	***Eriope***	***Eriope***	*Laxiflorae*	***Hypenia***
paniculata Benth.	*Laxiflorae*	*Longiflorae*	*Laxiflorae*	***Hypenia***
pruinosa Pohl ex Benth.	*Densiflorae*	*Densiflorae*	*Densiflorae*	***Hypenia***
reticulata Mart.ex Benth.	*Laxiflorae*	*Longiflorae*	*Laxiflorae*	***Hypenia***
salzmannii Benth.	*Laxiflorae*	*Laxiflorae*	*Laxiflorae*	***Hypenia***
salzmannii var. *filipes* Benth.	*Laxiflorae*		syn. of *salzmannii*	
simplex St.Hil.ex Benth.	*Laxiflorae*	*Laxiflorae*	*Laxiflorae*	***Eriope***
vitifolia Pohl ex Benth.	*Laxiflorae*	*Longiflorae*	*Laxiflorae*	***Hypenia***

Names in bold are generic epithets

§ Sections of *Hyptis*

§§ Subsection of section *Siagonarrhen*

All other infrageneric names refer to subsections of *Hyptis* section *Hypenia*.

Note on the name *Hyptis*

The epithet *Hyptis* was first used by Jacquin (1786) but the name *Mesosphaerum* was adopted by Kuntze in *Revision Genera Plantarum* (1891) on the basis of the earlier publication of one species, *Mesosphaerum hirsutum* (a synonym of *Hyptis suaveolens*), by P. Browne in his work on Jamaica (1756). The name *Hyptis* Jacq. was conserved against *Mesosphaerum* P. Browne and another earlier name, *Condea* Adams.

ii) Specimens seen

Specimens were seen at the following herbaria in the UK: K, E, BM; and in Brazil: R, RB, SPF, SP, MBM, UEC, UB, HEPH, IBGE and UEFS. Exsiccatae were obtained on loan from: P, G, S, NY, UC, G, W and M. All specimens cited have been seen unless stated otherwise.

Abbreviations

Abbreviations of books follow Stafleu and Cowan (1976-1988) and journals follow the *Botanico-Periodicum-Huntianum* (Lawrence et al. 1965) and its Supplement (Bridson 1991). Abbreviations for herbaria follow Holmgrem et al. (1990).

iii) Classification of the species of *Hypenia*

The classification of *Hypenia* and *Eriope* discussed in chapter 8 has been used as the basis for the taxonomic accounts which follow. The infra-generic classification and character distribution is summarised in table 10-2. This thesis does not constitute effective publication of new names or combinations.

Table 10-2 : Summary of current classification of *Eriope* and character distribution between sections

Section	Character distribution					
	Stylopodium	Corolla shape	Corolla colour	Corolla orientation	Cymes	Branched hairs
Vitifoliae (2)	0	0	Lilac	0	0	0
Micranthae (1)	0	0	Lilac	0	0	1
Irregulares (2)	0	1	White or pink	0	1	0
Salzmanniae (1)	0	1	Blue	0	1	0
Densiflorae (3)	0	1	White or lilac	0	0/1	0
Hypenia: Laxiflorae (8)	0	1	Red (white or lilac)	0	1	0
Hypenia: Ellipticae (6)	0	1	Red (yellow)	1	1	0
Eriope * (37)	1 (0 *Eriopdion)*	0 (1*Eriopidion*)	Lilac (pink)	0	1	0/1

Species numbers are in brackets. Stylopodium: 0 absent, 1 present; corolla shape: 0 funnel-shaped, 1 tubular; corolla orientation: 0 not resupinate, 1 resupinate; cymes: 0 multi-flowered, 1 single-flowered; branched hairs: 0 absent; 1 present.

* The infra-sectional classification of section *Eriope* is not covered in this account. Species numbers in *Eriope* are from Harley's unpublished scheme in appendix I and include *Eriopidion*.

B. Generic description

Eriope *Humb. & Bonpl. ex Benth.*

Type: *Eriope crassipes* Benth., Labiat.Gen. Spec.:144 (1833).
Eriope Humb. & Bonpl. ex Benth. Labiat. Gen. Spec.:142 (1833). Benth. in DC. Prodr. 12, 140 (1848); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85:188 (1936); Harley in Hooker's Icon. Pl. XXXVIII part III (1976).

Hyptis section *Hypenia* Mart. ex Benth., Labiat.Gen. Spec.: 136 (1833); Benth. in DC. Prodr. 12:137 (1848); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85:224 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7:219 (1949). Type: *Hyptis reticulata* Mart. ex Benth.
Hyptis section *Paniculatae* Schmidt in Fl. Bras. 8(1):146 (1858). **Lectotype:** *Hyptis reticulata* Mart. ex Benth.
Eriopidion Harley in Hooker's Icon. Pl. XXXVIII part III:103 (1976). Type: *Eriopidion strictum* (Benth.) Harley loc. cit. *Eriope stricta* Benth. in DC, Prodr. 12: 142 (1848).
Hypenia (Mart. ex Benth.) Harley in Bot. J. Linn. Soc. 98:91 (1988). Type: *Hypenia reticulata* (Mart. ex Benth.) Harley loc. cit. *Hyptis macrantha* St. Hil. ex Benth., Labiat. Gen. Spec.:140 (1833).

Subshrubs, shrubs or herbs with woody rootstocks. Stems often virgate, often terete, and glabrous with a waxy coating, fistulose and sometimes with globose swellings; or stems not virgate, somewhat angled and with dense or moderate indumentum and internodes not fistulose or swollen. Indumentum of simple and glandular hairs, branched hairs rarely present. Leaves petiolate, rarely more or less sessile, simple. Inflorescence an indeterminate thyrse; often large with well-developed branches and widely spaced cymes, sometimes branches reduced and cymes crowded. Inflorescence phyllomes < 5 mm long, subulate, caducous or persistent; rarely > 5 mm long, ovate and persistent. Cymes often consisting of a single flower, sometimes many-flowered; first axes of many-flowered cymes either conspicuously developed or cyme more or less sessile. Flowers subtended by paired bracteoles immediately below the calyx, rarely bracteoles absent; sometimes whole flower resupinate. Calyx ten-nerved; equally five-lobed, or posterior three lobes more or less connate, posterior lobes frequently expanded and deflexed if connate; calyx throat naked or obscured by long hairs; calyx tube cylindrical or campanulate; calyx accrescent in fruit.

Corolla white, pink, violet, blue, red or yellow, sometimes yellow or brown in bud; corolla tube cylindrical or funnel-shaped, 3 — 26 mm long; posterior lobes erect; lateral lobes deflexed, rarely pointing forward; anterior lobe boat-shaped with thickened base and fimbriate margins holding stamens under pressure, retracting with a mechanical stimulus and releasing stamens explosively. Stamens dorsifixed, opening by longitudinal slits; posterior filaments densely hairy. Stylopodium at base of style overtopping nutlets or absent. Chromosome numbers of 2n = 12, 20, 22 and 28.

DISTRIBUTION. Circa 60 species in South America, mostly Brazil, between 5°N and 24°S and 35°W and 65°W.

C. Section descriptions

i) Section Vitifoliae *R. Atkinson* sect. nov.

Type: *E. vitifolia* (Pohl ex Benth.) R. Atkinson comb. nov.
Hyptis vitifolia Pohl ex Benth.

Sectioni Eriopi affinis sed cymae floribus pluribus, bracteolis et stylopodiis absentibus differt.

Stems terete, glabrous and waxy; internodes fistulose, often swollen. Branched trichomes absent. Inflorescence large with well-developed branches and widely spaced cymes; inflorescence phyllomes subulate, caducous. Cymes multiflowered, first axes conspicuously developed; paired bracteoles absent from individual flowers. Flowers not resupinate. Calyx equally five-lobed. Corolla violet, brown in bud; corolla tube funnel-shaped, 3 — 4 mm long. Stylopodium absent. Chromosome numbers of 2n = 22 and 28.

This section consists of two species and is closely allied to *Eriope* section *Eriope*. It is readily distinguished by the multi-flowered cymes and the lack of bracteoles subtending individual flowers. In addition the habit of both species is very distinct and is characterised by the swollen internodes (which differ in shape between the two species), conspicuously waxy stems, many setose trichomes and relatively large, often lobed leaves.

Distribution: Savanna (cerrado and campo rupestre) in the Serra do Espinhaço in Minas Gerais and Bahia, northeastern Brazil.

ii) Section Micranthae *R. Atkinson* sect. nov.

Type: *E. micrantha* Benth. in DC.

Sectioni Vitifoliis affinis sed caulibus pubescentibus, internodiis non fistulosis, nec tumidis, trichomatibus ramosis presentibus, phyllomatibus ovatis, cymae floribus singularibus et bracteolis presentibus differt.

Stems somewhat angled with dense indumentum; internodes not fistulose or swollen. Branched trichomes present. Inflorescence large with well-developed branches and widely spaced cymes; inflorescence phyllomes ovate > 5 mm long, persistent. Cymes one-flowered, first axes conspicuously developed. Paired bracteoles present. Flowers not resupinate. Calyx equally five-lobed. Corolla violet; corolla tube funnel-shaped, ca. 3.5 mm long. Stylopodium absent.
Chromosome number not known.

The relatively small, funnel-shaped corolla suggests the close relationship of this section with section *Vitifoliae* but the single-flowered cymes and bracteolate flowers distinguish it from the preceding section. The absence of the stylopodium distinguishes it from section *Eriope* and the presence of branched trichomes is unique in those species of *Eriope* which lack a stylopodium (i.e. *Hypenia* sensu Harley 1988a).

Distribution: one locality in savanna (cerrado) near Cuiabá, Mato Grosso, western Brazil.

iii) Section Irregulares *(Briq.) R. Atkinson* comb. & stat. nov.

Type: *E. irregularis* (Benth. in DC.) R. Atkinson comb. nov.
Hyptis irregularis Benth. in DC.
Hyptis section *Hypenia* subsection *Irregulares* Briq. in Nat. Pflanzenfam. ed. 1, IV, 3a: 335 (1896). **Lectotype:** *Hyptis irregularis* Benth. in DC, chosen here.

Stems somewhat angled with moderate indumentum; internodes not fistulose or swollen. Branched trichomes absent. Inflorescence large with well-developed branches and widely spaced cymes or branches reduced and cymes crowded. Cymes one-flowered, more or less

sessile. Flowers not resupinate. Calyx equally five-lobed. Corolla violet or white; corolla tube cylindrical, ca. 5 mm long. Stylopodium absent. Chromosome number not known.

The tubular flowers of section *Irregulares* suggest close relationship with section *Densiflorae* but is distinguished by the regular calyx lobes and absence of waxy stems and swollen internodes. The inflorescence structure can be superficially similar to that of section *Densiflorae* (in *E. irregularis*) but differs in the absence of multi-flowered cymes.

Distribution: Savanna (cerrado) in western Bahia and northern Goiás, central Brazil.

iv) Section Salzmanniae *R. Atkinson* sect. nov.

Type: *E. salzmannii* (St. Hil. ex Benth.) R. Atkinson comb. nov.
Hyptis salzmannii St. Hil. ex Benth.

Sectioni Hypeniae affinis sed caulibus glabris et ceraceis, internodiis fistulosis atque semper tumidus, corollis azureis recedit.

Stems terete, glabrous and waxy; fistulose, often swollen. Branched trichomes absent. Inflorescence large with well-developed branches and widely spaced cymes. Cymes one to three-flowered, first axes well-developed. Flowers not resupinate. Calyx equally five-lobed. Corolla blue, yellow in bud; corolla tube cylindrical, 4— 5 mm long. Stylopodium absent. Chromosome number 2n = 12.

Section *Salzmanniae* shares a similar inflorescence and floral morphology with section *Hypenia* but is readily distinguished by the smaller, blue flowers and globose swellings on the stem.

Distribution: Savanna (cerrado, caatinga, campo rupestre, restinga) throughout northeastern Brazil with a disjunction in Venezuela.

v) Section Densiflorae *(Benth. in DC.) R. Atkinson* comb. & stat. nov.

Type: *E. densiflora* (Pohl ex Benth.) R. Atkinson comb. nov.
Hyptis densiflora Pohl ex Benth.
Hyptis section *Hypenia* subsection *Densiflorae* Benth. in DC. Prodr.12: 135 (1848).

Hyptis section *Paniculatae* subsection *Confertiflorae* Schmidt in Fl. Bras. 8(1): 146 (1858). **Lectotype:** *Hyptis densiflora* Pohl ex Benth., chosen here.
Hyptis section *Hypenia* subsection *Densiflorae* Briq. in Nat. Pflanzenfam. ed. 1, IV, 3a: 336 (1896). **Lectotype:** *Hyptis densiflora* Pohl ex Benth., chosen here.

Stems terete, glabrous and waxy; internodes fistulose, often swollen or sometimes stems somewhat angled with moderate indumentum, in which case swollen internodes absent. Branched trichomes absent. Inflorescence large with reduced branches and cymes crowded. Cymes one to three-flowered (or more), more or less sessile. Flowers not resupinate. Calyx with posterior three lobes connate at base, rarely not connate. Corolla white or violet; corolla tube cylindrical, 6 — 12 mm long. Stylopodium absent. Chromosome number 2n = 20.

Section *Densiflorae* is similar to section *Irregulares* but differs in the presence of multi-flowered cymes which are densely crowded on the indeterminate branches. In addition the three posterior calyx lobes are usually connate at the base. Stems are usually distinctly waxy and have conspicuous swellings which vary in shape characteristically for each species. Distribution: Savanna (cerrado) in central and northern Goiás and Distrito Federal, central Brazil.

vi) Section Hypenia *(Mart. ex Benth.) R. Atkinson* comb. nov.

Type: *E. reticulata* (Mart. ex Benth.) R. Atkinson comb. nov.
Hyptis reticulata Mart. ex Benth.
Hyptis section *Hypenia* Mart. ex Benth., Labiat.Gen. Spec., 136 (1833); Benth. in DC. Prodr., 12: 137 (1848); Briq. in Nat. Pflanzenfam., ed. 1, IV, 3a: 334 (1896); Epling in Repert. Spec. Nov. Regni Veg. Beih., 85: 224 (1936); Epling in Revista Mus. La Plata, secc. Bot., 7: 219 (1949). Type: *Hyptis reticulata* Mart. ex Benth.
Hyptis section *Paniculatae* Schmidt in Fl. Bras. 8(1): 146 (1858).
Hypenia (Mart. ex Benth.) Harley in Bot. J. Linn. Soc. 98: 91 (1988).

Stems terete, glabrous and waxy; internodes fistulose, often swollen or sometimes stems somewhat angled with moderate indumentum, in which case internodes not fistulose or swollen. Branched trichomes absent. Inflorescence large well-developed branches and cymes widely spaced. Cymes one-flowered, rarely two or three-flowered, first axes well-developed. Flowers resupinate or not resupinate. Calyx more or less equally five-lobed. Corolla red,

rarely yellow or white; corolla tube cylindrical, 9 — 26 mm long. Stylopodium absent. Chromosome number 2n = 20.

Section *Hypenia* has similar tubular flowers and large inflorescence with single-flowered cymes and well-developed indeterminate axes to section *Salzmanniae.* Uniquely in *Eriope,* the majority of species have red flowers which are considerably larger than flowers of all other species in the Hyptidinae. One species, *E. subrosea,* has white flowers which are similar in size and shape to those in section *Densiflorae* but the place of this species in *Hypenia* is doubtful. The two subsections are distinguished by the orientation of the corolla which is resupinate in subsection *Ellipticae* and non-resupinate in subsection *Laxiflorae.* Additionally, the flowers of subsection *Ellipticae* tend to be larger, held closer together and be more erect in the infloresecence.

Distribution: Savanna (cerrado and campo rupestre) in parts of the states of São Paulo and Minas Gerais and throughout Goiás and Mato Grosso, central Brazil. Savanna (cerrado) regions in Paraguay and Bolivia.

Subsection Laxiflorae *(Benth. in DC.) R. Atkinson* comb. nov.

Type: *E. reticulata* (Mart. ex Benth.) R. Atkinson comb. nov.
Hyptis reticulata Mart. ex Benth.

Hyptis section *Hypenia* subsection *Laxiflorae* Benth. in DC. Prodr., 12: 137 (1848); Briq. in *Nat. Pflanzemfam.* ed. 1, IV, 3a: 335 (1896); Epling in Repert. Spec. Nov. Regni Veg. Beih., 85: 228 (1936); Epling in Revista Mus. La Plata, secc. Bot., 7: 229 (1949). Type: *Hyptis reticulata* Mart. ex Benth.
Hyptis section *Paniculatae* subsection *Laxiflorae* Schmidt in Fl. Bras. 8: 152 (1858).
Lectotype: *Hyptis reticulata* Mart. ex Benth., chosen here.
Hyptis section *Hypenia* subsection *Longiflorae* Briq. in Nat. Pflanzenfam. ed 1, IV, 3a: 335 (1896).
Lectotype: *Hyptis reticulata* Mart. ex Benth., chosen here.

Flowers not resupinate.
Distribution: Savanna (cerrado) in central Brazil and neighbouring parts of Bolivia and Paragauy.

Subsection Ellipticae *(Briq.) R. Atkinson* comb. nov.

Type: *E. macrantha* (St. Hil. ex Benth.) R. Atkinson comb. nov.

Hyptis macrantha St. Hil. ex Benth.

Hyptis section *Hypenia* subsection *Ellipticae* Briq. in Nat. Pflanzenfam. . ed. 1, IV, 3a: 335 (1896). **Lectotype** *Hyptis macrantha* St. Hil. ex Benth., chosen here.

Hyptis section *Hypenia* subsection *Coarctatae* Briq. in Nat. Pflanzenfam. . ed. 1, IV, 3a: 335 (1896). **Lectotype:** *Hyptis crispata* Pohl ex Benth., chosen here.

Flowers resupinate.

Distribution: Savanna (cerrado) in central Brazil, especially Distrito Federal, neighbouring parts of Goiás and northwestern Minas Gerais.

D. Key to sections of *Eriope*

A key is presented here to the sections of *Eriope*. The stylopodium is an invaluable aid to identification and is used as the first lead in the key. However, the stylopodium is absent in *Eriopidion* which, for the purposes of this study, is treated as part of section *Eriope*. As a result *Eriopidion* is keyed out separately from the rest of section *Eriope*. The constituent species of the two monotypic sections, *Micranthae* and *Salzmanniae*, are given in brackets after the section names.

1 Stylopodium present ..... sect. *Eriope* (less *Eriopidion*)

Stylopodium absent 2

2 Fruiting calyx with lobes obscuring throat, opening with a hygroscopic mechanism. Long virgate stems absent. Nutlets triquetrous in section ..... *Eriopidion*

Fruiting calyx with lobes not obscuring throat, hygroscopic mechanism absent. Long virgate stems present. Nutlets ovoid in section 3

3 Corolla funnel-shaped, </= 4 mm long, violet 4

Corolla cylindrical, > (/=) 4 mm long, white, blue, yellow, red, violet 5

4 Cymes 1-flowered; paired bracteoles present. Branched hairs present on leaf upper surface. Stems with wax absent; internodes not fistulose or swollen ....sect. *Micranthae* **(3. micrantha)**

Cymes many-flowered; paired bracteoles absent. Branched hairs absent. Stems with wax present; internodes fistulose, sometimes swollen ..... A. sect. *Vitifoliae*

5 Cymes 1.5 — 60 mm apart, branches visible, cymes 1-flowered. Posterior three calyx lobes not connate..... 6

Cymes densely clustered and obscuring branches, most cymes > 1-flowered. Posterior three lobes usually connate at base.....B. sect. *Densiflorae*

6 Corolla < = 5 mm 7

Corolla > = 9 mm .....D. sect. *Hypenia*

7 Corolla sky blue. Stems with wax present, internodes fistulose with slightly to moderately inflated, hard, swellings ..... sect. *Salzmanniae* **(6. salzmannii)**

Corolla white. Stems with wax absent; internodes not fistulose or swollen ..... C.sect. *Irregulares*

E. Key to species of *Eriope* formerly included in *Hypenia*

A key is presented to the species of *Eriope* formerly included in *Hypenia.* The species are keyed out according to section. Monotypic sections (i.e. *Micranthae* and *Salzmanniae*) are not included here, see preceding key. The most problematic part of the key is in section *Hypenia* (the 'macrantha complex'). There are lamentably few simple presence / absence characters in section *Hypenia,* particularly subsection *Laxiflorae.* This introduced difficulties in producing effective contrasting leads in the key. Leaf characters and inflorescence arrangement were important in delimiting species. However, neither set of characters provided simply described leads and the distribution of indumentum was found to be a useful additional source of characters for the key. Some quantitative characters are described by qualitative adjectives in the key, particularly the arrangement of cymes in subsection *Ellipticae*. The figures given in the descriptions include the full range of variation in cyme distance and tend to obscure the overall pattern of the cyme arrangement. Thus, even in species with crowded cymes there will be some cymes which are isolated. In this case using quantitative figures obscures the value of the character.

Unfortunately, however, the key to subsection *Laxiflorae* is not easy to use. Reference must be made to species descriptions and geographical distribution for reliable identification in the subsection.

i) Section *Vitifoliae*

Internodes fistulose with moderately to extremely inflated, easily compressible, sometimes asymmetric, swellings ..... **1. vitifolia**

Internodes fistulose, swollen along their length ..... **2. longicaulis**

ii) Section *Densiflorae*

1 Calyx teeth broadly deltoid, posterior 3 connate at base 2

Calyx teeth aristulate, symmetric ..... **8. concinna**

2 Leaves petiolate; lamina lanceolate to ovate, 30 — 65 x 7 — 20 mm; base acute or truncate; apex acute to rounded; margin serrate. Flowering calyx greenish-yellow. Corolla tube white ..... **9. densiflora**

Leaves sessile or shortly petiolate; lamina ovate, 4 — 30 x 2.5 — 25 mm; base shallowly cordate; apex obtuse or mucronate, sometimes acute; margin crenate. Flowering calyx vinaceous. Corolla tube violet ..... **7. brachystachys**

iii) Section *Irregulares*

Inflorescence few-branched; cymes overlapping or spaced, 1.5 — 30 mm apart; branches moderately pubescent. Leaf lower surface sparsely to moderately pubescent with simple hairs and subsessile glands ..... **4. irregularis**

Inflorescence branched; cymes spaced 3 — 30 mm apart; branches glabrous or sparsely pubescent. Leaf lower surface hispid with simple hairs and dense subsessile glands ..... **5. gracilis**

iv) Section *Hypenia*

Flower not resupinate. Corolla tube 8 — 18 mm .....subsect. *Laxiflorae*

Flower resupinate. Corolla tube 15 — 26 mm .....subsect. *Ellipticae*

Subsection *Laxiflorae*

1 Corolla tube white, sometimes tinged violet or pink. Stem internodes with conspicuous globose swellings ..... **17. subrosea**

Corolla tube red, apricot, salmon-pink, pink, yellow or violet, never white. Stem internodes without conspicuous globose swellings (rarely present, in which case corolla tube red) 2

2 Calyx teeth aristulate, 3 — 7 mm; sparse setose hairs on upper parts of stem ..... **10. aristulata**

Calyx teeth broadly deltoid or narrowly deltoid-rostrate, < = 3 mm; setose hairs on upper parts of stem absent 3

3 Paired bracteoles deflexed, ovate. Leaf lamina oblong-elliptic ..... **15. recoria**

Paired bracteoles erect, lanceolate or narrowly lanceolate. Leaf lamina ovate or lanceolate 4

4 Inflorescence upper branches frequently aborted and forming an irregular umbellate shape, minute glandular hairs present ..... **11. caiaponiensis**

Inflorescence upper branches regularly developed (forming a regular pyramidal shape), minute glandular hairs absent 5

5 Leaf lamina narrowly lanceolate; margin coarsely serrate. Fruiting calyx narrowly cylindrical, >= 11 mm ..... **12. hatschbachii**

Leaf lamina ovate or lanceolate, not narrowly lanceolate; margin crenate or dentate. Fruiting calyx cylindrical, < =10 (rarely < 13) mm 6

6 Leaf lamina frequently lobed; margin coarsely dentate, sometimes irregular ..... **13. macrosiphon**

Leaf lamina never lobed; margin crenate or serrate, regular 7

7 Flowering calyx moderately pubescent with conspicuous and persistent long, contorted hairs; teeth narrowly deltoid ..... **14. paniculata**

Flowering calyx pubescent, conspicuous and persistent long, contorted hairs absent; teeth broadly deltoid ..... **16. reticulata**

Subsection *Ellipticae*

1 Paired bracteoles broadly ovate, overlapping to form a cup-shaped "epicalyx" ..... **18. calycina**

Paired bracteoles lanceolate to ovate, never overlapping to form a cup-shaped "epicalyx" **2**

2 Corolla tube 16 — 18 mm; yellow, apricot, pink or red. Cymes crowded ..3

Corolla tube 15 — 26 mm; red. Cymes not crowded **4**

3 Corolla yellow. Leaves shortly petiolate; base truncate or cordate, clasping stem ..... **19. crispata**

Corolla deep pink, apricot or red (yellow in bud). Leaves petiolate; base cuneate or truncate not clasping stem ..... **23. sclerophylla**

4 Stems upper parts pubescent; internodes not fistulose or swollen **22. niquelandiensis**

Stems upper parts glabrous; internodes fistulose, sometimes swollen 5

5 Leaf lamina ovate, base cordate..... **20. indaiaensis**

Leaf lamina lanceolate or oblong; base cuneate ..... **21. macrantha**

F. Species descriptions

i) Section *Vitifoliae* R. Atkinson sect. nov.

1. Eriope vitifolia *(Pohl ex Benth.) R. Atkinson* comb. nov.

Type: Brazil, Goyaz, Santa Cruz, May/June 1820, *Pohl* 6059 (holotype W).

Hyptis vitifolia Pohl ex Benth., Labiat. Gen. Spec.: 138 (1833); Bentham in DC. Prodr.12: 138 (1848); Schmidt in Fl. Bras. 8(1): 153 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 230 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 227 (1949).
Mesosphaerum vitifolium (Pohl ex Benth.) Kuntze, Revis. Gen. Pl. 2: 527 (1891).
Hypenia vitifolia (Pohl ex Benth.) Harley in Bot. J. Linn. Soc. 98: 91 (1988).
Hyptis calophylla St. Hil. ex Benth., Labiat. Gen. Spec.: 138 (1833); in DC. Prodr. 12: 137 (1848); Schmidt in Fl. Bras. 8(1): 153 (1858); Type: Brazil, Minas Geraes, Minas Novas, Morro d'Andaia, *St Hilaire* 1155 (P).
Mesosphaerum calophyllum (St. Hil.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hyptis glaziovii Briq. in Bull. Herb. Boissier 2: 716 (1894); Type: Brazil, São Paulo, Campos da Bocaina, S. José dos Barreiros, *Glaziou* 13047 (holotype G, isotypes K, P).

Woody herb to 2.5 m. *Stems* erect, often branched near base, moderately robust; basal parts pubescent with simple and glandular hairs and long setose hairs; upper parts glaucous green, drying reddish-brown, terete, glabrous, waxy; internodes fistulose with moderately to extremely inflated, easily compressible, sometimes asymmetric swellings. *Leaves* petiolate, spreading, distant; lamina ovate, sometimes lobed, 12 — 120 x 6 — 90 mm; base cordate; apex cuspidate to rostrate or obtuse; margin dentate, irregular; upper surface moderately pubescent with simple and glandular hairs; lower surface tomentose with lanate hairs; petiole 7 — 90 mm, glabrous or moderately pubescent with simple, glandular and occasional setose hairs. *Inflorescence* a large, open, indeterminate thyrse; phyllomes at lowest node 18 — 85 x 4 — 50 mm, caducous on other nodes; branches spreading, arched, 2 (sometimes 4) per node with cyme in axil, regularly developed, glabrous; cymes dichasial, many-flowered, regularly spaced 3 — 50 mm apart; peduncle bracts lanceolate, ca. 1 mm long; peduncle erect, 5 — 28 mm, glabrous; pedicel erect, 1 — 3 mm; paired bracteoles absent. *Flowering calyx* vinaceous, horizontal, campanulate, 2 — 3 mm, densely pubescent with simple hairs and some glandular hairs; teeth 0.3 — 1 mm, symmetrical, broadly deltoid. *Fruiting calyx*

drooping, cylindrical, swollen at base, 1 — 5 per cyme, 6 — 7 mm; teeth ca. 0.3 — 1 mm, apiculate; throat with sparse long hairs. *Corolla* tube white, tinged purple, brownish-yellow in bud, funnel-shaped, 3 — 4 mm; throat white; posterior lobes purple with dark lines, 1.5 to 2 mm, rounded, reclinate; lateral lobes purple, spreading; anterior lobe purple, ca. 3 mm. *Style* white, tinged purple at distal end. *Stamen* filaments white with white hairs; anthers yellow at anthesis becoming dark brown. *Nutlets* ovoid in section, 2.5 — 3.5 x 1.5 — 2.5 mm. Figure 10-1.

DISTRIBUTION. Brazil, Minas Gerais and Bahia in the Serra do Espinhaço with a disjunct occurrence near Niquelândia, Goiás (figure 10-6).

HABITAT. Upland savanna (campo rupestre) and savanna (cerrado). Altitude 650 — 1500 m.

BRAZIL.

BAHIA. Abaíra: Catolés, 5 Jan.1993, *Ganey* 1798 (K); Abaíra: Gerais do Pastinho, 31 Jan. 1992, *Hind* et al. 51419 (K); Abaíra: Campo da Pedra Grande, 19 Feb. 1992, *Stannard & Queiroz* 52115 (K); Água Quente: Pico das Almas, Vertente Norte, 16 Dec. 1988, *Hind* 27260 (SPF); Caetité: Brejinho das Ametistas district, ca. 3km from sede, 18 Feb. 1992 *Carvalho* et al. 3743 (MBM); Mucugê: 13.3 km N of Cascavel on road to Serra do Sincorá, , 25 March 1980, *Harley* et al. 20951 (K, NY); Morro do Chapéu: ca. 7 km S of town, 17 Feb. 1971, *Irwin* 32425 (K, NY, UC); Mucugê: 8 km SW on road from Cascavel, nr Fazenda Paraguacú, 6 Feb.1974, *Harley* et al. 16084 (K); Piatã: Quebrada da Serra do Atalho, 26 Dec. 1991, *Harley* et al. 50383 (K); Rio de Contas: 12-14 km N on road to Mato Grosso, 17 Jan. 1974, *Harley* et al. 15160 (K, NY, M, RB); Rio de Contas: 2.5-5 km S on side road to W of road to Mato Grosso, 28 March 1977, *Harley* et al. 20088 (E, K, NY, UEC); Rio de Contas: 6-10 km NW, on road to Pico das Almas, 21 July 1979, *Mori* et al. 12434 (K, NY); Seabra: ca. 26 km N, road to Agua de Rega, near Rio Riachão, 23 Feb. 1971, *Irwin* 30819 (K, NY, UC).

GOIÁS. Macêdo: ca.15km N of Niquelândia, 21 April 1988, *Brooks* et al. BRASPEX 142 (NY); Niquelândia: Jacuba, 24 Feb.1956, *Macêdo* 4408 (S);

MINAS GERAIS. Belo Horizonte: Sant. Hugo Werneck, 21 April 1956, *Roth* 16523a (SPF); Bocaiuva: 10 km N on BR 135, 21 Jan. 1978, *Hatschbach* 40804 (K); Diamantina: ca. 24 km SW on road to Gouveia, 20 Jan. 1969, *Irwin* et al. 22326 (K, NY); Francisco Sá: 50km NE on road to Salinas, 13 Feb. 1969, *Irwin* et al. 23260 (NY); Grão Mogol: road to Cristália, 22 April 1978, *Hatschbach* 41396 (K, UC); Januária: 28 Oct. 1964, *Heringer & Rizzini* 9882 (UB); Jequitaí: BR 365, near km 66, 13 March 1995, *Hatschbach* et al. 61819 (MBM); Joaquim Felício: ca. 3 km S, 6 March 1970, *Irwin* 27062 (K, NY); Lagoa Santa: *Glaziou* 989 (P); near Formiga (= Montes Claros): July 1840, *Gardner* 5081 (K, BM); Montes Claros: ca. 30 km SE, road to Juramento, 15 May 1977, *Gibbs* et al. 5120 (UEC); Montes Claros: ca. 48 km W on road to Aguas Claras, Serra do Espinhaço, 25 Feb. 1969, *Irwin* 23880 (UB); Montezuma: ca. 30km NW of the town on way to Espinosa, Minas Gerais, 14 March 1994, *Souza* et al. 5503 (K, SPF); Morro d'Andaia: dans les Minas Novas, Minas Geraes, 1816-1821, *St.Hilaire* 1133 (P); Santo Hipólito - Diamantina road: km 81, 30 Nov. 1976, *Shepherd* et al. 3851(or 9851?) (NY, UEC); Santana do Riacho: 28 km S José de Almeida on road to Santana, 16 Feb. 1982, *Giulietti* et al. CFSC 7779 (K); Varzea da Palma: Fazenda Mãe d'Agua, 22 Nov. 1962, *Duarte* 7491 (RB).

RIO DE JANEIRO. São José dos Barreiros: Campo da Bocaina, 6 April 1892, *Glaziou* 13047* (K, G, P).
* Glaziou specimens are notorious for unreliable localities (see Wurdack 1970) and because this locality lies well outside the usual range of *E. vitifolia*.it is unlikely to represent a collection actually made from the state of Rio de Janeiro.

E. vitifolia is isolated in *Eriope* and can be readily identified. Uniquely in the genus, both *E. vitifolia* and *E. longicaulis* have many-flowered cymes, the individual flowers of which lack paired bracteoles below the calyx. The leaves are variable in size and shape but the conspicuously swollen stems, with often assymetric fistulae, make *E. vitifolia* one of the most vegetatively distinct species in the Hyptidinae.

Hyptis calophyllum and *Hyptis glaziovii* have both been placed as synonyms of *E. vitifolia* by Epling (1936). They were described on the basis of their relatively large leaves but further collections have confirmed the decision to place them in synonymy. Collections of *E. vitifolia* from Minas Gerais tend to have larger leaves but the variation overlaps with that found in other localities.

E. vitifolia is distributed through the mountains of the Serra do Espinhaço from central Minas Gerais through to northern Bahia with a disjunction to Niquelândia in the Serra da Mesa in northwestern Goiás.

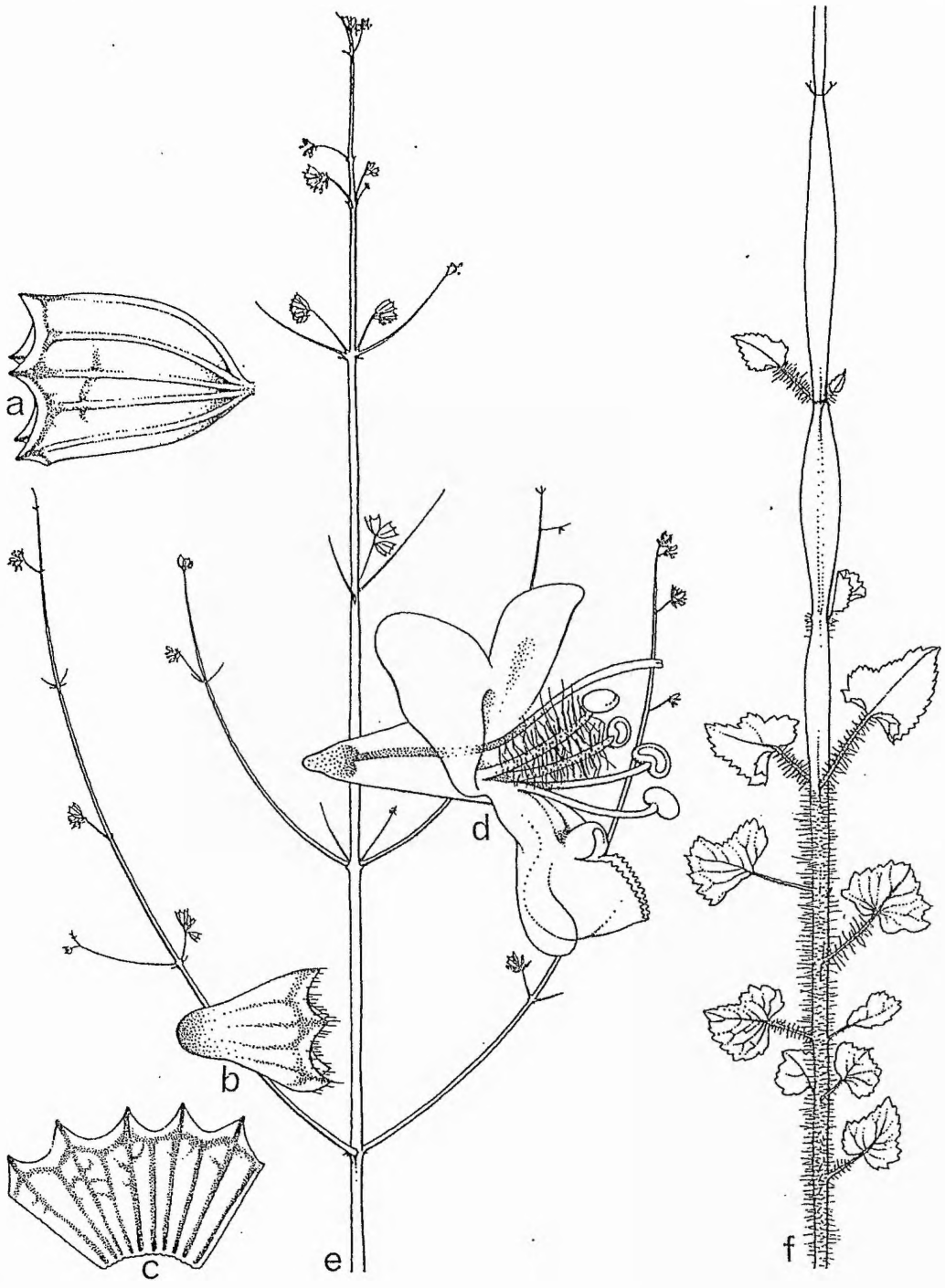


Figure 10-1: ***Eriope vitifolia***

a) fruiting calyx (x8); b) flowering calyx (x8); c) flowering calyx (x8); d) corolla (x8); e) inflorescence (x½); f) stem and leaves (x½) (a - f *Irwin* et al 32425). Drawn by Emmanuel Papodopoulos.

2. Eriope longicaulis *Harley* & R. Atkinson* sp. nov.

Type: Brazil, Bahia, 22 km NW of Lagoinha, road to Minas do Mimoso, 6 March 1974, *Harley* et al.16873 (holotype SPF, isotype K).

E. vitifoliae affinis sed internodiis caulium non tumidis e statura majore distinguitur.

Shrub to 4 m. *Stems* erect, frequently branched, moderately robust; basal parts pubescent with simple and glandular hairs and long setose hairs; upper parts glaucous green, drying reddish-brown, terete, glabrous, waxy; internodes fistulose, swollen along their length. *Leaves* petiolate, spreading, distant; lamina ovate, sometimes lobed, 12 — 35 x 5 — 20 mm; base cordate; apex cuspidate to rostrate or obtuse; margin dentate, irregular; upper surface moderately pubescent with simple and glandular hairs; lower surface tomentose with lanate hairs; petiole 4 — 25 mm, glabrous or moderately pubescent with simple, glandular and occasional setose hairs. *Inflorescence* a large, open, indeterminate thyrse; phyllomes caducous; branches spreading, arched, 2 (sometimes 4) per node with cyme in axil, regularly developed, glabrous; cymes dichasial, many-flowered, regularly spaced 3 — 50 mm apart; peduncle bracts lanceolate, ca. 1 mm long; peduncle erect, 2 — 4 mm, glabrous; pedicel erect, 1 — 3 mm; paired bracteoles absent. *Flowering calyx* vinaceus, horizontal, campanulate, 2 — 3 mm, densely pubescent with simple hairs and some glandular hairs; teeth 0.3 — 1 mm, symmetrical, broadly deltoid. *Fruiting calyx* drooping, cylindrical, swollen at base, 1 — 5 per cyme, 6 — 7 mm; teeth ca. 0.3 — 1 mm, apiculate; throat with sparse long hairs. *Corolla* tube white, tinged purple, brownish-yellow in bud, funnel-shaped, 3 — 4 mm; throat white; posterior lobes purple with dark lines, 1.5 to 2 mm, rounded, reclinate; lateral lobes purple, spreading; anterior lobe purple, ca. 3 mm. *Style* white, tinged purple at distal end. *Stamen* filaments white with white hairs; anthers yellow at anthesis becoming dark brown. *Nutlets* ovoid in section, 2.5 — 3.5 x 1.5 — 2.5 mm.

DISTRIBUTION. Brazil, Bahia, endemic to a small area in northern Bahia (figure 10-6).
HABITAT. Savanna (cerrado). Altitude: 950 — 1000 m.

BRAZIL.

BAHIA. Lagoinha: 16 km NW (5.5km SW of Delfino), side road to Minas do Mimoso, 4 March 1974, *Harley* et al.16690 (K, R); Lagoinha: 22 km NW, road to Minas do Mimoso, 6 March 1974, *Harley* et al.16873 (K); Lagoinha: Delfino area, 9 March 1997, *Harley* et al. PCD 6177 (K).

* This species was first recognised in the field by Harley.

The two species of section *Vitifoliae* differ principally in the length and shape of the internodes. In *E. longicaulis* the internodes are swollen along their length rather than developing the conspicuous fistulae of *E. vitifolia.* This has an impact on the habit of the taxa and *E. longicaulis* is much taller and less compact than *E. vitifolia.* There is a significant cytological difference between the two species (see chapter 3), *E. longicaulis* has 2n = 22, an anomalous number in the Hyptidinae and *E. vitifolia* has 2n = 28, a common number in *Hyptis.* This difference supports taxonomic recognition of the two forms although the morphology of the two taxa is very similar. *E. longicaulis* may represent an isolated population of *E. vitifolia* which maintains reproductive isolation through cytological differentiation. Alternatively the cytological difference may reflect a greater divergence between the species than that indicated by morphology. In the absence of further information it seems most sensible to emphasise the distinctiveness of *E. longicaulis* by recognising it as a species.

E. longicaulis occurs at the northern extremity of the Serra do Espinhaço in northwestern Bahia and is separated from the nearest *E. vitifolia* locality at Morro do Chapéu by 125 km (see figure 10-6).

ii) Section *Micranthae* R. Atkinson sect. nov.

3. Eriope micrantha *Benth.in DC.*

Type: Brazil, Chapada, Mato Grosso, *Reidel* 87 (holotype K). Benth. in DC., Prodr. 12: 141 (1848).

Hypenia micrantha (Benth. in DC.) Harley in Bot. J. Linn. Soc. 98: 92 (1988).
Hyptis effusa S.Moore in Trans. Linn. Soc. London, Bot. Ser. 2, 4: 441 (1895); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 230 (1936); Epling in Revista Mus. La Plata, Secc. Bot. 7: 229 (1949). Type: Brazil, Chapada, Mato Grosso, *Moore* 159 (holotype BM).

Woody herb to 2 m. *Stems* erect, unbranched with long, virgate axes, slender; basal parts pubescent with dense to moderate branched, glandular and simple hairs; upper parts reddish to dark brown, pubescent with moderately dense branched, glandular and simple hairs, angled, wax absent; internodes not fistulose or swollen. *Leaves* petiolate, spreading, distant; lamina ovate, 25 — 90 mm x 15 — 55 mm; apex acute; base cordate to truncate; margin serrate, regular; upper surface sparsely pubescent with branched, simple and glandular hairs; lower surface moderately to densely pubescent with long hairs and subsessile glands; petiole 5 — 15 mm with branched and simple hairs. *Inflorescence* a large open, indeterminate thyrse; phyllomes ovate, 4 — 15 x 2 — 8 mm, persistent; branches spreading, straight, 2 per node, very regularly developed, glabrous or with sparse short hairs; cymes 1-flowered evenly spaced 5 — 12 mm apart; pedicel drooping, 8 — 20 mm, glabrous; paired bracteoles narrowly lanceolate, ca. 0.5 mm long. *Flowering calyx* vinaceous, deflexed, 1.5 — 2.5 mm, campanulate, pubescent with simple and glandular hairs; teeth ca. 0.2 mm, symmetrical, broadly deltoid. *Fruiting calyx* declinate, 4 — 5 mm, broadly campanulate; teeth ca. 0.5 mm, broadly deltoid ; throat glabrous. *Corolla* tube purple, funnel-shaped, ca. 3.5 mm long; posterior lobes with dark lines, 2.5 to 3 mm, rounded; lateral lobes 2.5 — 3 mm; anterior lobe purple, ca. 2 mm long. *Stamen* filaments with long hairs. *Nutlets* not seen.

DISTRIBUTION.Brazil, Mato Grosso, endemic to Chapada dos Guimarães (figure 10-6).
HABITAT. Savanna (cerrado). Altitude: 600 — 680 m.

BRAZIL.

MATO GROSSO. Chapada dos Guimarães: Colégio Evangélico de Burití, 7 May 1983, *Barcia* et al. 1319 (R); Chapada, *Riedel* 87 May/June 1827 (K); Cuiabá: NE of the town near Burití, June 1927, *Dorrien Smith* 267 (K); s.loc., *Gaudichaud* 64, 1833 (P); Cuiabá: Santa Anna da Chapada, 12 May 1903 *Malme* (S); Cuiabá: Burití by Santa Anna da Chapada, 26 June 1894 *Malme* 1704C (S); Cuiabá: Santa Anna da Chapada, 30 July 1902, *Malme* s.n. (S); Cuibá: Santa Anna da Chapada, 27 May 1903 *Malme* s.n. (S); Plateau of Chapada, Sept. 1891, *Moore* 159 (BM).

E. micrantha is a very distinct species in the genus and is easily recognised by the exceptionally regular panicle with single-flowered cymes and the large regular, ovate leaves and phyllomes which are unique in *Hypenia*. It was the only species of *Hypenia* to have branched hairs on the leaves but there are a number of *Eriope* section *Eriope* species which have branched hairs, e.g. *E. latifolia*. The flowers of *E. micrantha* are small and campanulate and very similar to those found in sections *Vitifoliae* and *Eriope*. The inflorescence structure

of *E. micrantha* is typical of *Eriope* section *Hypenia* subsection *Laxiflorae,* differing only in its exceptional regularity, long peduncles and the broad, leaf-like phyllomes.

E. micrantha is morphologically and geographically isolated and is mostly restricted to a very small area of cerrado in the Chapada dos Guimarães in central Mato Grosso (figure 10-6).

iii) Section *Irregulares* (Briq.) R. Atkinson comb. & stat. nov.

4. Eriope irregularis *(Benth. in DC.) R. Atkinson* comb. nov.

Type: Brazil. Bahia, Santa Rosa, *Gardner* 2926, Sept. 1839 (holotype K, isotypes BM, G).
Hyptis irregularis Benth. in DC. Prodr.12: 137 (1848); Schmidt in Fl. Bras. 8: 152 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 226 (1936); Epling in Revista Mus. La Plata, Secc. Bot. 7: 224 (1949).
Mesosphaerum irregulare (Benth.) Kuntze, Revis. Gen.Pl. 2: 526 (1891).
Hypenia irregularis (Benth. in DC.) Harley in Bot. J. Linn. Soc. 98: 91 (1988).

Woody herb or shrub to 2.5 m. *Stems* erect, much-branched, slender; basal parts pubescent with dense simple hairs and setose hairs; upper parts drying dark brown, moderately pubescent with dense simple hairs, slightly angled, wax absent; internodes not fistulose or swollen. *Leaves* sessile, erect, persistent, overlapping, sometimes small leaves clustered in axils; lamina ovate to broadly deltoid, 8 — 25 x 5 — 11 mm; base cordate or truncate; apex acute or obtuse; margin crenate, regular; upper surface sparsely to moderately pubescent with simple hairs; lower surface sparsely to moderately pubescent with simple hairs and subsessile glands. *Inflorescence* a little-branched, open, indeterminate thyrse; phyllomes caducous; branches erect, straight, 2 per node, regularly developed, moderately pubescent with simple hairs; cymes 1-flowered, clustered or spaced 1.5 — 30 mm apart; peduncle bracts lanceolate, 1 — 1.5 mm; peduncle erect, 1 — 5 mm, pubescent with simple hairs; paired bracteoles erect, narrowly lanceolate, 0.5 — 1 mm. *Flowering calyx* erect, straight, cylindrical, 4 — 5 mm, pubescent with simple hairs and subsessile glands; teeth symmetric, 1 — 1.5 mm, broadly deltoid. *Fruiting calyx* erect, straight, 6.5 — 9 mm; teeth 1.5 — 2 mm, broadly deltoid; throat glabrous. *Corolla* tube white or violet, ca. 5 mm, cylindrical; posterior and lateral lobes white or violet, ca. 1.5 mm; anterior lobe white or violet, ca. 1 mm. *Stamen* filaments with sparse long hairs. *Nutlets* trigonous, ca. 2 x 1 mm.

DISTRIBUTION. Brazil, north-eastern Goiás and neighbouring parts of Bahia and Minas Gerais and an outlying record in Maranhão (figure 10-7).

HABITAT. Savanna (cerrado). Altitude: 600 — 950 m.

BRAZIL.

BAHIA Barreiras: ca. 32 km W, upper slopes of Espigão do Mestre, 5 March 1971, *Irwin* et al. 31565 (K, P, NY, UC); Corrego do Brejo: Goyaz, 7 May 1895, *Glaziou* 21920 (K, G, P); Correntina: ca. 15 km E on road to Goiás, Chapadão Occidental, 25 April 1980, *Harley* et al. 21720 (K, NY, SPF); Correntina: Fazenda Jatobá, 26 June 1992, *Silva* et al. 1375 (K); Correntina: Fazenda Jatobá, 5 June 1992, *Silva* et al. 1270 (K); Cristópolis: Engo. Velho, 3 July 1979, *Hatschbach* 42317 (K, NY, UC); Dianópolis - Barreiras road, ca. 41 km after GO-BA boundary, 12 Feb. 1987, *Pirani* et al. 1972 (K); Rio Roda Velha: ca. 150 km SW of Barreiras, 15 April 1966, *Irwin* et al. 14932 (K, NY);. Santa Rosa: Sept. 1839, *Gardner* 2925 (K, BM); Santa Rosa, Sept. 1839, *Gardner* 2926 (K, BM, G);

GOIÁS. Posse: 10 km S on BR-020, 8 Oct. 1976, *Hatschbach* 39067 (K); Posse: 15 km N on BR-020, 12 July 1979, *Hatschbach & Guimarães* 42276 (K, NY, UC, MBM).

MARANHÃO. Carolina: 35 km from town on BR-010, Transamazônica, Pedra Caida, 15 April 1983, *Silva* et al. 1117 (K).

E. irregularis is superficially similar to *E. brachystachys* but is distinguished by the symmetric calyx. *E. brachystachys* always has the upper three teeth connate at the base. *E. irregularis* also differs from *E. brachystachys* in the inflorescence structure. Although the cymes frequently overlap they are not clustered to the extent that they are in *E. brachystachys* and *E. densiflora* and they are always single-flowered. The leaf is also a different shape and is rhomboid rather than clearly ovate in *E. brachystachys.* The specimens cited above have short, pubescent inflorescence branches and small, sessile leaves and are readily placed into *E. irregularis.* They are all collected from an area to the north and east of the centre of distribution of *E. brachystachys* and can clearly be identified as distinct from it (figure 10-7).

5. Eriope gracilis *R. Atkinson* sp nov.

Type: Brazil, Mato Grosso, Corrego Rancho (mun. Alto Araguaia), *Hatschbach* 34695 (holotype MBM, isotype K).

E. irregulari affinis sed inflorescentiis ramosis, cymis distantibus et foliis magis glandulosis differt.

Woody herb or shrub to 1.5 m. *Stems* erect, unbranched, slender; basal parts pubescent with moderately dense simple hairs, glandular hairs and setose hairs; upper parts drying dark brown, glabrous or sparsely pubescent with simple hairs and glandular hairs, slightly angled, wax absent; internodes not fistulose or swollen. *Leaves* petiolate, spreading, persistent, distant; lamina ovoid to broadly deltoid , 17 — 35 x 11 — 18 mm; base cordate or truncate; apex acute; margin crenate, regular; upper surface hispid with simple hairs; lower surface hispid with simple hairs and dense subsessile glands. *Inflorescence* a much branched, open, indeterminate thyrse; phyllomes caducous; branches spreading, straight, 2 or 4 per node, regularly developed, glabrous or sparsely pubescent with simple hairs; cymes 1-flowered, widely spaced 3 — 30 mm apart; peduncle bracts lanceolate, ca. 1 mm; peduncle erect 2 — 10 mm, glabrous; paired bracteoles erect, narrowly lanceolate, 0.5 — 1 mm. *Flowering calyx* erect, straight, cylindrical, ca. 4 mm, pubescent with simple hairs and subsessile glands; teeth symmetric, ca. 1 mm, broadly deltoid . *Fruiting calyx* erect, straight, 8 — 9 mm; teeth ca. 1.5 mm, broadly deltoid ; throat glabrous. *Corolla* tube white or violet, ca. 4 mm, cylindrical; posterior and lateral lobes white or violet, ca. 1.5 mm; anterior lobe white or violet, ca. 1 mm. *Stamen* filaments with sparse long hairs. *Nutlets* trigonous, ca. 2 x 1 mm.

DISTRIBUTION. Brazil, west-central region of the Planalto Central. Northern Goiás and eastern Mato Grosso (figure 10-7).
HABITAT. Savanna (cerrado). Altitude not recorded.

BRAZIL.
MATO GROSSO. Alto Araguaia: Corrego Rancho, 22 July 1974, *Hatschbach* 34695 (MBM, K).
GOIÁS. Colinas do Sul: Serra da Mesa, 15 June 1993, *Hatschbach* 59601 (MBM).

E. gracilis can be identified by its relatively large, distantly spaced leaves with hispid indumentum and dense subsessile glands on the lower leaf surface. The inflorescence branches are more or less glabrous and much longer than those of *E. irregularis* and the cymes are distantly spaced along their length. The two collections differ in morphology. *Hatschbach 59601* has much longer and more slender inflorescence branches but the leaves are a similar size and shape to *Hatschbach 34695* although they are distinctly petiolate. The inflorescence has a markedly similar structure to that of species from subsection *Laxiflorae*.

This species is close to *E. irregularis* and shares the same calyx and corolla characters. *Hatschbach 34695* is intermediate in morphology, although not in geographical location,

between *Hatschbach 59601* and *E. irregularis*. However the divergent inflorescence structure is very marked in both collections and makes the species clearly recognisable.

E. gracilis occurs further west than *E. irregularis* and the two collections are somewhat isolated from each other. Further collections from intermediate localities would be very useful to increase the understanding of the range of variation in this taxon.

iv) Section *Salzmanniae* R. Atkinson sect. nov.

6. Eriope salzmannii *(Benth.) R. Atkinson* comb. nov.

Type: Brazil, Bahia, *Salzmann* s.n. (holotype K, isotypes G, P).

Hyptis salzmannii Benth., Labiat.Gen. Spec.: 138 (1833); Benth. in DC. Prodr. 12: 137 (1848); Schmidt in Fl. Bras. 8(1): 152 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 230 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 230 (1949).
Mesosphaerum salzmannii (Benth.) Kuntze, Revis. Gen. Pl. 2: 527 (1891).
Hypenia salzmannii (Benth.) Harley in Bot. J. Linn. Soc. 98: 91 (1988).
Hyptis salzmannii var. *filipes* St. Hil. ex Benth. in DC. Prodr. 12: 137 (1848); Schmidt in Fl. Bras. 8(1): 152 (1858). Type: Brazil, Piauhy, Oeiras, *Gardner* 2283 (holotype K, isotypes BM, G, P).

Woody herb to 2m. *Stems* erect, branched near base below long, virgate axes, slender; basal parts pubescent with short simple and glandular hairs and long setose hairs; upper parts glaucous green, drying reddish-brown, terete, glabrous, waxy; internodes fistulose with slightly to moderately inflated, hard, swellings. *Leaves* petiolate, spreading, smaller leaves clustered in axils, mostly overlapping; lamina ovate to lanceolate, 5 — 50 x 3 — 22 mm; base cuneate; apex acute or obtuse; margin +/- regular, crenate; upper surface sparsely to moderately pubescent with short simple hairs and glandular hairs; lower surface moderately to densely pubescent with subsessile glands and short simple hairs and glandular hairs; petiole 2 — 40 mm, glabrous or with short simple hairs and glandular hairs. *Inflorescence* a large, open, indeterminate thyrse; phyllomes lanceolate, 6 — 8 x 1.5 — 3 mm, frequently caducous; branches spreading, straight, 2 per node, sometimes with cymes in axils, regularly developed, glabrous; cymes dichasial, 1 —3-flowered, evenly spaced 3 — 40 mm apart, often subtended by leaves on lower parts of plant; peduncle bracts lanceolate, ca. 1 mm;

peduncle erect, 0 — 35 mm, glabrous; paired bracteoles ca. 0.5 mm (in 3-flowered cymes middle flower lacks bracteoles). *Flowering calyx* green or vinaceous, horizontal, campanulate, 2 — 3 mm, moderately to densely pubescent with short simple hairs and glandular hairs; teeth ca. 1mm, symmetrical, broadly deltoid. *Fruiting calyx* erect, broadly campanulate, 4 — 8 mm, chartaceous; teeth 1 — 2 mm, broadly deltoid ; throat with sparse to moderate long hairs. *Corolla* tube pale yellow in bud maturing to sky blue, cylindrical, 4 — 5 mm; throat white; anterior lobes blue with dark lines into throat, vertical; lateral lobes blue, 2 — 3 mm, rounded, held forward of corolla mouth; lower lip blue, ca. 1 mm. *Style* white. *Stamen* filaments white, with white hairs; anthers yellow. *Nutlets* trigonous, 2.5 — 3.5 mm x 1 — 1.5 mm.

Figure 10-2.

DISTRIBUTION. *E. salzmannii* is the most widespread species in section *Hypenia* and occurs in the drier parts of Northeastern Brazil with a disjunction to the llanos of Venezuela and one isolated record in the Pacaraima mountains in Guyana. There is one unlocalised record for Colombia, the other, for Angostura, is mapped in Venezuela, Angostura is the old name for Ciudad Bolivar in Venezuela (figure 10-8).

HABITAT. Seasonally dry woodland (caatinga), savanna (cerrado), upland savanna (campo rupestre), coastal scrub (restinga) and disturbed areas. Altitude: 50 — 1180 m.

BRAZIL.

BAHIA. S. loc. , 1834, *Blanchet* 111 (G); s. loc., s.d. *Blanchet* 167 (K, BM, G, W); s.loc., 1839, *Gardner* 715 (K); s. loc., s.d., *Martius* 641 (K, BM); s. loc., s. dat. *Salzmann* s.n. (K, G, P); Abaíra: Cabrália, Piatã - Boninal road, 19 March 1992, *Ganev & Queiroz* 52715 (K); Água Fria: road to Cia.de Celulose, 20 Aug. 1984, *Lima & Messias Santos* 186 (NY); Aracatú: ca.20 km along Brumado - Vitoria da Conquista road, 29 Dec. 1989, *Carvalho* et al. 2701 (K, MBM); Aracatú: 14 May 1983, *Hatschbach* 46374 (K, MBM); Barreiras: Rio das Ondas, 12 March 1979, *Hatschbach* 42126 (K, NY, UC); Barreiras: ca. 10 km W, 2 March 1971, *Irwin* et al. 31298 (K, NY, UC); Cactité: 20 km E, road to Brumado, 20 Nov. 1992, *Arbo* et al. 5661 (SPF); Caetité: Brumado, road to Caetité, 16 June 1986, *Hatschbach & Silva* 50437 (K, MBM); Caetité: Tucano, 15 March 1995, *Hatschbach* et al. 61915 (MBM); Camaçari: BA-099 (cocoa road), Guarajuba, 14 July 1983, *Bautista* et al. 823 (K, NY); Canudos: ca. 10 km S, 10 July 1985, *Gonzaga* 12 (RB); Canudos: Raso da Catarina, 18 June 1981, *Silva Guedes* s.n. (RB); Delfino: 3km NW of Lagoinha (5.5km SW of Delfino), road to Minas do Mimoso, 5 March 1974, *Harley* et al. 16740 (K, E, NY, M); Feira de Santana: 8 Oct. 1982, *Noblick & Britto* 2069 (K); Feira de Santana: Campus de UEFS, 7 Aug. 1985, *Noblick* 4249 (K, NY); Feira de Santana: Rod. BR28, 5 Oct. 1963, *Silva Santos* 28005 (K, M); Gentio do Ouro: ca.4km NE , road to Central, 22 Feb. 1977, *Harley* et al. 18930 (K, E, NY, UEC, SPF); Jacobina: ca. 10 km away on road to Morro do Chapéu, 14 March 1990, *Carvalho & Saunders* 2800 (K); Jacobina: Lage do Batata road, km 15, 28 June 1983, *Coradin* et al. 6169 (K, NY, SP); Jacobina: Serra da Jacobina, Morro da Pousada, 24 Dec. 1984, *Harley* et al. CFCR 7559 (K, UEC, SPF); Jacobina: Serra do

Tombador, 26 Oct. 1978, *Martinelli* 5180 (RB); Juázeiro; 53 km NE on BA210, 9 Feb.1972, *Pickersgill* RU 72-97 (K); Lençois: BR 242 between junction to Lençois & Pai Inácio, 19 Dec. 1984, *Harley* et al. CFCR 7121 (K, SPF); Juázeiro: 7 km S on BR 407 to Senhor do Bonfim, grounds of Pousada, 24 Jan. 1993, *Thomas* et al. 9624 (MBM); Livramento do Brumado: ca.4 km along road from to Rio de Contas, 28 March 1991, *Lewis & Andrade*, 1924 (K, UEC, SPF); Maracás: 26 km away on way to Tamboré, 24 Jan. 1965, *Pereira & Pabst* 9709 (K, M); Milagres: road to Itaberaba, km5, BR 116, 13 Dec. 1981, *Carvalho & Lewis* 965 (K); Milagres: BA 046, 18 July 1982, *Hatschbach* 45129 (MBM); Mimoso: between Rio Roda Velha & Rio de Pedras, road between Posse, GO & Barreiras, BA, 13 Feb. 1971, *Irwin* et al. 33270 (K, NY); Morro do Chapéu: 22 km W, 20 Feb. 1971, *Irwin* et al. 30682 (K, NY, UC); Morro do Chapéu: 26 July 1980, *Orlandi* 277 (RB); Pedra Azul: on way to airport, 20 April 1964, *Trinta & Fromm,* 802 (NY, R); Piatã: Gerais da Inúbia, 22-26 km from Catolés, 10 March 1992, *Stannard* et al. 51847 (K); Planalto: 9km NE along BR 116, 30 March 1976, *Davidse* et al.11641 (K); Ponco d'Arcias: June 1844, *Blanchet* 3882 (G, P); Rio de Contas: Estrada de Fraga, ca. 2km SE of town, 13 July 1985, *Graças* et al. 870 (K); Rio de Contas: ca.2km N, flood plain of Rio Brumado, 19 Jan. 1974, *Harley* et al. 15313 (K, E, NY, M); Rio de Contas: ca. 2 km N, flood plain Rio Brumado, 22 March 1977, *Harley* et al. 19841 (K); Rio de Contas: 5km S on road to Livramento do Brumado, 16 April 1991, *Lewis & Andrade* 1988 (K, UEC); Rio de Contas: Barrigudinha, *Martius* 1888 (M); Seabra: Queimada Nova, 13 Jan 1977, *Hatschbach* 39535 (K, UC, MBM); Serra de São Inácio, Feb. 1907, *Ule* 7553 (K, G); Sobradinho: km 48 on road to Juazeiro, 8 Aug. 1994, *da Silva* et al. 2442 (K, CEN); Tangine Novo: janzeiro, Barrigudinho, 1914, *Luetzelburg* 8 (K, M); Xique Xique: ca.4km N of Sao Inácio on road to Xique Xique, 25 Feb. 1977, *Harley* et al. 19070 (K, NY, UEC, SPF).

CEARÁ. s.col., s.d. 1131/8256 (R); s.col., s.d., 1139/8253 (R); Barbalha: between Barbalha and Curiri-Crato, 6 Aug. 1948, *Duarte* 1280 (RB); Cedro: 31 May 1933, *Luetzelberg* 23640 (M); Missão Velha: 23 July 1964, *Duarte & Castellanos* 485 (K); Orós: Orós to Icó road (10 km from Orós), 14 July 1984, *Silva* 228 (RB); Riacho do Porco: 25 April 1910, *Löfgren* 684 (S).

GOIÁS. Posse: between Rio Roda Velha and Rio de Pedras, road from Posse, 13 Feb. 1971, *Irwin* et al. 30239 (UC).

MINAS GERAIS. S. loc., 1878-9, *Glaziou* 11298 (K,P); Diamantina: Pinheiro, 1892, *Glaziou* 19686 (K, R); Grão Mogol: 1 km SW of the city, 21 May 1982, *Mamede* et al. CFCR 3405 (K, MBM); Itaobim: km 4 on way to Jequitinhonha, 9 Feb. 1977, *Shepherd* et al. 4407 (UEC).

PARAÍBA. Campina Grande: 16 km W, 27 July 1990, *Agra* 1270 (K); Souza: 1935, *Seccas* 50 (UC, RB).

PERNAMBUCO. S. loc., 1912, *Luetzelberg* 1477 (RB); Gravatá: July 1926, *Pickel* 1140 (K); Recife: Caruaru, W of Recife, 25 Sept. 1976, *Davis & Andrade-Lima* 61134 (K, E, UEC); Santa Maria de Boa Vista: 24.7 km NNE of Lagoa Grande, 7 March 1970, *Eiten & Eiten* 10865 (K, SP).

PIAUÍ. S. loc. *Gardner* s.n. (K); s. loc. April 1839, *Gardner* 2283 (K, BM, G, P); s. loc. 1912, *Luetzelberg* s.n. (RB); Serra de Santa Marita, 8 April 1978, *Orlandi* 32 (RB).

LOC. INCERT. Serra de Tiriria, 11 May 1912, *Zehintura* s.n. (R).

SERGIPE. Estância: ca. 19.4 km on BR 101 to Praia do Abais, 28 Nov. 1993, *Amrorim* et al. 1538 (K).

GUYANA. Pacaraima, s.d., *Schomburgk* 177 (K).

COLOMBIA. S. loc., 1865, *Moritz* 419 (G).

VENEZUELA. Bords de l'Orénoque, *Chaffanjon* s.n. (P); Caicara del Orinoco: 22.5 km SW, Cerro Medano, 2 Sept. 1985, *Steyermark* et al. 131210 (NY); Ciudad Bolívar: Bolívar, Sept.1929, *Holt* 148 (UC); Ciudad Bolívar: Nov. 1898, *Sprague* s.n. (K); Cumaná: Cerro Impossível, Sucre Prov., 1843, *Funck* 715 (G); El Furrial: 6 km E, 35 km E Maturín, Monagas, 19 April 1973, *Agostini & Agostini* 1692 (K); El Tigre: 33km S, Anzoategui, 6 April 1985, *Manara* et al. 2279 (NY); Piar: Hato Morichito, May 1986, *Fernandez* 2853 (K); Angostura, 1841, *Otto,*

s.n. (UC); Santa Fé de Guaicú: entre rio Cani y Cantaura, Anzoátegui, 22 Aug. 1942, *Pittier* 15105 (UC); Maturin: 10 km S, between Maturin and Barrancas, 26 March 1970, *Rojas* 791 (K); Urica: 11.5 km SE on highway 13, Anzoategui, 3 Oct. 1977, *Steyermark* et al. 114275 (K).

E. salzmannii is morphologically isolated in *Eriope,* although it has a superficial resemblance to *E. vitifolia* and some species of section *Eriope,* notably *E. hypenioides.* All three species have well-developed 'greasy pole' characters and small flowers borne in a large, lax inflorescence but *E. salzmannii* is readily identified by its blue, tubular corollas, one (sometimes two or three) - flowered cymes, its small, ovate leaves and hard, globose swellings of the stem. It does not differ significantly in its inflorescence morphology from section *Hypenia* subsection *Laxiflorae,* or section *Eriope,* but the floral morphology is divergent. The flowers of *E.salzmannii* are small and blue with a cylindrical corolla tube and the lateral lobes point forward. This floral morphology is also seen in *Eriopidion strictum.*

A variety of *E. salzmannii,* var. *filipes,* was described by Bentham (1848) on the basis of one specimen which is scarcely distinguishable from other specimens of *E. salzmannii* and the variety was reduced to synonymy by Epling (1936).

E. salzmannii is unusual in *Eriope* for its distribution. It is common on roadsides and other disturbed areas and it is the most ecologically catholic species in the genus, occurring in cerrado, campo rupestre, restinga and caatinga in Northeastern Brazil and the llanos of northern South America (figure 10-8).

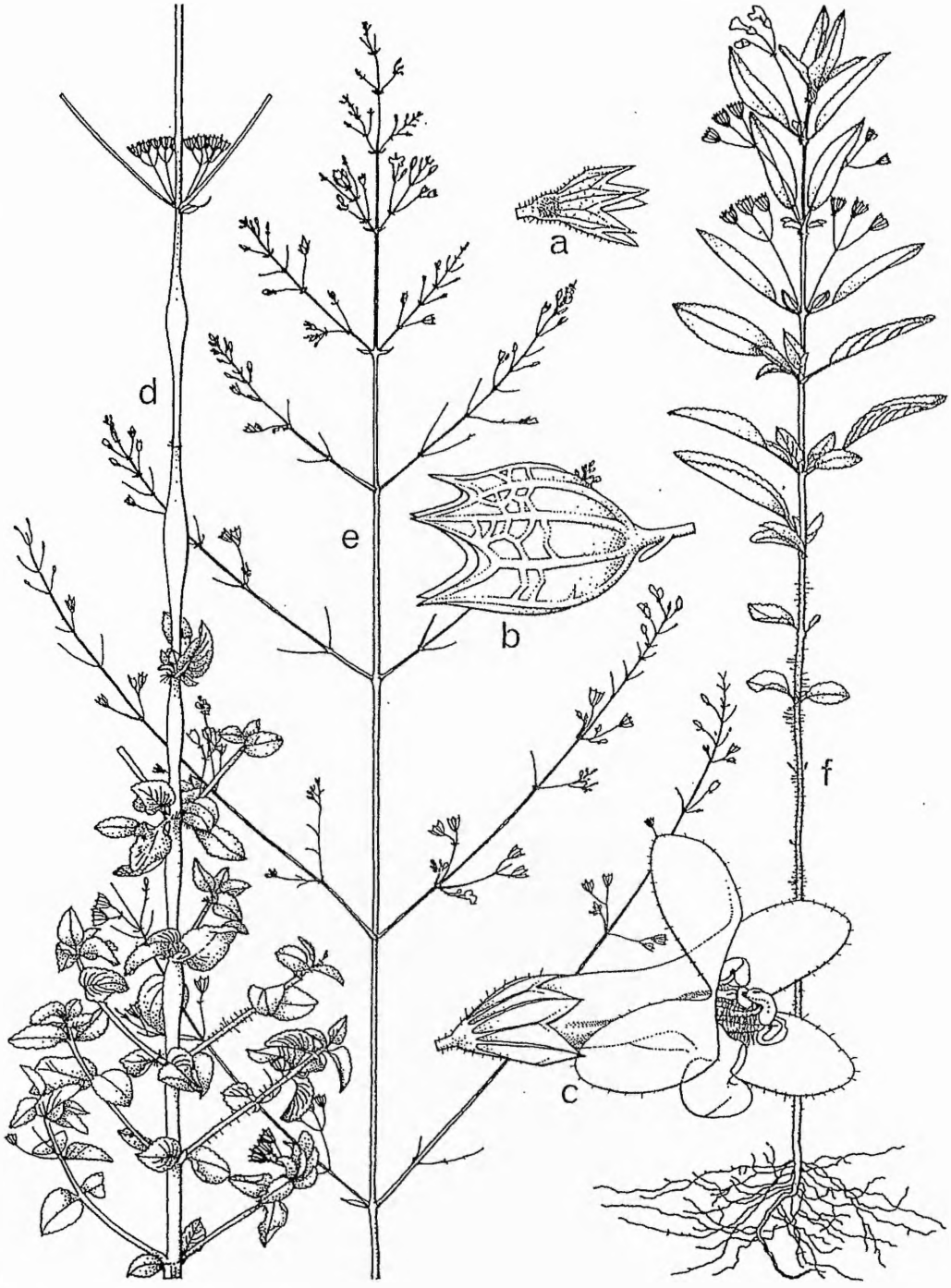


Figure 10-2: ***Eriope salzmannii***

a) flowering calyx (x5) (*Coradin* et al. 6169); b) fruiting calyx (x5) (*Coradin* et al. 6169); c) flower (x6) (*Coradin* et al. 6169); d) stem and leaves (x½) (*Coradin* et al. 6169); e) inflorescence (x½) (*Coradin* et al. 6169); immature plant with axillary cymes (x½) (*Irwin* et al. 31298). Drawn by Emmanuel Papodopoulos.

v) Section *Densiflorae* (Benth. in DC.) R. Atkinson comb. & stat. nov.

7. Eriope brachystachys *(Pohl ex Benth.) R. Atkinson* comb. nov.

Type: Brazil, entre Rio Crixas e Rio Maranhão, Goyaz,1819, *Pohl* s.n. (holotype W, isotype K).

Hyptis brachystachys Pohl ex Benth., Labiat.Gen. Spec.: 137 (1833); Bentham in DC. Prodr.12: 136 (1848); Schmidt in Fl. Bras. 8(1): 151 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 226 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 224 (1949).
Mesosphaerum brachystachyum (Pohl ex Benth.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hypenia brachystachys (Pohl ex Benth.) Harley in Bot. J. Linn. Soc. 98: 91 (1988).
Hyptis marifolia Benth. in DC. Prodr.12: 136 (1848); Schmidt in Fl. Bras. 8(1): 150 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 225 (1836); Epling in Revista Mus. La Plata, secc. Bot. 7: 225 (1949); **synon. nov.** Type: Brazil, Villa Rica, Goyaz, March 1840, *Gardner* 3932 (holotype K, isotypes E, BM, P, NY).
Mesospherum marifolium (Benth.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hypenia marifolia (Pohl ex Benth.) *Harley* in Bot. J. Linn. Soc. 98: 91 (1988).
Hyptis pruinosa Pohl ex Benth., Labiat.Gen. Spec.: 137 (1833); Bentham in DC. Prodr.12: 136 (1848); Schmidt in Fl. Bras. 8(1): 151 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 227 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 223 (1949); **synon. nov.** Type: Brazil, Serra San Feliz, Goyaz, July 1819, *Pohl* 2017 (holotype K).
Mesosphaerum pruinosum (Pohl ex Benth.) Kuntze, Revis. Gen. Pl. 2: 527 (1891).
Hypenia pruinosa (Pohl ex Benth.) Harley in Bot. J. Linn. Soc. 98: 91 (1988).
Hyptis inelegans Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 226 (1936); Epling in Revista Mus. La Plata, Secc. Bot. 7: 223 (1949); **synon. nov.** Type: Brazil, between As Sages and the Rio Moquém, Goyaz, 24 Sept. 1828, *Burchell* 7822 (holotype K).
Hypenia inelegans (Epling) Harley in Bot. J. Linn. Soc. 98: 91 (1988).
Hyptis paradisi Harley in Kew Bull. 29: 126 (1974); **synon. nov.** Type: Brazil, ca. 20 km N of Alto do Paraíso, Chapada dos Veadeiros, Goiás, 20 March 1971, *Irwin* et al. 32943 (holotype K, isotypes NY, UC).
Hypenia paradisi (Harley) Harley in Bot. J. Linn. Soc. 98: 91 (1988).

Woody herb to 3 m. *Stems* erect, main axes unbranched or with short lateral branches below long, virgate axes, or intricately branched to immediately below short inflorescence

branches, robust or slender; basal parts pubescent with long simple hairs, glandular hairs and setose hairs; upper parts drying dark purplish-brown, terete, rarely angled, glabrous and waxy, rarely pubescent; internodes fistulose, often with globose swellings. *Leaves* sessile or shortly petiolate, spreading, persistent, overlapping or distant; lamina ovate, 4 — 30 x 2.5 — 25 mm; base shallowly cordate; apex obtuse or mucronate, sometimes acute; margin crenate, regular; upper surface pubescent to tomentose with lanate hairs, glandular hairs and subsessile glands; lower surface pubescent to tomentose with dense subsessile glands and lanate hairs and glandular hairs; petiole 1 — 4 mm long, pubescent with lanate hairs, glandular hairs and subsessile glands. *Inflorescence* a condensed, indeterminate thyrse; phyllomes caducous; branches erect, arched, 2 per node, regularly developed, glabrous and waxy; cymes (1—)2(—9)-flowered, dichasial, densely clustered and obscuring branches; peduncle bracts narrowly lanceolate, 1 — 5 mm; peduncles erect, 3 — 5 mm, glabrous and waxy; paired bracteoles narrowly lanceolate, 1 — 3 mm. *Flowering calyx* vinaceous, erect, cylindrical, straight or sigmoid, 4 — 7 mm, pubescent or tomenotose with simple hairs and glandular hairs; teeth asymmetrical, the posterior 3 connate at base, narrowly deltoid, 1 — 4 mm. *Fruiting calyx* erect, 6 — 9 mm; teeth 1 — 5 mm; throat glabrous. *Corolla* tube violet, 6 — 8 mm, cylindrical, straight or sigmoid; posterior and lateral lobes violet, sometimes with white on margin of tube mouth, 2 — 3 mm; anterior lobe violet, ca. 1.5 mm. *Stamen* filaments white with white hairs, anthers yellow. *Style* white or violet. *Nutlets* trigonous, rarely winged, 2 — 3 x 1 — 1.5 mm.

DISTRIBUTION. Brazil, northern Goiás, Distrito Federal and neighbouring parts of Minas Gerais.

HABITAT. Savanna (cerrado), one record from serpentine rocks. Altitude: 500 — 1500 m.

VERNACULAR NAME. Barbatimão.

BRAZIL.

DISTRITO FEDERAL. Brasília: Reserva Ecologica do IBGE, 16 Aug. 1995 *Atkinson & Giorgio* 150 (K, UNB, IBGE); Brasília: Reserva Ecológica do IBGE, 2 July 1990 *Azevedo & Lopes* 687 (IBGE); Brasília: Região de Palma, 9 June 1981 *Bongatto* 16 (K); Brasília: Fazenda Agua Limpa, ca. 30 km S of Brasília, 12 July 1976, *Davis* 60203 (E, UEC); Brasília: Aguas Emendadas, 25 May 1972, *Fereira* 1146 (HEPH); Brasília: Convênio Florestal de Brasília, 1 June 1960, *Gomes* 1086 (RB); Brasília: Reserva Ecológica do Roncador, 8 May 1978, *Heringer* et al. 477(K); Brasília: Imprensa Oficial, 11 April 1961 *Heringer* 8243/437 (NY, UB); Brasília: road from Sobradino, 10 July 1961, *Heringer* 8466/660 (UC, UB, HEPH); Brasília: Reserva Biologica das Aguas Emendadas, 2 Aug. 1975 *Heringer* 14800 (NY, UB); Brasília: Estação Experimental de Biologia (UNB), 5 May 1976 *Heringer* 15509 (K); Brasília: near Palace Hotel, 18 May 1966, *Hunt* 5515 (K, NY, UC); Brasília: W margin of Lagoa Paranoá, 11 March 1966, *Irwin* et al. 13882 (NY); Brasília: ca. 20 km NE, Córrego Landim, 11

May 1966, *Irwin* et al..15773, (K, NY, UB); Brasília: Cabeça do Veado, 2km from Escola Fazendaria, 9 May 1980, *Kirkbride* 1223 (G); Brasília: Fazenda Água Limpa, 24 July 1980 *Kirkbride* 1343 (UB); Brasília: Fazenda Água Limpa, 25 Sept. 1981, *Kirkbride Jr.* 4451 (K); Brasília: ca.15km W of Radiobras antenna, 14 April 1982, *Kirkbride Jr.* 4728 (K, NY); Brasília: area of Nova Cap, 13 May 1957, *Magalhaes* 9760 (K); 18 May 1982, *Pereira* 13 (K); Brasília: Campus of University, 16 April 1963, *Pires* et al. 9146 (RB, UB); Brasília: Campus of University, 28 April 1963, *Pires* 9503 (UC); Brasília: Fazenda Água Limpa, 13 May 1976, *Ratter & Fonsêca* 2972 (E, K); Brasília: Fazenda Água Limpa, near Vargem Bonita, ca. 18 km SSW of Brasília, June 1976, *Ratter* et al. 3459 (E); Brasília: Córrego Fazendinha, 1984, *Sato* 15 (UB); Brasília: Reserva Ecológica do IBGE, 18 May 1988, *Silva* 662 (IBGE); Brasília: road from the Peninsula, close to Clube do Congresso, 29 May 1965, *Sucre* 376 (UB, RB); Brasília: Zoo-Botânico, 20 July 1965, *Sucre* 748 (RB); Planaltina: BR 020 Sobradinho - Planaltina, km 15, 1 June 1982 *Almeida* et al. 328 (UEC); Planaltina: BR 020 Sobradinho - Planaltina, km 15, 1 Aug. 1985 *Almeida* et al. 1097 (UEC); Planaltina: GO-118, 8-10 km S of São Gabriel, 12 June 1993 *Hatschbach* et al. 59289 (MBM); Sobradinho: ca.10km E of Brasília, 6 July 1966, *Irwin* et al. 18031 (NY).

GOIÁS. Alto Paraíso: GO-12, Chapada dos Veadeiros, 20 Feb. 1975, *Hatschbach* 36338 (K, UC, MBM); Alto Paraíso: km 5-10 S, GO-12, 24 May 1975 *Hatschbach* 36787 (K, MBM); Alto Paraíso: 5 km S on GO-12, 24 May 1975, *Hatschbach* 36807 (K); Alto Paraíso: road to Nova Roma, Rio Bartolomeu, 13 June 1993 *Hatschbach* et al. 59474 (MBM); Alto Paraíso: ca. 1 km W, Chapada dos Veadeiros, 13 Feb. 1966 *Irwin* et al. 12762 (K, NY, UB); Alto Paraíso: ca.19km N, Chapada dos Veadeiros, 20 March 1971, *Irwin* et al. 32801 (K, UC); Alto Paraíso: ca. 20km N, Chapada dos Veadeiros, 21 March 1971, *Irwin* et al. 32886 (K, NY, UC); Alto Paraíso: ca. 20km N, Chapada dos Veadeiros, 20 March 1971, *Irwin* et al. 32943 (K, NY, UC); Alto Paraíso: ca.10km N, Chapada dos Veadeiros, 23 March 1971, *Irwin* et al. 33097 (K, NY); Alto Paraíso: 7 km away on road to Nova Roma, 21 May 1994 *Munhoz* et al. 107 (UB); Alto Paraíso: Chapada dos Veadeiros, 18 July 1964, *Prance & Silva* 58183 (K, NY); Alto Paraíso: 2 km away, Chapada dos Veadeiros, 18 July 1964, *Prance & Silva* 58202 (K, NY, UC); Alto Paraíso: 12 km from Alto Paraíso, road to Nova Roma, 20 May 1994 *Proença* et al. 1142 (UB); Anápolis: 10 km away on BR-153, 22 April 1975 *Hatschbach* 36678 (K, UC, MBM); Anápolis: 11 March 1978, *Mianguago* 187 (RB); Campos anayas: March 1840 *Gardner* 3932 (K, E, BM, P, NY); Cavalcante: between As Araras (= Arraias ?) and Cavalcante, 26 Sept. 1828, *Burchell* 7876 (K); Cavalcante: between As Araras (= Arraias ?) and the Rio Moquém (= Bagagem), 24 Sept. 1828, *Burchell* 7822 (K); Cavalcante: Pasto do Agripino, near Corrego Matthias, 3 May 1986 *Cares* 8 (UB); Cavalcante: Cana Brava (= rio Canabrava), 14 June 1990, *Brooks* et al. TMEX 489 (K); Cocalzinho: road to Braslândia, 8 March 1978, *Paulo* 52 (RB); Cristalina, 18 March 1964, *Pereira* 8982 (RB); Colinas do Sul: 2-3 km N, 15 June 1993 *Hatschbach* et al. 59561 (MBM); Colinas do Sul: 3 - 4 km N, 15 June 1993, *Hatschbach* et al. 59567 (MBM); Corumbá de Goiás: 9 July 1951, *Macedo* 3289 (K); Corumbá de Goiás: ca. 15km N, Serra dos Pirineus, 14 May 1973 *Anderson* 10286 (K, NY,UB); Formosa: ca. 35 km N, Rio Paraná, Goiás, 30 March 1966, *Irwin* et al. 14310 (NY, UB); Formosa: 20 km E, southern Serra Dourada, 16 May 1956, *Dawson* 14872 (UC); Luziânia: road to Luziânia, 1 km after the DF border, 20 July 1990, *Melo & França* 315 (UB); Luziânia: between Gama and Luziânia, 12 July 1964 *Duarte & Mattos* 8430 (K, RB); Luziânia: Fazenda da Pinguela, 22 June 1976 *Heringer* et al. 15874 (K, UB); Niquelândia: km 8 road to Uruaçu, Fazenda Trarías, 13 April 1996, *Fonseca* et al. 871 (K, IBGE); Niquelândia: between Rio Crixas and Rio Maranhão, 1819, *Pohl* s.n. (W, K); Pirenópolis: ca. 20km E, Serra dos Pirineus, 14 Jan. 1972, *Irwin* et al. 34021 (NY); Pirenópolis: Cachoeira do Abade, 25 May 1968, *Onishi* et al. 60 (K); São Felix: Serra San Feliz, July 1819, *Pohl* 2017 (K); Sao Joao d'Aliança: ca. 3km S, 16 March 1971, *Irwin* et al. 31919 (K, P, UC); Sao Joao d'Aliança: ca. 58km N, Serra Geral do Paraná, 18 March 1971, *Irwin* et al. 32112, (K, NY, UC, M); São Jorge: ca.10 km away on road to

Aliança, Chapada dos Veadeiros, 23 July 1995 *Atkinson & Silva Jr.* 143 (K, UNB, IBGE, SPF); Terezina: 13km S, Chapada dos Veadeiros, 16 March 1973 *Anderson* 7242 (NY).
MATO GROSSO. Barra do Garças: ca. 9km NE, 6 May 1973, *Anderson* 9799 (K, NY).
MINAS GERAIS. s. loc., 1816 - 182, *St.Hilaire* 612 (P); s. loc. 1816 - 1821, *St.Hilaire* s.n. (P, UC); Paracatu: BR 040, 24 June 1983, *Hatschbach & Kummrow* 46628 (K, MBM).

E. brachystachys is a very variable species, particularly in habit and the size and indumentum of the leaves. It is defined by its assymetrical calyx teeth, globose fistulae and small, sessile, ovate leaves with cordate bases. It is readily distinguished from *E. densiflora* by the leaf shape and in the field can also be identified by its violet, rather than white, corolla and vinaceous calyx as compared to the yellow-green calyx of *E. densiflora.* It is a plant of savanna (cerrado) and is found in open savanna (campo limpo) through to savanna woodland (cerradão).

As *E. brachystachys* is circumscribed here it includes *Hypenia marifolia, H. pruinosa, H. paradisi* and *H. inelegans* in synonymy. The type of *H. marifolia* has very small leaves and is distinguished by the tomentose indumentum on all parts, particularly the leaves and calyx. Specimens similar to the type of *H. marifolia* occur throughout the range of *E. brachystachys* and there are many intermediates and it seems impossible to justify their maintenance as separate species. *E. concinna* could also represent a similar situation although the aristulate calyx teeth are unusual and support its specific status, at least until further collections become available.

H. pruinosa is very similar to *E. brachystachys* from Distrito Federal, differing principally in the lack of branches on the lower parts and the distant leaves. The collection made by *Brooks et al. TMEX 489* on an expedition to investigate the vegetation of serpentine rock was made in northern Goiás, close to the border with Tocantins and close to where *Pohl 2017*, the type of *H. pruinosa,* was collected. The two collections are very similar and may represent a form of *E. brachystachys* restricted to serpentine rock. *H. inelegans* was distinguished from *H. pruinosa* by the pubescent inflorescence branches, a character state found on several *E. brachystachys* collections. Neither taxon seems sufficiently differentiated to merit maintenance as separate species.

Hypenia paradisi as described by Harley (1974) is localised to the area around Alto Paraíso, in the Chapada dos Veadeiros, but more typical *E. brachystachys* (i.e. with short calyx teeth) is also found in the same area. There are also several collections which it is difficult to assign

with certainty to either taxon and there seems insufficient reason to maintain the specific status of *H. paradisi.*

E. brachystachys is very variable in habit. Plants observed in the field in the vicinity of Niquelândia and Chapada dos Veadeiros in northern Goiás are intricately branched subshrubs with small leaves and the stems bearing the inflorescence are very short so that the overall height is rarely above 1.5 metres. The type specimen represents this variant and was collected from near Niquelândia. Specimens from Distrito Federal are only branched at the base of the stem, with short, straight branches bearing larger leaves. In these plants the inflorescence stems are very long, sometimes up to three metres, and virgate with well-developed globose fistulae. The difference in habit may reflect a taxonomically recognisable difference but all the characters so far identified to define the two types are continuously variable. Until further intensive field work is undertaken, particularly in the area of Goiás to the north of Brasília, it seems most appropriate to maintain *E. brachystachys* as a single, variable species. Hybrids are common in *Eriope* sensu stricto (Harley pers. comm.) and it may be that the variation observed in *E. brachystachys* as it is circumscribed here is a result of hybridisation between distinct species. However no evidence of hybridisation was observed in the field and it is not possible to ascertain its prevalence in *E. brachystachys.*

8. Eriope concinna *(Benth.) R. Atkinson* comb. nov.

Type: Brazil, Serra do Duro, Goyaz, Sept. 1839, *Gardner* 3385 (holotype K).

Hyptis concinna Benth. in DC. Prodr.12: 136 (1848); Schmidt in Fl. Bras. 8(1): 154 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 226 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 225 (1949).
Mesosphaerum concinnum (Benth.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hypenia concinna (Benth.) Harley in Bot. J. Linn. Soc. 98: 91 (1988).

Woody herb to 1.25 m. *Stems* erect, main axes with short lateral branches, slender; basal parts sparsely pubescent with simple and glandular hairs and setose hairs; upper parts drying dark brown, pubescent with simple and glandular hairs, terete; internodes not fistulose or swollen. *Leaves* sessile, spreading, persistent, overlapping, sometimes with smaller leaves clustered in axils; lamina ovate, 3 — 7 x 2 — 5 mm; base cordate; apex obtuse; margin crenate, regular; upper surface pubescent with simple and glandular hairs; lower surface

white-tomentose with simple and glandular hairs. *Inflorescence* a condensed, indeterminate thyrse; phyllomes ovate, 2 — 4 x 1.5 — 3 mm, persistent; branches erect, straight, 2 per node, pubescent with simple and glandular hairs; cymes 1 (— several)-flowered, densely clustered and obscuring branches; peduncle bracts narrowly lanceolate 3 — 4 x 0.5 mm; peduncle erect, 1.5 — 2 mm, sparsely pubescent with simple hairs and subsessile glands; paired bracteoles narrowly lanceolate, ca. 1 mm. *Flowering calyx* not seen. *Fruiting calyx* erect, straight, 7 — 8 mm, pubescent with simple hairs and subsessile glands; teeth symmetric, aristulate, ca. 3 mm; throat with sparse long hairs or glabrous. *Corolla* not seen. *Nutlets* not seen.

DISTRIBUTION. Single locality in western Bahia close to the border with Tocantins (formerly the northern part of the state of Goiás = Goyaz). Figure 10-9.
HABITAT. "In rocky places".

BRAZIL.
TOCANTINS. Serra do Duro, Goyaz (=Bahia), Sept. 1839, *Gardner* 3385 (K).

Unfortunately *E. concinna* is only represented by one specimen. The tiny, leaves with tomentose indumentum on the lower surface and aristulate calyx teeth are very distinctive. However some specimens of *E. brachystachys* have a similar leaf size and shape, particularly those previously described as *H. marifolia*. The indumentum is only tomentose on the lower surface in *E. concinna* compared to both surfaces in specimens described as *H. marifolia*. The calyx teeth are much narrower than those of some collections of *E. brachystachys* (formerly *H. paradisi*). Further collections may prove *E. concinna* to belong within the very variable *E. brachystachys* but for the present it seems sufficiently distinct to be maintained as a separate species.

Serra do Duro is ca. 200 km north of the nearest *E. brachystachys* locality and *E. concinna* may represent an outlier of *E. brachystachys* which is sufficiently isolated to maintain morphological distinctiveness. Collections for intermediate localities are lacking and further collecting in the area of Serra do Duro and neighbouring parts of Tocantins could help to confirm the uniqueness of *E. concinna* or justify its relegation into *E. brachystachys*.

9. Eriope densiflora *(Pohl ex Benth.) R. Atkinson* comb. nov.

Type: Brazil, Carretão, Engenho São Antonio e Trahiras, Goyaz, July 1819, *Pohl* 1759 (holotype W, isotype K).

Hyptis densiflora Pohl ex Benth., Labiat. Gen. et Sp.:137 (1833); Benth. in DC. Prodr.12: 135; Schmidt in Fl. Bras. 8(1): 149 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 228 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 220 (1949).
Mesosphaerum densiflorum (Pohl ex Benth.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hypenia densiflora (Pohl ex Benth.) Harley in Bot. J. Linn. Soc. 98: 91 (1988).
Hyptis melochioides St.Hil. ex Benth., Labiat.Gen. Spec.: 137 (1833); Bentham in DC. Prodr. 12: 135 (1848); Schmidt in Fl. Bras. 8(1): 150 (1858). Type: Brazil, Minas Geraes, 1816 - 1821, *St. Hilaire* s.n. (holotype P).
Hyptis densiflora var. *dolichodon* Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 228 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 221 (1949). Type: Brazil, Serra Geral near San Domingos, Goyaz, May 1840, *Gardner* 4316 (holotype K, isotypes BM, E, G, P, NY, UC).

Woody herb to 3 m. *Stems* erect, main axes with short lateral branches below long, virgate axes, robust; basal parts pubescent with dense simple hairs and setose hairs; upper parts dark reddish-brown, terete; glabrous and waxy; internodes fistulose with elongate swellings. *Leaves* petiolate, spreading, persistent, overlapping or distant; lamina lanceolate to ovate, 30 — 65 x 7 — 20 mm; base acute or truncate; apex acute to rounded; margin serrate, regular; upper surface densely pubescent with simple hairs; lower surface tomentose with lanate hairs and sparse minute glandular hairs; petiole 3 — 25 mm long, with simple hairs. *Inflorescence* a condensed indeterminate thyrse; phyllomes caducous; branches erect, 2 or 4 per node, regularly developed, glabrous and waxy; cymes 1— 3-flowered, densely clustered and obscuring branches; peduncle bracts narrowly lanceolate, 2 — 3 mm, persistent, peduncles erect, 2 — 4 mm, with sparse glandular hairs; paired bracteoles narrowly lanceolate, ca. 2 mm. *Flowering calyx* greenish-yellow, erect, cylindrical, sigmoid, 7 — 8 mm, sparsely pubescent with short hairs and minute glandular hairs; teeth 2.5 — 3 mm, asymmetrical, the posterior 3 connate at base, narrowly deltoid. *Fruiting calyx* erect, 8 — 9 mm, sigmoid; teeth 2.5 — 5 mm, narrowly deltoid; throat glabrous. *Corolla* tube white, 10 — 12 mm, cylindrical, sigmoid; posterior and lateral lobes white, sometimes with pale pink spots, ca. 2 mm. *Stamens* filaments white, long hairs sparse or absent; anthers yellow or brown. *Style* white. *Nutlets* trigonous, 1.5 — 2 x 0.75 — 1 mm. Figure 10-3.

DISTRIBUTION. Brazil; Goiás, Distrito Federal, Minas Gerais, between the rivers Araguaia and São Francisco on the Central Plateau of Brazil (figure 10-9).

HABITAT. Savanna (cerrado), on the margin of gallery forest, one record from serpentine rocks. Altitude: 600 — 1150 m.

BRAZIL.

DISTRITO FEDERAL. Brasília: Jardim Botânico, ca. 1 km S of administration buildings, 8 Aug. 1995, *Atkinson & Giorgio* 146 (UNB, IBGE, K); Brasília: Reserva Ecológica do IBGE, 2 Sept. 1995, *Atkinson & Giorgio* 151 (UNB, IBGE, K); Brasília: 6 May 1969, *Chaves* 2572 (K); Brasília: Parque do Gama, 45 km S of Brasília, 12 July 1976, *Davis* 60156 (E, UEC); Brasília: Parque do Gama, 45 km S of Brasília, 12 July 1976, *Davis* 60188 (E, UEC); Águas Emendadas: 2 June 1972, *Fereira* 1256 (HEPH); Brasília: Gama, 24 April 1972, *Fereira* 1361 (HEPH); Brasília: IBGE, start of forest at rio São Bartolomeu, 11 July 1979, *Heringer* et al.1489 (K); Brasília: Águas Emendadas National Park, 17 July 1979, *Heringer* et al.1851 (K); Brasília: IBGE, basin of rio São Bartolomeu, 22 May 1980, *Heringer* et al.4855 (K, UEC); Brasília: 20 km E, Chapada de Contagem, 19 Sept. 1964, *Irwin & Soderstrom* 5319 (K, NY); Brasília: ca. 15 km E , 17 Sept. 1965, *Irwin* et al.7817 (K, NY); Brasília: Paranoá Lake, 24 Aug. 1961, *Irwin* et al.8392 (K, NY); Brasília: Bananal stream, N of Buraco stream, 21 June 1992, *Kirkbride* 1308 (UB); Brasília: Palmeiras stream, 17 June 1980, *Kirkbride Jr.* 4365 (UB); Brasília: rio São Bartolomeu, Quebrada dos Neri, BR 251, 21 June 1992, *Melo & Franca* 768 (UB); Brasília: between city and calcareous zone, 26 April 1963, *Pires* et al.9404 (RB, UB); Brasília: road to Unaí, 18 July 1993, *Proença* 856 (UB); Brasília: Águas Emendadas Biological Reserve, 20 June 1983, *Ramos* 294 (HEPH); Brasília: Olympic Centre, UNB campus, 20 June 1980, *Ribeiro* s.n. (UB); Planaltina: ca. 12 km NNE on DF2, 18 June 1976, *Ratter* et al.3178 (K, E).

GOIÁS. Barro Alto: 11 - 12 km SW, 26 June 1990, *Brooks & Reeves* TMEX 666 (K); Caiapônia: 35 km W, 25 July 1977, *Hatschbach* 40075 (K, UC, MBM); Carre: road to Rio Carto, Goyaz, 15 May 1895, *Glaziou* 21919 (K, G, P, R, S); Cavalcante: between São José and rio Bagagem, Goyaz, 17 Sept. 1828, *Burchell* 7662 (K); Cavalcante: between rio Chupeteiro and rio Bezerro (between Cavalcante, Goiás and Conceição, Tocantins), Goyaz, 10 Oct. 1828, *Burchell* 8004 (K); Cavalcante: Corrego Rica (between Cavalcante, Goiás and Conceição, Tocantins), Goyaz, Oct. 1828, *Burchell* 8065 (K); Chapada de São Marcos: Goyaz, Aug. 1834, *Riedel & Lund* 2857 (NY); Dona Barbara to Sobradinho, Goyaz, 22 Sept. 1894, *Glaziou* s.n. (P); Formosa: rio Tiquiri, 25 May 1967, *Heringer* 11461 (K, NY, UB); Goiás Velha: between town and Serra Dourada, 17 July 1964, *Duarte & Mattos* 8395 (UB, RB, NY, G, K); Luziânia: 6 June 1975, *Heringer* 14686 (K, NY, UEC, UB, HEPH); Luziânia: Fazenda da Pinguela, 22 June 1976, *Heringer* et al.15874* (K, NY, UEC); Niquelândia: 26 July 1952, *Macêdo* 3666 (UC, S); Niquelândia: road near nickel mine, *Fonseca* et al. 991, 31 May 1996; Porougatu: on way to Uruaçu, Belém-Brasília highway, 13 Aug. 1963, *Maguire* et al.56176 (NY); São Domingos: Goyaz, May 1840, *Gardner* 4316 (K, BM, E, G, P, NY, UC); São Felix: Carretão, Engenho São Antonio e Trahiras, July 1819, *Pohl* 1759 (W, K); São João d'Aliança: 21 km S, Chapada dos Veadeiros, 20 April 1956, *Dawson* 14476a (UC); São João da Aliança: highway GO-12, 23 May 1975, *Hatschbach* 36713 (K, UC, MBM); São João da Aliança: Buriti Alto, 15 Oct. 1990, *Hatschbach & Silva* 54535 (MBM); São João da Aliança: 3 km S, 16 March 1971, *Irwin* et al.31943 (K, P, NY, UC, S).

MINAS GERAIS. Paracatú: Brasília to Belo Horizonte road, 3 June 1960, *Heringer & Rizzini* 7558 (UB).

TOCANTINS. Mission of Duro, Goyaz (= Tocantins), Oct. 1839, *Gardner* 3384 (K).

Heringer et al.15874 at K is mixed with *E. brachystachys*.

E. densiflora is one of the most distinctive species in section *Hypenia*. Its leaves are distinctly petiolate and lanceolate with a cuneate base and serrate margin and much larger than the ovate leaves with cordate base and crenate margin of *E. brachystachys*. The colour of the calyx is very distinctive in the field, being a strong acid yellow-green. It contrasts markedly with the white corolla. It has only been observed in cerrado close to the margin of gallery forest.

E. densiflora, although the type species for section *Densiflorae* sensu Bentham and Epling, differs to a greater degree from the other species placed in this group than they do from each other. It seems to be somewhat isolated and has many characters in common with species of section *Hypenia,* particularly *E. subrosea,* notably the size and shape of the leaf. The closeness of *E. densiflora* to section *Hypenia* is confirmed by ITS sequences which place it in the middle of section *Hypenia* rather than with *E. brachystachys.* This result is based on limited sampling but it does seem to be reflected to some extent in morphology.

Hyptis densiflora var. *dolichodon* was described by Epling from a Gardner specimen. Other collections of *E. densiflora* with immature inflorescences are very similar to this collection and the variety has been reduced to synonymy. *Hyptis melochioides* was described by Bentham (1848) on the basis of a single specimen. This specimen has rather broad leaves but is otherwise typical of *E. densiflora* and was reduced to synonymy by Epling (1936).

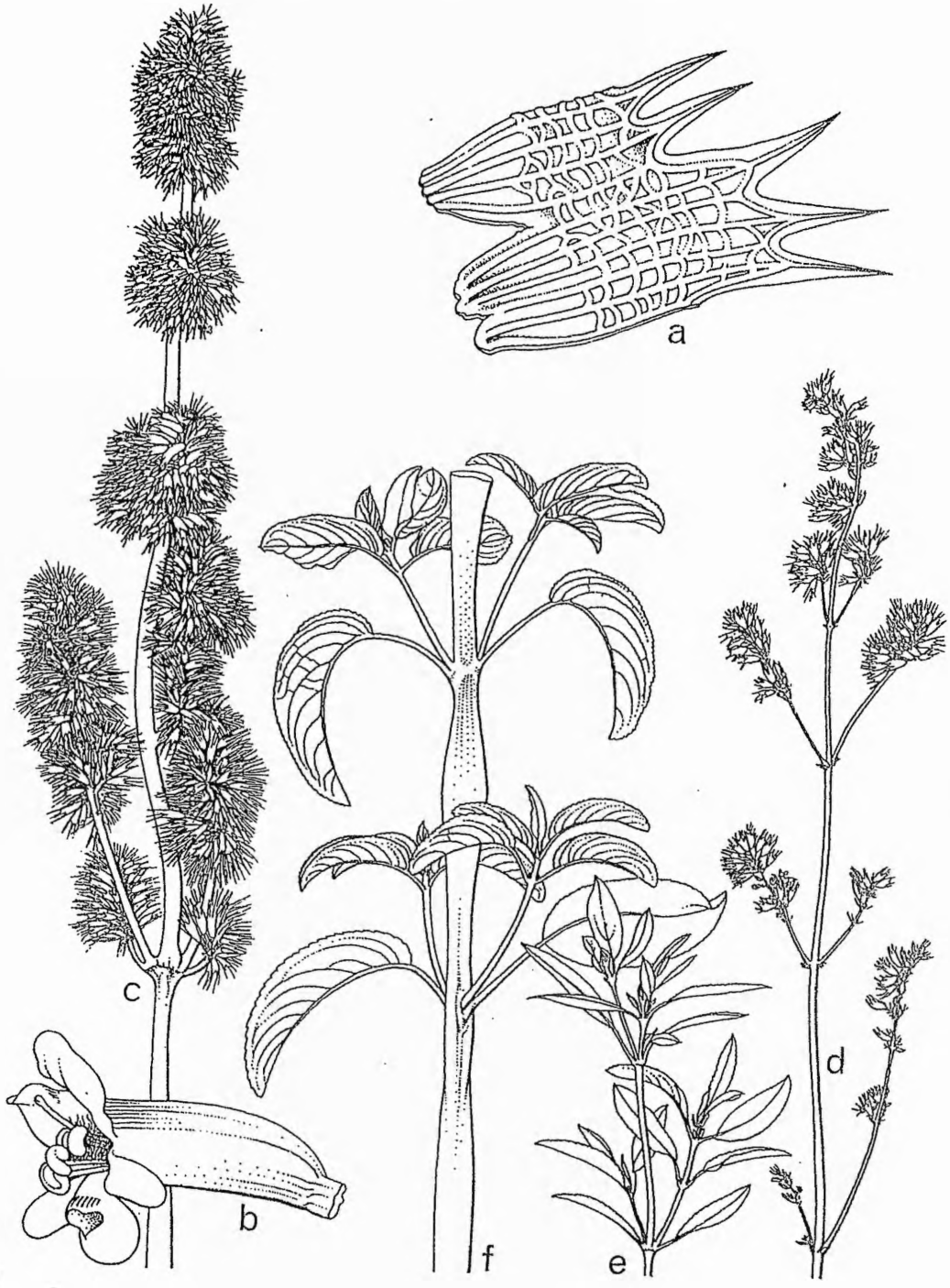


Figure 10-3: ***Eriope densiflora***

a) fruiting calyx (x5) (*Ratter* et al. 3178); b) corolla (x5) (*Ratter* et al. 3178); c) inflorescence (x½) (*Ratter* et al. 3178); d) inflorescence (x½) (*Irwin* et al. 31943); e) leaves and stem (*Irwin* et al. 31943); leaves and stem (*Irwin* et al. 31943). Drawn by Emmanuel Papodopoulos.

vi) Section *Hypenia* (Mart. ex Benth.) R. Atkinson comb. nov.

Subsection *Laxiflorae* (Benth. in DC.) R. Atkinson comb. nov.

10. Eriope aristulata *(Epling) R. Atkinson* comb. nov.

Type: Brazil, Morro de Canto Gallo, Goyaz, 1828, *Burchell* 7105 (holotype K, isotype UC).

Hyptis aristulata Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 231 (1936); Epling in Revista Mus. La Plata, Secc. Bot. 7: 231 (1949).
Hypenia aristulata (Epling) Harley in Bot. J. Linn. Soc. 98: 91 (1988).
Hyptis macrantha St.Hil.ex Benth. sensu Epling & Mathias in Brittonia 8: 298 (1957) **pro parte**.

Woody herb to 1.8 m. *Stems* erect, unbranched with long, virgate axes, slender; basal parts sparsely or moderately pubescent with subsessile glands and minute glandular hairs and dense setose hairs; upper parts light to reddish brown, terete, sparsely to moderately pubescent with subsessile glands, minute glandular hairs and sparse setose hairs, wax absent, rarely present; internodes not fistulose or swollen. *Leaves* petiolate, spreading, frequently caducous, distant; lamina lanceolate to ovate, 20 — 95 x 9 — 47 mm; base cuneate or truncate, sometimes cordate; apex acute or obtuse; margin dentate or serrate, regular; upper surface pubescent with dense, or sparse simple hairs and sparse subsessile and stalked glands; lower surface pubescent with dense simple hairs and sparse subsessile and stalked glands; petiole 3 — 11 mm, pubescent with setose hairs. *Inflorescence* an open, indeterminate thyrse, few-flowered; phyllomes narrowly lanceolate, frequently caducous; branches spreading, straight, 2 per node, frequently not regularly developed, pubescent with sparse to moderate subsessile glands and minute glandular hairs and sparse setose hairs; cymes 1-flowered, 2 — 30 mm apart along branches; peduncle bracts narrowly lanceolate, 1 — 2 mm; peduncle deflexed, 4 — 22 mm, with glandular hairs; paired bracteoles erect, narrowly lanceolate, 1 — 2 x 0.5 — 0.75 mm. *Flowering calyx* green with vinaceous tinge, deflexed, gibbous-campanulate, 7 — 10 mm, moderately pubescent with short simple hairs and glandular hairs; teeth 3 — 7 mm, symmetrical, aristulate. *Fruiting calyx* cylindrical, declinate, 14 — 15 mm; throat +/- glabrous; teeth 6 — 7 mm, aristulate. *Corolla* tube salmon-pink to red, 12 — 15 mm, cylindrical, slightly bulbous on posterior side; posterior and lateral lobes salmon-pink to red, ca. 5 mm, pointed, sometimes twisted, spreading; anterior lobe salmon-pink to red with yellow mark on retracted apex, 3 — 4 mm. *Stamen*

filaments pink with white hairs; anthers yellow. *Style* pink. *Nutlets* ovoid in section, ca. 3.5 x 2 mm.

DISTRIBUTION. Brazil, Goiás endemic to the vicinity of Niquelândia (figure 10-11).

HABITAT. Savanna (cerrado). Altitude: 350 — 400 m.

BRAZIL.

GOIÁS. Colinas do Sul: 2 - 3 km N, 15 June 1993, *Hatschbach* et al. 59553 (MBM); Niquelândia: 40 km N in Serra da Mesa, 22 July 1995, *Atkinson* et al.135 (UB, IBGE, SPF, K); Niquelândia: 40 km N in Serra da Mesa, 22 July 1995, *Atkinson* et al.138 (UB, IBGE, SPF, K); Niquelândia: ca. 25 km NE in Serra da Mesa, near Codemin, 22 July 1995, *Atkinson* et al. 141 (UB, IBGE, SPF, K); Niquelândia: peak near Morro de Canto Gallo, 1828, *Burchell* 7105 (K, UC); Niquelândia: near nickel mine, 13 Aug. 1996, *Mendonça* et al. 2565 (K, IBGE).

Specimen incertae sedis

BRAZIL.

GOIÁS. Niquelândia: 24 July 1952, *Macêdo* 3634 (NY, S).

E. aristulata is one of the most distinctive species in subsection *Laxiflorae*. *E. subrosea* has the closest geographical distribution of a species in the subsection but *E. aristulata* is readily distinguished by its long aristulate calyx teeth, long hairs on the leaves and stems, red or pink corolla and lack of globose swellings on the stem. The area between Niquelândia and Alto Paraíso is rich in species of section *Hypenia* and two species from subsection *Ellipticae* are found here, *E. niquelandiensis* and *E. calycina*. The latter two species are readily separated from *E. aristulata* in the field by their resupinate flowers and much more robust habit. Neither species has the long calyx lobes of *E. aristulata* or long hairs on the upper stems and leaves. *E. aristulata* is common in cerrado throughout its range (figure 10-11).

Macêdo 3634 is a collection from the Niquelândia area which differs significantly from collections assigned to *E. aristulata* and possibly represents a different species in subsection *Laxiflorae*. The frequently branched inflorescence, the glandular indumentum on the branches, the regular arrangement of the closely spaced cymes, the long, sparsely pubescent or glabrous peduncle, the large lobes of the corolla and the irregularly lobed leaves, all contribute to the distinctive appearance of this collection. The deltoid calyx lobes, <=3 mm long and lack of long hairs on the leaves and upper stems make it impossible to assign to *E. aristulata*. It is possibly more closely related to *E. macrosiphon* which has similar indumentum in the inflorescence and lobed leaves but the inflorescence arrangement and

long peduncle give it a very characteristic appearance. However vegetative parts are poorly represented on the replicates seen and further, more complete, collections are required before it can be described with confidence as a new species.

11. Eriope caiaponiensis *R. Atkinson* sp. nov.

Type: Serra do Caiapó, ca. 30 km due S of Caiapônia, Goiás, 29 April 1973, *Anderson* et al. 9395 (holotype UB)

E. reticulatae affinis sed inflorescentiis irregularibus ramosis confertis et ramis cum trichomatibus glandibus minutis differt.

E. caiaponiensis differs from *E. reticulata,* and all other species in subsection *Laxiflorae,* in its very irregular and crowded inflorescence and in the presence of minute glands on the inflorescence branches.

Woody herb to 1 m. *Stems* erect, sometimes branched below long, virgate axes, slender; basal parts pubescent with simple hairs and setose hairs; upper parts drying black or dark brown, terete, glabrous, waxy; internodes fistulose, sometimes slightly swollen. *Leaves* petiolate, spreading, persistent, overlapping or distant; lamina broadly to narrowly lanceolate, 35 — 125 x 5 — 35 mm; base cuneate; apex acute; margin serrate, regular; upper surface sparsely pubescent with short simple hairs and glandular hairs; lower surface tomentose with lanate hairs and glandular hairs. *Inflorescence* an irregular, open, indeterminate thyrse, floriferous; phyllomes narrowly lanceolate, frequently caducous; branches erect or spreading, straight, 2 or 4 per node, upper branches frequently aborted and forming an irregular umbellate shape, lower branches glabrous, waxy, upper branches with sparse simple hairs and minute glandular hairs; cymes 1-flowered, 2 — 35 mm apart towards apex of branch and in branch axils; peduncle bracts narrowly lanceolate, ca. 1.5 mm; peduncle erect, 3 — 9 mm, pubescent with simple hairs and minute glandular hairs; paired bracteoles erect, narrowly lanceolate, ca. 1.5 x 0.5 mm. *Flowering calyx* pale green, erect or horizontal, gibbous-campanulate, 4 — 6 mm, sparsely pubescent with simple hairs and minute glandular hairs; teeth broadly deltoid, 1 — 2 mm. *Fruiting calyx* erect, cylindrical, 8 — 9 mm; teeth broadly deltoid -rostrate, ca. 2 mm; throat with sparse long hairs or glabrous. *Corolla* tube red, cylindrical, straight, 12 — 13 mm; posterior and lateral lobes red, ca. 3 mm; anterior lobe red, ca. 3 mm. *Stamen* filaments with dense hairs. *Style* pink. *Nutlets* ovoid in section, ca. 3.5 x 2 mm.

DISTRIBUTION. Brazil, Goiás, Serra do Caiapó, south of Caiapônia (figure 10-11).
HABITAT. Savanna (cerrado). Altitude: 950 — 1200 m.

BRAZIL.
GOIÁS. Caiapônia: ca. 30 km due S, Serra do Caiapó, 29 April 1973, *Anderson et al.* 9395 (UB); Caiapônia: between Jataí and Caiapônia, 40 km from Caiapônia, June 1966, *Hunt & Ramos 6268* (K); Caiapônia: Serra do Caiapó, ca. 40 km S, *Irwin et al. 17772* (K).

E. caiaponiensis has an irregular inflorescence which has an umbellate shape because of the irregular development of the upper branches and the clustering of the flowers at the branch apices, rather than the usual pyramidal shape formed by regular development of all branches as in other species of subsection *Laxiflorae*. The inflorescence contributes to the distinctive appearance of this taxon. The lanceolate leaves (which are yellow when dried on herbarium sheets), short corolla and minute glandular hairs are also useful characters to identify *E. caiaponiensis*.

E. caiaponiensis is localised to a small area in southwestern Goiás on the Serra do Caiapó near Caiapônia and is relatively geographically separated from other species in subsection *Laxiflorae* (figure 10-11).

12. Eriope hatschbachii *R. Atkinson* sp. nov.

Type: Terenos, Mato Grosso do Sul, 20 Feb. 1970, *Hatschbach* 23882 (holotype MBM, isotypes K, UC).

E. macrosiphoni affinis sed foliis lanceolatis et calycibus longis recedit.

This species can be distinguised from *E. macrosiphon* by its narrowly lanceolate leaves and long calyx tube.

Woody herb to 2 m. *Stems* erect, unbranched with long, virgate stems, robust; basal parts with setose hairs; upper parts drying dark purplish-brown, terete, glabrous and waxy; internodes fistulose, swollen along length. *Leaves* petiolate, erect, persistent, distant; lamina narrowly lanceolate, 10 — 25 x 40 — 150 mm; base truncate; apex acute; margin coarsely serrate; upper surface sparsely pubescent with simple hairs; lower surface tomentose with lanate hairs and subsessile glands; petiole 8 — 50 mm long, glabrous and waxy.

Inflorescence a regular, open, indeterminate thyrse, few-flowered; phyllomes narrowly lanceolate, frequently caducous; branches erect, 2 per node, regularly developed, glabrous, waxy; cymes 1-flowered, regularly spaced 8 — 28 mm apart, sometimes in branch axils; peduncle bracts lanceolate, ca. 2 mm long; peduncle erect, 4 — 7 mm, sparsely pubescent with simple hairs, glandular hairs and minute glandular hairs along length or glabrous with tuft of hairs below calyx; paired bracteoles erect, narrowly lanceolate, 1.5 — 2 x 0.5 — 0.75 mm. *Flowering calyx* erect, gibbous-campanulate, 6 — 7 mm, moderately pubescent with simple hairs, glandular hairs and minute glandular hairs; teeth broadly deltoid -rostrate, 2 mm. *Fruiting calyx* erect, narrowly cylindrical, 11 — 13 mm; teeth broadly deltoid -rostrate, 2 — 3 mm; throat glabrous. *Corolla* tube red, narrowly cylindrical, straight or curved, 11 — 13 mm; posterior and lateral lobes red, 3 — 4 mm; anterior lobe red, ca. 2 mm. *Nutlets* ovoid in section ca. 3 x 2 mm.

DISTRIBUTION. Brazil, Mato Grosso do Sul, surroundings of Campo Grande (figure 10-11).

HABITAT. Savanna (cerrado). Altitude: not recorded.

BRAZIL.

MATO GROSSO DO SUL. Camapuã: road from Campo Grande to Cuiabá, 14 May 1973, *Hatschbach* 31890 (MBM); Campo Grande: way to Aquidauana, 9 Nov. 1977, *Rodrigues* 335 (K); Terenos: 20 Feb. 1970, *Hatschbach* 23882 (K, MBM, UC).

Specimen incertae sedis

BRAZIL.

MATO GROSSO DO SUL. Aquiduana: Col. Paxixi, 12 July 1970, *Hatschbach & Guimarães* 24587 (MBM).

E. hatschbachii can be recognised by the long, narrowly lanceolate leaves with their coarsely serrate margins and the long, narrow fruiting calyx. *Hatschbach & Guimarães 24587* is a much less robust plant than the other specimens, particularly the stems and the inflorescence, but has similar leaves and flowers and comes from the same area. However, it also differs in the lower leaf, instead of tomentose indumentum there are conspicuous subsessile glands and no lanate hairs. For these reasons this specimen has not been included in this description, although it may prove to belong to this taxon if further collections become available.

It is not clear whether *E. hatschbachii* flowers are resupinate or not. The inflorescence characters, particularly the regular distribution of cymes along the branches, suggest that this

taxon belongs subsection *Laxiflorae*. However, the position of several flowers on the herbarium sheet suggests that they could be resupinate. All these flowers have an obvious twist in the peduncle which could be an artefact of the pressing process or may occur in nature.

It is endemic to a relatively small area in Mato Grosso do Sul (figure 10-11) and is named for the prolific Brazilian collector, Gert Hatschbach, whose collections are always exemplary.

13. Eriope macrosiphon *(Briq.) R. Atkinson* comb. nov.

Type: Brazil, Matto Grosso, *Kuntze* s. n., July 1892, (holotype NY).

Hyptis macrosiphon Briq. in Bull. Herb. Boissier 4: 785 (1896); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 233 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 237 (1949).
Mesosphaerum macrosiphon Briq. loc. cit.
Hypenia macrosiphon (Briq.) Harley in Bot. J. Linn. Soc. 98: 92 (1988).
Hyptis lindmaniana Briq. in Ark. Bot. 2, 10: 4 (1904).
Mesosphaerum lindmanianum Briq. loc. cit. Type: Brazil, Mato Grosso, Serra de Itapirapuau, 1 May 1894, *Lindman* 3381 (K, UC).
Hyptis mattogrossensis Pilger in Bot. Jahrb. Syst. 30: 191 (1902). Type; Brazil, Matto Grosso, Cuyaba, *Pilger* 407 (B, not seen).
Hyptis macrantha St. Hil. ex Benth. sensu Epling & Mathias in Brittonia 8: 298 (1957) **pro parte.**

Woody herb to 2 m. *Stems* erect, unbranched with long, virgate axes, moderately robust; basal parts pubescent with simple hairs and setose hairs; upper parts drying dark brown or pinkish brown, moderately pubescent with simple hairs, wax absent, rarely present; internodes fistulose, globose swellings rarely present. *Leaves* petiolate, spreading; lamina ovate to lanceolate, frequently lobed, 9 — 130 x 8 — 75 mm; base cordate or truncate; apex acute or mucronate; margin coarsely dentate, sometimes irregular; upper surface sparsely to moderately pubescent with short simple hairs; lower surface moderately pubescent (rarely tomentose) with short simple hairs and subsessile glands; petiole 7 — 50 mm, glabrous or with short simple hairs. *Inflorescence* a regular, open, indeterminate thyrse, floriferous; phyllomes narrowly lanceolate, caducous; branches spreading, straight or slightly arched, 2 or 4 per node, lower branches moderately pubescent with simple hairs, upper branches with

glandular hairs; cymes 1-flowered, closely spaced 3 — 28 mm apart; peduncle bracts narrowly or broadly lanceolate ca. 1.5 mm long; peduncle erect, 3 — 15 mm, pubescent with glandular hairs; paired bracteoles narrowly or broadly lanceolate, 1.5 x 0.5 — 0.75 mm. *Flowering calyx* green with vinaceous tinge, erect, shortly gibbous-campanulate, 3.5 — 4.5 mm, moderately pubescent with glandular hairs; teeth symmetrical, broadly deltoid -rostrate, 1.5 — 2 mm. *Fruiting calyx* erect, cylindrical, frequently with swollen base, 9 — 10 mm long; throat with moderately dense long hairs; teeth ca. 2 mm. *Corolla* tube red, orange or pink, cylindrical, straight, 9 — 15 mm; posterior and lateral lobes 3 — 4 mm long; anterior lobe 3 — 4 mm. *Stamen* filaments with white hairs, anthers yellow. *Style* pink. *Nutlets* ovoid in section, ca. 3.5 x 2 mm.

DISTRIBUTION. Dispersed from western and northern Goiás throughout Mato Grosso and into southern Pará, eastern Rondônia, Mato Grosso do Sul and Paraguay. One record from savanna in northern Bolivia (figure 10-10).
HABITAT. Savanna (cerrado). Altitude 250 — 1300 m.

BOLIVIA. Beni, San Joaquin, Vuelta Grande, Mamore Province, *Leigh-Hunt et al.* 110, Aug. 1996 (E).

BRAZIL. s. loc., Oct. 1874, s. collector 751 (K).
GOIÁS. Balsas: Jataí, Rio Corrente, 19 July 1951, *Macêdo*, 3327 (UC); Caiapônia: ca. 35 km W, 25 July 1978, *Hatschbach* 40081 (K, MBM); Corumbá de Goiás: ca. 15km N, Serra dos Pirineus, 14 May 1973, *Anderson* 10293 (K, NY); Cristalina: 13 km west on BR-040, 13 Aug. 1980, *Hatschbach* 43092, (MBM, K); Cristalina: 1 km SE, km 107, *Mori* et al.16942, 24 July 1984, (K); Cristalina: Fazenda Lopo Botelho, *Pires & Mattos* 9821, 7 July 1963, (K, UB); Mineiros: BR 060 20 July 1974, *Hatschbach* 34629 (K, MBM); Pirenópolis: ca. 20km E, Serra dos Pirineus, 16 Jan.1972, *Irwin* et al. 34367 (K, NY).
MATO GROSSO. s. loc., 1892, *Briquet* s.n. (K); s.loc., July 1892, *Kuntze* s.n. (NY); s. loc., s.dat., *Smith* 34878 (R); Arcnápolis: 50-70 km W of junction for Arcnápolis on BR 364, Chapada do Parecis, 12 May 1995, *Hatschbach* et al. 62700 (MBM); Barra do Garças: 80 km N on road to Nova Xavantina, Vale de Sonhos, 26. Aug.1972, *Ratter & Fonsêca* 2281 (K, E); Cascalheira: RGS expedition base camp, 12.5km SW, 23 Sept.1968, *Harley & Souza* 10188 (K, RB, UB); Cascalheira: RGS expedition base camp, c.12km SW, near Lago de Leo, 28 Sept. 1968, *Harley & Souza* 10329 (K); Cascalheira: RGS expedition base camp, 1968, *Harley* s.n. (K); Cascalheira: RGS expedition, plot 4, 14 July 1968, *Taituba in Richards* 6417 (K); Chapada dos Guimarães: Colégio Evangélico de Burití, 9 May 1983, *Barcia* et al. 5025 (R); Chapada dos Guimarães: 13 July 1994, *Dubs* 1535 (E, K, Z); Chapada dos Guimarães: viewpoint, *Hatschbach* et al. 66612 (K, MBM); Chapada dos Guimarães: córrego Congonhas, *Hatschbach* 66704 (K, MBM); Chapada dos Guimarães: 2 Aug. 1902, *Malme* 2 Aug. 1902 (S); Chapada dos Guimarães: 31 May 1903, *Malme* (S); Chapada dos Guimarães: 20 July 1902, *Sladen* 387 (BM); Cuiabá: between town and Chapada dos Guimarães, 2 Feb. 1978, *Harley* 20423 (K); Cuiabá: Caxipó da Paute, March 1911, *Hoehne* 2845 (R); Cuiabá: Caxipó da Paute, March 1911, *Hoehne* 2848 (SP); Cuiabá: 9 Jan. 1894, *Malme* 1162 (R, S); Cuiabá: 30 Nov. 1893, *Malme* 1162 (S); Cuiabá: 24 Nov. 1893, *Malme* 1162 (S); Cuiabá: 11 Dec. 1894, *Malme* 1162 (S); Cuiabá: 14 June 1902, *Malme* 1730 (S); Cuiabá: May 1927, *Smith* 116

(K); Cuiabá: 1891-2 *Moore* 23 (BM); General Carneiro: Meruri, Sept. 1963, *Hartmann* s.n. (SP); Nova Xavantina: road to Cachimbo, 80km from Nova Xavantina, 4 June 1966, *Hunt* 5779 (K, UC, SP); Nova Xavantina: ca. 87 km N, Serra do Roncador, 2 June 1966, *Irwin* et al. 16474 (K, RB); Nova Xavantina: 8 km S, 31 Aug. 1982, *Kirkbride* 1613 (UB); Nova Xavantina: ca.1km S, 19 July 1967, *Ratter & Castro* 123 (K); Nova Xavantina: ca. 200 km N on road to São Felix, Rio Turvu, 4 June 1968, *Santos & Souza* 1642 (E, K, NY); Rio Amalar: near source of Rio Paraguay, May 1927, *Smith* 165 (K); Rondonópolis: 16 May 1973, *Hatschbach* 31993 (K); Serra de Itapirapuau, 1 May 1894, *Lindman* 3381 (K, UC).

MATO GROSSO DO SUL. Campo Grande: Estaça, 10 Sept. 1936, *Archer & Gehrt*, 36395 (SP); Campo Grande: 40 km N, Jatobá, 15 July 1966, *Goodland* 290 (K); Rio Brilhante: Rio Anhandui, 23 Oct. 1970, *Hatschbach* 25105 (K, UC); Amambaí: Rio Pandio 13 Feb 1983, *Hatschbach* 46178 (K, G, MBM); Amambaí: 20 km W, MT-642, 16 Dec. 1983, *Hatschbach & Callejas* 47290 (G, MBM).

PARÁ. S. loc., s.dat., s.n., (P) Serra do Cachimbo, SE, June 1955, *Alvarenga* (90546) (UC, RB);.

RONDÔNIA. Vilhena: 22 May 1979, *Silva & Rosário* 4565 (NY).

PARAGUAY.

Canendiyú: 50 km from Capitán Bado to Ype Hú, 12 Jan. 1979, s.nom., COTESU 19487 (NY); Canendiyú: 65 km S of Capitán Bado, 29 March 1983, *Simonis* et al. 234; Capitán Bado: on way to Ype Hú, 5 Feb. 1982, *Casas* 5988 (G, NY); Esperanza: Sierra de Amambay, 1907-8 *Hassler* 10256 (G, NY); Sierra de Amambay: 1907-8, *Hassler* 10855 (K, G, NY, UC, S, P); Sierra de Maracuyá, s.dat. *Hassler* 5159 (K, G, NY, UC, S, P).

Specimen incertae sedis

BRAZIL.

MATO GROSSO. Cuibá: between Cuiabá and Chapada de Guimarães, 2 Feb. 1978, *Harley* 20424 (K).

E. macrosiphon has a similarly widespread distribution and variability in its character states to *E. reticulata* but it can be distinguished from all other species of subsection *Laxiflorae* by a number of characters. These characters are: cordate leaves with an irregular, dentate margin which are frequently lobed; cymes close together in the inflorescence on short peduncles; and pubescence on the inflorescence branches. All these characters are seen on the specimens from Rondônia and Bolivia, the most westerly collections for any species of *Eriope* (figure 10-10). These latter collections have tomentose indumentum on the lower leaf and the leaves are not lobed but the inflorescence varies little from that of collections from Mato Grosso. Specimens from central Mato Grosso do Sul and Cristalina, Goiás, have conspicuous globose fistulae but the leaves, flowers and inflorescence are otherwise very similar to other collections of *E. macrosiphon*.

Hyptis mattogrossensis was described by Pilger from a specimen collected from Cuiabá in Mato Grosso and was placed in synonymy by Epling (1936) Although the type specimen of

H. mattogrossensis has not been seen it was collected in an area which has produced many collections of *E. macrosiphon* and it has been retained in synonymy.

Harley 20424 is a sterile collection from Chapada de Guimarães. The shape of the leaf is similar to other collections from the area but the identification cannot be confirmed in the absence of the inflorescence.

14. Eriope paniculata *(Benth.) R. Atkinson* comb. nov.

Type: Brazil, loc. incert. Sellow 1499 (holotype B, not seen) (Sellow s.n., probable isotype K).

Hyptis paniculata Benth., Labiat.Gen. Spec.: 139 (1833); Bentham in DC. Prodr.12: 138 (1848); Schmidt in Fl. Bras. 8(1): 155 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 233 (1935); Epling in Revista Mus. La Plata, secc. Bot. 7: 238 (1949).
Mesosphaerum paniculatum (Benth.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hypenia paniculata (Benth.) Harley in Bot. J. Linn. Soc. 98: 92 (1988).
Hyptis coccinea Mart. ex Benth., Labiat.Gen. Spec.: 139 (1833); Bentham in DC. Prodr.12: 138 (1848); Schmidt in Fl. Bras. 8(1): 155 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 233 (1935). Type: Brazil, Serra do Sincorá, Bahia, *Martius* s.n. 1818 (holotype M).
Mesosphaerum coccineum (Benth.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hyptis macrantha St.Hil. ex Benth. sensu Epling & Mathias in Brittonia 8: 298 (1957) **pro parte.**

Woody herb to 1.5 m. *Stems* erect, unbranched with long, virgate axes, slender; basal parts pubescent with simple hairs and setose hairs; upper parts drying dark brown, terete, glabrous, waxy; internodes fistulose, sometimes slightly swollen. *Leaves* petiolate, erect, frequently caducous, distant; lamina oval to ovate, 30 — 60 x 5 — 40 mm; base truncate or cordate; apex rounded or acute; margin crenate or serrate, regular; upper surface sparsely to moderately pubescent with short simple hairs and minute subsessile glands; lower surface tomentose with lanate hairs; petiole 10 — 20 mm, glabrous or pubescent with short simple and setose hairs. *Inflorescence* a regular, open, indeterminate thyrse, few-flowered; phyllomes caducous; branches few, spreading, 2 per node, very regular, glabrous, waxy; cymes 1-flowered, widely spaced 5 — 60 mm apart; peduncle bracts lanceolate, 1.5 — 3.5 mm long; peduncle erect, 3 — 15 mm, glabrous with tuft of lanate hairs at distal end or pubescent with lanate hairs along length; paired bracteoles erect, narrowly lanceolate, 2 —

2.5 x 0.5 — 0.75 mm. *Flowering calyx* vinaceous, erect, gibbous-campanulate, 6 — 10 mm, moderately pubescent with conspicuous and persistent long contorted hairs and glandular hairs; teeth symmetrical, narrowly deltoid -rostrate, 2 — 3 mm. *Fruiting calyx* erect, cylindrical, 9 — 13 mm; teeth 2 — 3 mm; throat with moderately dense long hairs. *Corolla* tube red, narrowly cylindrical, straight, 14 — 15 mm; posterior and lateral lobes red, ca. 4 mm; anterior lobe red, ca. 3 mm. *Stamen* filaments with dense hairs. *Nutlets* ovoid in section, ca. 4 x 2.5 mm.

DISTRIBUTION. Brazil, Serra do Cabral to Grão Mogol, northern Minas Gerais (figure 10-11).

HABITAT. Upland savanna (campo rupestre) and savanna (cerrado).

Altitude: 600 —1200 m.

BRAZIL.

Loc. incert., s. dat., *Sellow* 565 (P).

BAHIA. Serra do Sincorá, 1818, *Martius* s. n. (M).

MINAS GERAIS. Buenópolis: Serra do Cabral, 27 July 1976, *Davis* et al. UEC no. 2343 (E, UEC); Buenópolis: Serra do Cabral, ca. 10 km from town, road to Lapa Pintada, 13 Oct. 1988, *Harley* et al. 24934 (SPF); Diamantina: June 1964, *Brade* 13645 (RB); Grão Mogol: Mandacarú - Grão Mogol road, 20 June 1985, *Martinelli* et al. 11202 (RB); Joaquim Felício: Serra do Cabral - Bocaina, 5 July 1985, *Cerati* et al. 205 (K); Joaquim Felício: Serra do Cabral, road from Joaquim Felício to Pirapora, 28 July 1976, *Davis* et al. UEC no. 2502 (E, UEC); Joaquim Felício: road to Serra do Cabral, 17 May 1981, *Rossi* et al. CFCR 1161 (SPF, K); Joaquim Felício: Serra do Cabral, 31 Aug. 1985, *Zappi* et al. CFCR 8105 (SPF); Lassance: Serra do Cabral, 9 Dec. 1919, *Lutz* 1586 (R); Montes Claros: ca. 30 km SE, road to Juramento, 15 May 1977, *Gibbs* et al. 5117 (SP); Serra do Cabral: 1898, *Schwacke* 13407 (G); Serra do Calixto: Sept. 1945, *Lanstyak* s.n., RB no. 55967 (RB).

E. paniculata is close to *E. reticulata* morphologically and geographically and is separated by a hiatus scarcely greater than that between morpho-types within *E. reticulata.* However, it has well-defined, consistent characters which are not found elsewhere in the subsection, notably the long, hairs on the calyx. It is separated on the basis of the long, narrow calyx teeth, long hairs on the floral parts, short, erect peduncles and by the exceptionally regular inflorescence. It has a restricted distribution in northern Minas Gerais (figure 10-11).

The specimen at Kew, *Sellow s.n.*, is without locality details and possibly represents a duplicate of *Sellow 1449.* It could therefore be an isotype. The specimen is in rather poor condition but the floral parts that remain and the one mis-shapen leaf have the characteristic indumentum of other collections cited. The holotype from Berlin has not been seen and is

possibly destroyed. Sellow 565 is a complete fertile specimen and is a good example of *E. paniculata* as described here. Epling in his 1949 monograph cites *Glaziou 21957* as the only other collection for *E. paniculata* apart from the type, but I have cited *Glaziou 21957* as the type of *E. indaiaensis.*

Epling (1949) cites *Hyptis coccinea* as a synonym of *H. gardneriana.* The type of *H. coccinea, Martius* s. n. from Serra do Sincorá in Bahia is identical to specimens of *E. paniculata* cited from the Serra do Cabral. As a result I have cited *H. coccinea* as a synonym of *E. paniculata.*

15. Eriope recoria *R. Atkinson* sp. nov.

Type: Brazil: Distrito Federal, Brasília: Jardim Botânico, ca. 1 km S of administration buildings, 8 Aug. 1995, *Atkinson & Giorgio* 147 (holotype IBGE, isotypes K, UB).

E. reticulata affinis sed inflorescentiis multifloribus, flores majores, bracteolis ovatus et foliis majores, oblongus-ellipticus differt.

E. recoria can be distinguished from *E. reticulata* by its floriferous inflorescence with relatively large flowers, ovate bracteoles and large, oblong-elliptic leaves.

Woody herb to 3 m. *Stems* erect, unbranched with long, virgate axes, moderately robust; basal parts sparsely pubescent with simple hairs, sometimes with setose hairs; upper parts dark reddish-brown or glaucous green, terete, glabrous, waxy; internodes not fistulose or swollen. *Leaves* petiolate, spreading, persistent, distant or overlapping; lamina oblong-elliptic, 53 — 165 x 11 — 60 mm; base cuneate or truncate; apex mucronate; margin finely to coarsely serrate; upper surface sparsely pubescent with short simple hairs; lower surface sparsely pubescent to tomentose with simple hairs and lanate hairs; petiole 10 — 55 mm, glabrous or with sparse simple hairs. *Inflorescence* a large, open indeterminate thyrse, floriferous; phyllomes narrowly lanceolate, frequently caducous; branches spreading, straight, 2 per node, regularly developed, glabrous and waxy; cymes 1-flowered, 4 — 40 mm apart along branches, sometimes in the branch axils; peduncle bracts broadly lanceolate to ovate, 1.5 — 2.5 mm; peduncle 4 — 25 mm, sparsely pubescent with simple and glandular hairs; paired bracteoles deflexed, ovate, 1.5 — 2.5 x 1 — 2 mm. *Flowering calyx* vinaceous, horizontal, gibbous-campanulate, 5 — 7 mm, pubescent with simple and glandular hairs;

teeth symmetrical, 1.5 — 2 mm, broadly deltoid. *Fruiting calyx* erect, cylindrical, 9 — 11 mm; throat with sparse long hairs. *Corolla* tube salmon-pink to red, cylindrical, straight, not deeply channelled, 9 — 18 mm; throat marked yellow; posterior and lateral lobes salmon-pink to red, 5 — 8 mm; anterior lobe salmon-pink to red with yellow mark on retracted apex, 3 — 5 mm. *Stamen* filaments pink with white hairs; anthers yellow. *Style* pink. *Nutlets* ovoid in section, ca. 3.5 x 2 mm. Figure 10-4.

DISTRIBUTION. In Distrito Federal and neighbouring parts of Goiás (figure 10-11).
HABITAT. Savanna (cerrado), apparently restricted to the boundary with gallery forest. Altitude: 1000 — 1240 m.

BRAZIL

DISTRITO FEDERAL. Brasília: Jardim Botânico, ca. 1 km S of administration buildings, 8 Aug. 1995, *Atkinson & Giorgio* 147 (IBGE, K, UB); Brasília: road to agricultural school, 9 Sept. 1978, *Heringer* et al. 566 (HEPH, K, NY, UEC); Brasília: basin of the Rio São Bartolomeu, 8 August 1979, *Heringer* et al.1931 (K, NY); Brasília: basin of the Rio São Bartolomeu, 3 Apri l980, *Heringer* et al.4226 (K, UEC); Brasília: Fazenda da Sucupira, 21 July 1975, *Heringer* 14777 (K, NY); Brasília: Cabeça do Veado stream, IBGE Ecological Reserve, 21 Sept. 1982, *Kirkbride Jr.* 4908 (K, NY); Brasília: country club, 7 Sept. 1968, *Lima* 80 (K); Brasília: Fazenda Água Limpa, University of Brasília experimental farm, ca. 25 km SSE of Brasília, 3 May 1968, *Philcox & Onishi* 4891 (K); Brasília: Fazenda Água Limpa, University of Brasília experimental farm, ca. 25 km SSE of Brasília, *Ratter* et al.2918 (E, K); Brasília: Fazenda Água Limpa, University of Brasília experimental farm, ca. 25 km SSE of Brasília, *Ratter* et al.3124 (E, K).
GOIÁS. Luziânia: industrial town, 30 April, 1975, *Heringer* 14605 (K, NY, UEC); Pirenópolis: Caixa d'Água hill, 23 April 1976, *Heringer* 15566 (K, NY, SP); Posse: 6 km from the town on the road to Duzin, old road to Alvorada, 31 March 1997, *Harley* et al. 28557 (K, UEFS).

E. recoria has a large, floriferous inflorescence and broad, persistent leaves which distinguish it from the other species from subsection *Laxiflorae* in Distrito Federal, *E.reticulata,* and all other species in the subsection. The ovate, deflexed bracteoles are conspicuous on herbarium sheets and make this species readily identifiable. It has long corolla tubes which are always red (cf. short pink, red, or orange corolla of *E. reticulata*).

E. recoria has an unusual habitat preference for the margin of gallery forest which it shares with *E. densiflora.* The two have been observed growing together in the Botanic Garden, Brasília, with *E. reticulata* growing in neighbouring savanna (figures 10-10 and 10-11). The specific epithet is derived from the acronym RECOR, which signifies the Reserva Ecológica do IBGE, the type locality.

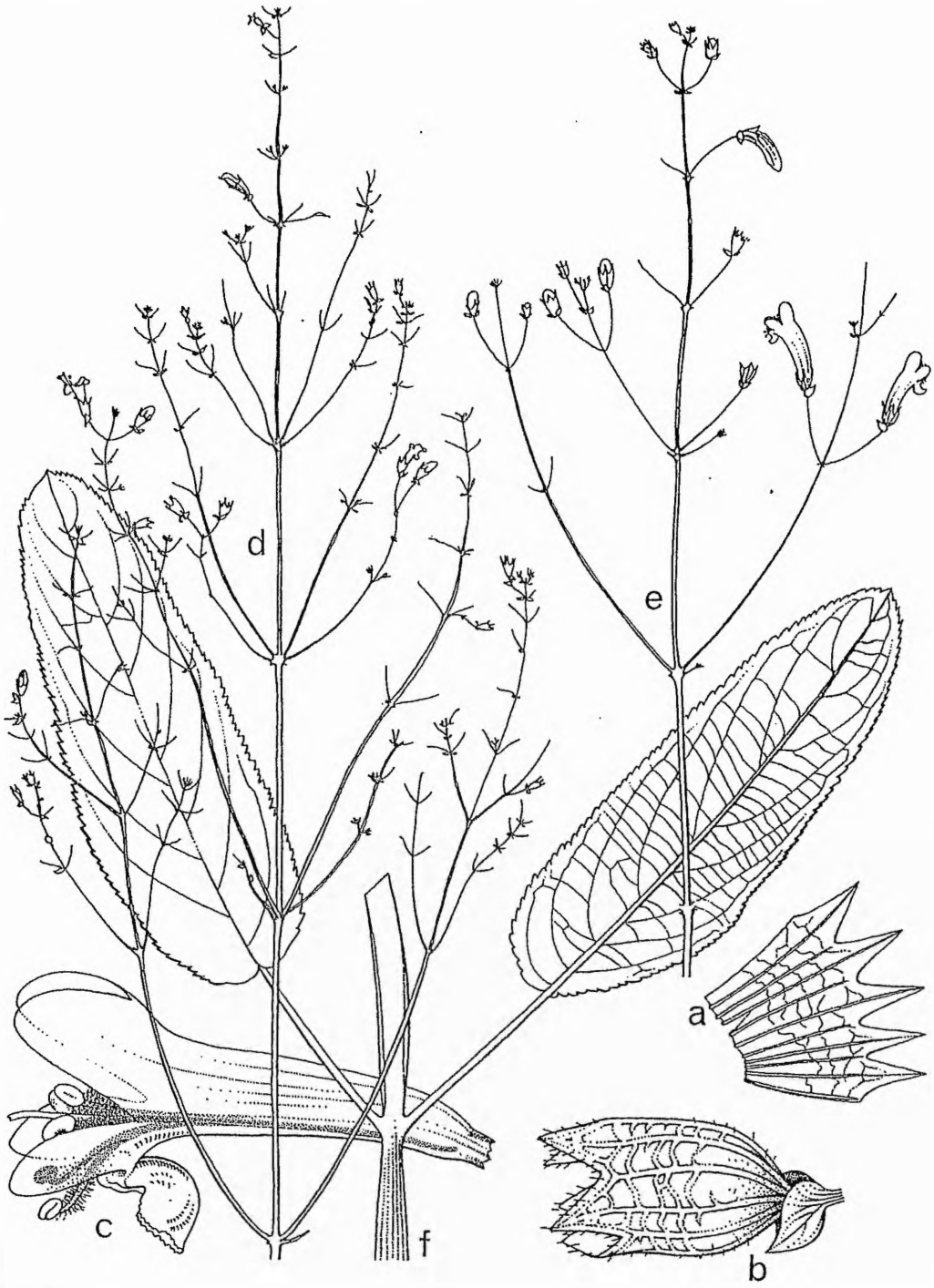


Figure 10-4: ***Eriope recoria***

a) flowering calyx (x4) (*Heringer* 566); fruiting calyx (x4) (*Heringer* 14777); c) corolla (x4) (*Heringer* 566); d) inflorescence (x½) (*Heringer* 566); e) inflorescence (x½) (*Ratter* et al. 3124); f) stem and leaves (x½) (*Heringer* 566). Drawn by Emmanuel Papodopoulos.

16. Eriope reticulata *(Mart. ex Benth.) R. Atkinson* comb. nov.

Type: Brazil, inter Rio do Peixe et Rio Verde, in via prob. São João del Rey, Minas Geraes, Feb. 1818, *Martius* 713 (holotype M).

Hyptis reticulata Mart. ex Benth., Labiat.Gen. Spec.: 141 (1833); Bentham in DC. Prodr.12: 139 (1848); Schmidt in Fl. Bras. 8(1): 154 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 232 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 236 (1949).
Hypenia reticulata (Mart. ex Benth.) Harley in Bot. J. Linn. Soc. 98: 92 (1988).
Hyptis longiflora Pohl ex Benth., Labiat.Gen. Spec.: 140 (1833); Benth. in DC. Prodr.12: 139 (1848); Type: Brazil, a Barbacena ad Paracatu, Minas Geraes, *Pohl* 440 (holotype W, isotype K).
Mesosphaerum longiflorum (Pohl) Kunth in *Revis.* Gen. Pl. 2: 526.
Hyptis laxiflora Mart. ex Benth., Labiat.Gen. Spec.: 139 (1833); Benth. in DC. Prodr.12: 138 (1848); Type: Brazil, in Campis montis ad Villa Rica, Minas Geraes, Martius (holotype M).
Hyptis glauca St. Hil. ex Benth., Labiat.Gen. Spec.: 141 (1833); Bentham in DC. *Prodr* 12: 139 (1848); Schmidt in Fl. Bras. 8(1): 155 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih., 85: 232 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 233 (1949) ; **synon. nov.** Type: Brazil, Caxambú, St. Paul, 1816 - 1821, *St. Hilaire* 1447 (holotype P).
Mesosphaerum glaucum (St. Hil. ex Benth.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hypenia glauca (St. Hil. ex Benth.) Harley in Bot. J. Linn. Soc. 98: 92 (1988).
Hyptis glauca var. *gardneriana* Benth. in DC. Prodr. 140 (1848); Type: Brazil, near Mission Duro, Goyaz, *Gardner* 3391 (holotype K, isotypes G, BM, NY, P).
Hyptis gardneriana (Benth. in DC.) Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 233; Epling in Revista Mus. La Plata, secc. Bot. 7: 236 (1949).
Hyptis perplexa Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 231 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 232 (1949) ; **synon. nov.** Type: Brazil, Diamantina, Olaria, Minas Gerais, 8 May 1931, *Mexia* 5796 (holotype UC, isotypes K, G, S).
Hypenia perplexa (Epling) Harley in Bot. J. Linn. Soc. 98: 92 (1988).
Hyptis pauliana Epling in Revista Mus. La Plata, secc. Bot. 7: 236 (1949) ; **synon. nov.** Type: Brazil, between the Riberão de Veravinha and Villa Franca, São Paulo, 1828, *Burchell* 5522 (holotype K).
Hypenia pauliana (Epling) Harley in Bot. J. Linn. Soc. 98: 92 (1988).

Hyptis macrantha St.Hil.ex Benth. sensu Epling & Mathias in Brittonia 8: 298 (1957) **pro parte.**

Woody herb to 2.5 m. *Stems* erect, unbranched or branched below long, virgate axes, moderately robust or slender; basal parts pubescent with simple hairs, sometimes glandular hairs and setose hairs; upper parts glaucous-green, drying pinkish-brown, glabrous, waxy; internodes fistulose, sometimes swollen. *Leaves* petiolate, erect, overlapping, or distant; lamina ovate, ovate-lanceolate, lanceolate or oblong, 8 — 120 x 5 — 55 mm; base cuneate or truncate or shallowly cordate; apex obtuse or acute, sometimes asymmetric; margin crenate or serrate, regular; upper surface sparsely pubescent with simple hairs and subsessile glands; lower surface sparsely or moderately pubescent with simple hairs and subsessile glands, or tomentose with simple and lanate hairs and glandular hairs; petiole 2 — 40 mm, pubescent with simple hairs or glabrous and waxy. *Inflorescence* a regular, open, indeterminate thyrse, moderately floriferous; phyllomes lanceolate, frequently caducous; branches erect, straight, 2 per node, regularly developed, glabrous and waxy, sometimes upper branch pubescent with glandular hairs and minutely gland-tipped hairs; cymes 1-flowered, regularly spaced 4 — 50 mm apart; peduncle erect or deflexed, (3) —10 — 27 mm, glabrous and waxy, sometimes pubescent with glandular and minutely gland-tipped hairs; peduncle bracts lanceolate, 1.5 — 3 mm; paired bracteoles, narrowly lanceolate, 1.5 — 2.5 x 0.5 — 0.75 mm. *Flowering calyx* vinaceous, gibbous-campanulate, 4 — 8 mm, pubescent with simple hairs, subsessile glands and minutely gland-tipped hairs; teeth symmetrical, broadly deltoid -rostrate, (0.5) —2 — 3 mm. *Fruiting calyx* broadly cylindrical, 8 — 13 mm; teeth broadly deltoid, 2 — 3 mm; throat with dense long hairs or sometimes +/- glabrous. *Corolla* tube red, pink or apricot, straight, 8 — 15 mm; posterior lobes red, 4 — 7 mm; lateral lobes red, pink or apricot, spreading, 3 — 6 mm; anterior lobe red, carmine or apricot with yellow or cream mark on retracted apex, ca. 3 — 4 mm long. *Stamen* filaments pink with dense white hairs, anthers yellow. *Style* pink. *Nutlets* ovoid in section, 2.5 — 4 x 1.5 — 2.5 mm.

DISTRIBUTION. Brazil, southern and central Brazil, including the states of Paraná, São Paulo, the southern half of Minas Gerais, Distrito Federal and neighbouring parts of Goiás (figure 10-10).

HABITAT. Savanna (cerrado) and upland savanna (campo rupestre).

Altitude: 550 — 1570 m

BRAZIL.

s.loc.1816 -1821, *St.Hilaire* s.n. (P); s. loc., s. d, *Sellow* s.n. (P).

BAHIA. Rio de Contas, Nov. 1818, *Martius* s.n. (M).

DISTRITO FEDERAL. Brasília: Cabeça do Veado, to Papuda, 2 Sept. 1960, *Andrade & Emmerich* 322 (K); Brasília: Reserva Ecológica do IBGE, 3 Sept. 1995, *Atkinson & Giorgio* 145 (K, IBGE, UB); Brasília: Fazenda Água Limpa, University of Brasília Experimental Farm, 1 Aug. 1995 *Atkinson & Giorgio* 154 (K, IBGE, UB); Brasília: area of Cristo Redentor, Reserva Ecológica do IBGE, 16 Aug. 1988, *Azevedo* 90 (K); Brasília: Jardim Botânico, 20 Sept. 1988, *Azevêdo* 137 (K); Brasília: Fazenda Água Limpa, *França & Mello* UPCB 20219 (NY); Brasília: Parque Naçional, 26 July 1980, *Guimarães* 1080 (RB); Brasília: forestry road, Cabeça do Veado, , 22 July 1975, *Heringer* 14826 (K); Brasília: University, 23 Aug. 1965, *Irwin* et al. 7845 (K); Brasília: ca. 20 km NE, Chapada de Contagem, 5 Sept. 1965, *Irwin* et al. 7987 (K); Brasília: Mirante Sobradinho, near Jockey Club, 16 Aug. 1963, *Maguire* et al.56200 (K); Brasília: area of Cristo Redentor, Reserva Ecológica do IBGE, 16 Aug. 1988, *Mendonça* 1043 (K); Brasília: Reserva Ecológica do IBGE, 26 June 1989, *Pereira* 1376 (K); Planaltina: 22 km W, CPAC cerrado reserve, 20 July 1980, *Mori* et al. 16903 (K, NY).

GOIÁS. Caldas Novas: Serra de Caldas, 9 July 1976, *Hatschbach* 38749 (K, NY, UC, MBM); Catalão: ca. 25 km NE, Contraforte Central, 21 Jan. 1970, *Irwin* et al. 25021 (K, G); Morrinhos: 2 July 1951, *Macêdo* 3229 (UC); Serra das Araras, Goyaz, June 1840, *Gardner* 5080 (K, BM).

MINAS GERAIS. s. loc. 1833, *Claussen* s.n. (G); s. loc. 14 Oct. 1927, *Jarneby* (S); s. loc. June 1824 *Reidel* 237 (NY); s. loc., 1833, *Vauthier* 416 (G, P); Abaité: 1844, *Weddell* 1781 (G, P); Belo Horizonte: 1903, *Damazio* 978 (G); Belo Horizonte: Pampulha, 17 Aug. 1958, *Pabst* 4530 (K); Belo Horizonte: Bento Pires, 30 May 1945, *Williams & Assis* 7108 (UC); Belo Horizonte:10 km E, 4 July 1945, *Williams & Assis* 8105 (UC); Betim: Serra da Mutuca above Barreiros, 3 June 1945, *Williams & Assis* 7282 (NY, UC); Caldas: 9 Jan. 1919, *Hoehne* 2734 (UC); Caldas, Nov. 1854, *Lindberg* 146 (S); Caldas: 20 Sept.1875, *Mosén* 672 (UC); Caldas: 1861, *Regnell* 316 (K, NY, S, P); Cambuquira: 17 km E on Highway 267 to Caxambu, 26 Feb. 1976, *Davidse & Ramamoorthy* 10671 (K, SP); Carandaí: Palmeira 5 Nov. 1952, *Duarte* 4277 (UC, RB); Caxambu: St Paul, 1816 -1821, *St.Hilaire* 1447 (P); Corandai-Crespo: 15 Nov. 1946, *Duarte* 474 (UC, RB); Corinto: ca. 20km W, Rio Bicud, 3 March 1970, *Irwin* 26799 (K); Couto de Magalhães: Fazenda das Abóboras, 16 July 1984, *Harley* et al. CFCR 4586 (K, SP); Couto de Magalhães: Chapada do Couto, 17 July 1984, *Harley* et al. CFCR 4612 (K, SPF, SP, MBM); Curvalinho: next to bridge, 23 March 1978, *Menezes* 812 (K); Diamantina: 10 km SW of Rio Jequití & Mendonha, 15 April 1973, *Anderson* 8945 (K, NY, SP); Diamantina: June 1934 *Brade* 13644 (UC); Diamantina: ca. 7km NE, 29 Jan. 1969, *Irwin* et al. 22822 (K); Diamantina: ca.18km E, 14 March 1970, *Irwin* et al. 27483 (K, NY); Diamantina: road to Curvelo, 17 km from Diamantina, 1 Nov. 1981 *Menezes* et al. CFCR 2629 (K, SPF); Diamantina: Olaria, 8 May 1931, *Mexia* 5796 (K, BM, G, NY, UC, S, P); Diamantina: 20 May 1955, *Pereira* 1370 (UC); Diamantina: road to Conselheiro da Mata, ca 4km from Diamantina - Belo Horizonte road, 19 Nov. 1984, *Stannard* et al. CFCR 6199 (K, SPF); Ituiutaba: 3 Sept. 1944, *Macêdo* 82 (NY, UC, S); Jaboticatubas: 10 km N of Lagoa Santa, 28 April 1952, *Smith* et al. 6972 (NY, UC, S); Jandarela: 13 July 1972, *Emygdio* et al. 3293 (NY); Jandarela: 15 July 1972, *Emygdio* 3378 (NY); Lagoa Santa: near airfield, 10 Sept. 1945, *Williams & Assis* 7437 (UC); Magalhães: Usina, near dam, 12 July 1973, *Menezes* 4250 (SP, UEC); Nova Lima: Pico de Belo Horizonte, 6 June 1945, *Williams & Assis* 7162 (UC); Ouro Preto: 1857, *Casaretto* 2914 (G); Paracatu: on way to Barbacena, Nov. - Dec. 1818, *Pohl* 440 (K†, W); Paroapeba: s.nom. 227 (K); Patrocínio: 12 km N, 31 Jan. 1970, *Irwin* et al. 25758 (K); Poços de Caldas: Véu das Noivas, 19 Jan. 1980, *Krapovickas* 35382 (K); Retiro de Bras, mountains in western parts of Minas Geraes, 1816 - 1821, *St.Hilaire* 439 (P); Santana do Riacho: Serra do Cipó, 13 April 1935, *Brade* 14458 (UC); Santana do Riacho: Serra do Cipó, 23 July 1905, *Damazio* 34 (UC); Santana do Riacho: km 108 Belo

Horizonte to Conceição do Mato Dentro, 1 July 1981, *Giulietti* et al. CFSC 7416 (SPF, K); Santana do Riacho: km 144 Lagoa Santa to Conceição do Mato Dentro, 5 June 1970, *Joly* et al. 34 (E, NY, SP); Santana do Riacho: km 116, Lagoa Santa to Conceição do Mato Dentro, 6 May 1970, *Joly* et al. 187 (SP); Santana do Riacho: near source of Veú da Noiva, s.dat., *Simão* et al. CFSC 10462 (SPF); São João del Rey: between Rio do Peixe and Rio Verde, Feb. 1818, *Martius* 713 (M); São Sebastião do Paraiso: 18 April 1945, *Brade* et al. 17731 (UC); São Tomé das Letras: Baepundi, 13 July 1950, *Brade & Duarte* 20451 (UC); São Tomé das Letras: 5 Feb. 1973, *Hatschbach* 31295 (K, NY,MBM); São Tomé das Letras: 3 Nov. 1984, *Pirani* et al. CFCR 5847 (SPF); Serra do Cabral, 1898, *Schwacke* 13407 (G); Serra do Curral, BR 3 Km 420, 18 June 1964, *Duarte* 8086 (K, RB); Serra do Palmital, 20 May 1884, *Glaziou* 15314 (G, R, P); *Widgren* s.n. (M, S, P).

Additional citations from: Santana do Riacho: Belo Horizonte to Conceição do Mato Dentro. *Chukr* et al. CFSC 9827; *Cordeiro* 6112; *Diacui* et al. CFSC 8595; *Forero* 7841; *Forero* 8090; *Freire-Fierro & Esteves* CFSC 11838 (SPF); *Giulietti* et al. CFSC 5681; Santana do Riacho: km 116 Belo Horizonte to Conceição do Mato Dentro, 6 June 1970, *Joly* et al. 144 (SP); *Joly* et al. 1745; *Joly* et al.; *Menezes* 688; *Pirani* et al. CFSC 12396; *Pirani* et al. CFSC 6158; *Semir* et al. 2780; *Semir* et al. 2829; *Zappi* et al. CFSC 9331. São Tomé das Letras. *Hatschbach & Anderson* 36552; *Monteiro* 31; *Monteiro* 214; *Mello Silva* et al. CFCR 5681.

PARANÁ. Arapoti: Chapadão São Antonio, 11 Oct. 1968, *Hatschbach* 19997 (UC, MBM); Capão Bonito: 16 May 1914, *Dusén* 15037 (BM, S); Itararé: 26 April 1911, *Dusén* 11644 (NY, S); Jaguariaíva: 26 Oct. 1910, *Dusén* 10515 (E, K, G, S); Jaguariaíva: 8 Dec. 1910, *Dusén* 11009 (S); Jaguariaíva: 23 Oct. 1911, *Dusén* 13261 (S); Jaguariaíva: 19 Nov. 1914, *Dusén* 16035 (S); Jaguariaíva: Rio das Mortes, 5 Dec. 1965, *Hatschbach* 11985 (MBM); Jaguariaíva: Rio das Mortes 27 May 1975, *Hatschbach* 39955 (K, MBM); Jaguariaíva: 6 Nov. 1928, *Hoehne* 23438 (UC); Jaguariaíva: 11 May 1914, *Jönsson* 310a (S); Jaguariaíva: Rio das Mortes, 18 Dec.1965, *Reitz & Klein* 17933, (K); Jaguariaíva: 17 Jan. 1965, *Smith* et al. 14682 (NY); Piraguara: s.dat. *Dombrounfi* 10036 (K); Senges: Rio Velame, 17 June 1971, *Hatschbach* 26801 (K, NY, UC, S); Tibagí: Fazenda Monte-Alegre, Harmonia, 16 Sept. 1952, *Hatschbach* 2781 (UC); Tibagí: road from Castro, Rio Tibagí, 1 Nov. 1964, *Hatschbach* 11807 (K, MBM); Tibagí: Guartelá, canyon of Rio Iapó 17 March 1991, *Hatschbach & Barbosa* 58214 (MBM); Tibagí: 8 Nov. 1935, *Reiss* 146 (NY); Tibagí: Guartelá canyon 17 March 1991, *Ziller* 196 (MBM).

SÃO PAULO. S. loc., 1861/2, *Weir* s.n. (K); s. loc., 1836, Sellow s.n. (E, K); loc. incert., between the Ribeirão de Veravinha and Villa Franca, 1828, *Burchell* 5522 (K); Batatais: between Araracoara and Batatais, 5 June 1834, *Reidel & Lund* 2333 (NY); Botucati: 18 km N & 14 km W of São Miguel, 24 Oct. 1974, *Gottsberger & Campos* 129-24107 (UB); Buri: Fazenda Ipuaçu, 17 July 1983, *Marcondes* et al. 14775 (UEC); Campinas: 7 Nov.1904, *Heiner* 303 (S); Conchal: Conchal - Padua Sales road, 3 Dec. 1961, *Eiten* 3508 (SP); Conchal: Conchal - Padua Sales road, 3 Dec. 1961, *Eiten* 3504 (SP); Franca: 28 March 1997, *Harley* et al. 28499 (K, UEFS); Itararé: 1 km SE, 29 Oct. 1965, *Mattos & Moura* 14989 (SP); Itirapina: road to the island, 2 Nov. 1990, *Joly* 1203 (SPF); Itirapina: Itirapina - Rio Claro road, 2 May 1962, *Labouriau* 1058 (RB); Itiripina: 14 Feb. 1975, *Leitão Filho* 1470 (UEC); Itirapina: 22 May 1965, *Paula* 109 (SP); João Simão: Fazenda Aritozina, 29 Jan 1982, *Leitão Filho* et al. 13269 (UEC); Itirapina: 25 Sept. 1940, *Toledo & Gehrt* 43220 (K, SP); Itiripina: 8 July 1987 *Válio* et al. 19255 (UEC); Itupeva: between Orisanga and Itupeva, 20 Aug. 1828, *Burchell* 5212 (K); Jales: 29 Oct. 1951, *Hoehne* 3927 (SPF); Moji Mirim: 15 Aug. 1828, *Burchell* 5151 (K); Moji Mirim: 15 Nov. 1901, *Hammar* 36 (UC); Moji Mirim: 21 May 1927, *Hoehne* 20438 (UC); Pirassununga: 25 Sept. 1946, *Pickel* 2722 (UC); Porto Ferreira: 22 April 1954, *Hoehne & Wasicky* 15339 (SPF); Rincão: 25 Jan. 1928, *Toledo* 22876 (SP); Santa Rita do Passa Quatro: Parque Estadual de Vaçunuga, July 1985, *Castro* 19736 (UEC); São Carlos: km 157 on road to Ribeirão Preto, 20 April 1960, *Campos & Felipe* 216 (K, SP); São Carlos: Mogi-Guaçu, Fazenda Campininha, 11

Feb. 1960, *Eiten* 1720 (NY, SP); São Carlos: Ibaté, ca.1km NE. (12.8km NE of centre of São Carlos), 14 June 1961, *Eiten* et al. 2926 (K, SP); São Carlos: 15km NNW of village of Santa Eudoxia, 3.5km W of Rio Moji-Guaçu, 20 June1961, *Eiten* et al. 3079 (K, SP); São Carlos: near border with mun. de Araraquara, 20 June 1961, *Eiten* et al. 3082 (SP); São Carlos: 8km N on road to Água Vermelha, 3 Oct. 1961, *Freitas Campos* 83 (P); São Carlos: district of Santa Eudóxia, Rio Mogi-Guaçu, 28 March 1962, *Labouriau* 51 (SP); São Carlos: 8 May 1940, *Viegans* 44369 (SP); São Paulo: 22 April 1954, *Wasicky* 15339 (K, SPF); Vassununga: 30 June 1978, *Moretes* 19710 (SPF).

Additional citations from: São Carlos: Mogi-Guaçu, Fazenda Campininha, *Custodio Filho* 213, 419, 432, 434; *Eiten* 3556; *Figueiredo-Ribeiro* 196112; *Forero* et al. 8169; *Gibbs & Leitão Filho* 3395; *Handro* 437; *Hoehne* 6124; *Kirizawa* 122; *Leitão Filho* et al. 6085; *Mantovani* 181, 736, 821, 1114, 1192, 1338, 1848; *Mantovani & Sugyiama* 1797; *Mattos & Mattos* 8246, 8283, 8307, 8442; *de Paulo* 188; *Sakane* 689; *Sugyiama & Mantovani* 133; *Sugiyama & Kirizawa* 425; *Tamashiro* et al. 17683.

TOCANTINS.Mission of Duro, Goyaz (= Tocantins), Sept. 1839, *Gardner* 3391 (K, BM, G, NY, P).

† one sheet at K of this collection also includes leaves from *E. calycina*.

E. reticulata is the most widespread and morphologically variable species in section *Hypenia*.It is characterised by its diffuse, more or less regular inflorescence, glabrous and usually waxy inflorescence branches and ovate to lanceolate leaves with crenate to serrate margins. However, all these characters are variable, especially the leaves which vary in size, shape and indumentum and the arrangement of the cymes in the inflorescence which can cause considerable divergence in overall appearance of specimens of this species from different localities. Morphological characters are more or less correlated with geographical distribution but the discontinuities are not sufficently defined to recognise different species (see chapter 9 and figure 10-10).

Gardner 3391, the type of *H. gardneriana,* here sunk into synonymy, is the most northerly record for any species in section *Hypenia,* and is very isolated from the main distribution of *E. reticulata.* However, northeastern Goiás, Tocantins and western Bahia are very poorly known regions botanically and further collections in these areas are likely to extend the range of *E. reticulata* from its nearest locality in Distrito Federal. Collections cited as *H. gardneriana* by Epling have been placed under *E. sclerophylla* in this revision. The Martius collection from Rio de Contas in Bahia is another northern record but there is some doubt as to its veracity. The area of Rio de Contas has been intensively collected by Harley et al. (Stannard 1995) and I have also visited the area. *E. reticulata* has never been found by these recent visits. Habitat disturbance may account for its loss since Martius' time but *E. salzmannii* and *E. vitifolia* are still common in the area.

The other widespread, variable species in subsection *Laxiflorae* is *E. macrosiphon* but *E. reticulata* differs with its glabrous inflorescence branches, laxly-spaced cymes and unlobed leaves with a serrate or crenate, rather than dentate, margin. Some populations of *E. reticulata* occur in northern Minas Gerais, close to *E. paniculata,* but *E. reticulata* is distinguished by its sparsely pubescent calyx, long peduncles and laxly-spaced cymes.

17. Eriope subrosea *(Harley) R. Atkinson* comb. nov.

Type: Brazil, Chapada dos Veadeiros, ca. 19 km N of Alto Paraíso, Goiás, 20 March 1971, *Irwin* et al. 32783 (holotype K, isotypes NY, P, UB)

Hyptis subrosea Harley in Kew Bull. 29: 128 (1974).
Hypenia subrosea (Harley) Harley in Bot. J. Linn. Soc. 98: 92 (1988).

Woody herb to 1.25 m. *Stems* erect, unbranched with long, virgate axes, slender; basal parts pubescent with simple hairs and glandular hairs, no setose hairs; upper parts yellowish, drying dark brown, terete, glabrous and waxy; internodes fistulose with conspicuous globose swellings. *Leaves* petiolate, erect, persistent, overlapping; lamina lanceolate or obovate, 30 — 60 x 7 — 20 mm; base cuneate; apex obtuse or cuspidate; margin coarsely crenate, regular; upper surface moderately pubescent with simple hairs and glandular hairs; lower surface densely pubescent with simple hairs and subsessile glands; petiole 3 — 20 mm, densely pubescent with simple hairs and glandular hairs. *Inflorescence* a regular, open, indeterminate thyrse, moderately floriferous; phyllomes caducous; branches few, spreading, straight or slightly arched, 2, sometimes 4, per node, regularly developed, glabrous, waxy; cymes 1-flowered, regularly spaced 3 — 35 mm apart; peduncle bracts broadly lanceolate, 1.5 — 2 mm; peduncle erect, 3 — 5 mm, glabrous or sparsely pubescent with glandular hairs; paired bracteoles narrowly lanceolate, 1 — 3 x 0.5 — 0.75 mm. *Flowering calyx* vinaceous, erect, gibbous-campanulate, 4 — 6 mm, moderately pubescent with short simple and glandular hairs; teeth symmetrical, narrowly detoid, 1.5 — 2 mm. *Fruiting calyx* erect, gibbous-cylindrical, 9 — 11 mm; teeth ca. 2 mm; throat with moderately dense long hairs. *Corolla* tube white sometimes with violet spots in throat, cylindrical, straight, 9 — 11 mm; posterior and lateral lobes white, tinged pink or violet on inner surface, ca. 3 mm; anterior lobe white tinged pink or violet, ca. 3 mm. *Stamen* filaments white; anthers purple. *Style* white. *Nutlets* ovoid in section, ca. 3.5 x 2 mm.

DISTRIBUTION. Brazil, Goiás, endemic to the Chapada dos Veadeiros, near the town of Alto Paraíso (figure 10-11).

HABITAT. Upland savanna (campo rupestre). Altitude: 1000 — 1500 m.

BRAZIL.

GOIÁS. Alto Paraíso: Chapada dos Veadeiros, 20 Feb. 1975, *Hatschbach* et al. 36331 (K, MBM); Alto Paraíso: 2 - 5 km W on GO-327, 15 Oct. 1990, *Hatschbach* et al. 54596 (MBM); Alto Paraíso: ca. 15 km W, Chapada dos Veadeiros, 8 Feb. 1966, *Irwin* et al. 12310 (K, RB); Alto Paraíso: ca. 19 km N, Chapada dos Veadeiros, 20 March 1971, *Irwin* et al. 32783 (K, P, NY, UB); Alto Paraíso: road to Campo Belo, km 8, 28 Nov. 1976, *Shepherd* et al. 3696 (UEC).

E. subrosea is unique in subsection *Laxiflorae* for its white or violet corolla, lanceolate leaves more or less clustered at the base of the stem, calyx which is long relative to the size of the corolla and distinctly globose swellings. The corolla size and colour and dense subsessile glands on the underside of the leaf suggest the possibility of a close relationship with *E. densiflora.* However it is retained in section *Hypenia* because of its lax inflorescence and large flowers. It has a restricted distribution to the surroundings of Alto Paraíso in the Chapada dos Veadeiros in northern Goiás (figure 10-11). This area is particularly rich species from both sections *Densiflorae* and *Hypenia* and a taxon such as *E. subrosea* with intermediate characteristics of both sections could be of hybrid origin between *E. densiflora* and a species from subsection *Laxiflorae.*

Subsection Ellipticae *(Briq.) R. Atkinson* comb. nov.

18. Eriope calycina *(Pohl ex Benth.) R. Atkinson* comb. nov.

Type: Brazil, Goyaz, Serra dos Cristães, May-June 1818, *Pohl* 6161(holotype W, isotypes K, UC).

Hyptis calycina Pohl ex Benth., Labiat.Gen. Spec.: 140 (1833); Bentham in DC. Prodr.12: 138 (1848);Schmidt in *Fl.bras.* 8(1): 156 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 234 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 234 (1949).
Mesosphaerum calycinum (Pohl ex Benth.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hypenia calycina (Pohl ex Benth.) Harley in Bot. J. Linn. Soc. 98: 92 (1988).
Hyptis macrantha St.Hil. ex Benth. sensu Epling & Mathias in Brittonia 8: 298 (1957) **pro parte**.

Woody herb to 2.5 m. *Stems* unbranched with long, virgate axes, robust; basal parts sparsely pubescent with simple hairs, setose hairs absent; upper parts drying pinkish-brown to black, terete, glabrous, waxy; internodes fistulose, swollen along length. *Leaves* petiolate, spreading, caducous, distant; lamina oblong or ovate to obovate, 40 — 230 mm x 10 — 50 mm; apex rounded or acute; base cuneate; margin crenate or dentate, often irregular; upper surface moderately to densely pubescent with lanate and simple hairs and sparse glandular hairs; lower surface moderately to densely pubescent with lanate and simple hairs and sparse glandular hairs; petiole 10 — 60 mm, densely pubesent with lanate hairs or glabrous and waxy. *Inflorescence* a large open, indeterminate thyrse, floriferous or few-flowered; phyllomes lanceolate or broadly ovate, 4 — 10 x 1.5 — 6 mm, frequently caducous; branches erect, straight, 2 or 4 per node, regularly developed, glabrous and waxy; cymes 1(—3)-flowered, regularly spaced 4 — 20 mm apart, bracts broadly ovate 3 — 6 x 3 — 6 mm; peduncle erect, 5 — 20 mm, pubescent with minute gland-tipped hairs; paired bracteoles broadly ovate, 3 — 6 x 3 — 6 mm, overlapping to form a cup-shaped "epicalyx". *Flowering calyx* yellowish-green, horizontal to erect, campanulate, 8 — 14 mm, sparsely to densely pubescent with minute gland-tipped hairs; teeth 2 — 4 mm, symmetrical, broadly deltoid , spreading. *Fruiting calyx* yellowish-brown, erect, 10 — 15 mm, papery; teeth 3 — 5 mm, broadly deltoid and sometimes winged; throat with sparse long hairs. *Corolla* tube apricot or salmon-pink to red with yellow mark on apex of retracted lip, cylindrical, curved along length, 20 — 25 mm; posterior and lateral lobes apricot or salmon-pink to red, spreading and

rounded, ca. 8 mm; anterior lobe apricot or salmon-pink to red with yellow mark on retracted apex, 3 — 4 mm. *Stamen* filaments white; anthers yellow. *Style* pink. *Nutlets* ovoid in section, 3 — 4 x 2 — 3 mm.
Figure 10-5.

DISTRIBUTION. Brazil, northeastern Goiás (figure 10-12).
HABITAT. Savanna (cerrado). Altitude: 750 — 1200 m.

BRAZIL.
GOIÁS. Loc. incert., between Rio do Padre and Rio Preto, 1828, *Burchell* 7773 (K); loc. incert., Belle Vallée de Chico Costa, 10 Sept. 1895, *Glaziou* 21923* (K, P, G); loc. incert., Fazenda da Taboquinha, 20 June 1895, *Glaziou* 21924* (K, S, P, R); loc. incert. Serra dos Cristães, Goyaz, May-June 1819, *Pohl* 6161(W, K, UC); Água Fria: repeater station of Telebrasília de Roncador 12 June 1993, *Hatschbach* 59304 (MBM); Alto Paraíso: road to Nova Roma, Serra da Laranjeira, 13 June 1993, *Hatschbach* 59054 (MBM); Alto Paraíso: 2 km away, Chapada dos Veadeiros, 18 July 1964, *Prance & Silva* 58221 (K, UC); Alto Paraíso: 2 km away, Chapada dos Veadeiros, 18 July 1964, *Prance & Silva* 58222 (K); Cavalcante: between As Araras (= Arraias ?) and Rio Moquem (= Bagagem), 24 Sept. 1828, *Burchell,* 7836 (K); Colinas: road from Niquelândia, ca. 5 km N of road to São Luis do Tocantins, 23 July 1995, *Atkinson* et al. 142 (UB, IBGE, SPF, K); Niquelândia: ca. 15 km S, 22 Jan. 1972, *Irwin* et al. 34761 (NY); Niquelândia: Cravias, 24 July 1952, *Macêdo* 3625 (UC, S); Paracatú: between Barbaçena and Paracatú, Nov.-Dec. 1818, *Pohl* 4040 (K) (mixed collection, see *E. reticulata*); close to Lagoa Paraim, 24 May 1980, *Kirkbride* et al. 3472 (K, NY); São João d'Aliança: road to Fazenda Mata Serena, 22 July 1992, *Vaz* 994 (RB); São João d'Aliança: Hotel Atos, 20 July 1992, *Vaz* (RB); Villa Boa: 1839, *Vindol* (M).
MINAS GERAIS. Guarda-Mor: between Guarda-Mor and Riberão de São João, 1828, *Burchell*, 7597 (K).
* *Glaziou* 21923, 21924 are mixed collections with *E. sclerophylla*. Replicates from each herbarium are cited under *E. calycina* or *E. sclerophylla* according to their identification.

E. calycina is one of the most distinctive members of subsection *Ellipticae.* The flowers are clearly resupinate, even from herbarium sheets and the floriferous panicle and variably-coloured flowers are conspicuous. The most notable feature of *E. calycina* is the large overlapping bracteoles which form a kind of "epicalyx". The corolla is unusual in *Eriope* for being curved along its length, a character presumably associated with its pollinator.

The distribution of *E. calycina* is centred on the Chapada dos Veadeiros, an area very rich in the *Eriope* species covered in this account (figure 10-12). It seems to be the only resupinate-flowered species in the area but localities on the western side of Chapada dos Veadeiros are very close, and possibly overlap, with those of *E. niquelandiensis*. The two species are easily separated and *E. calycina* is readily distinguished by the above-mentioned characters.

Figure 10-5: ***Eriope calycina***

a) fruiting calyx (x3) (*Kirkbride* 3472); b) flower (x3) (*Kirkbride* 3472); c) inflorescence (x½) (*Pohl* s.n.); d) inflorescence (x½) (*Pohl* s. n.); e) leaf (x½) (*Burchell* 7773); f) stem and leaves (*Pohl* s.n.). Drawn by Emmanuel Papodopoulos.

19. Eriope crispata *(Pohl ex Benth.) R. Atkinson* comb. nov.

Type: Brazil, Goyaz, Serra Dourada, *Pohl* 1566 (holotype W).

Hyptis crispata Pohl ex Benth., Labiat.Gen. Spec.: 139 (1833); Bentham in DC. Prodr. 12: 139 (1848); Schmidt in Fl.Bras 8(1): 153 (1858); Epling in Repert. Spec. Nov. Regni Veg Beih.85: 234 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 235.
Mesosphaerum crispatum (Pohl ex Benth.) Kuntze, Revis. Gen. Pl. 2: 526 (1891).
Hypenia crispata (Pohl ex Benth.) Harley in Bot. J. Linn. Soc. 98: 92 (1988).
Hyptis macrantha St.Hil. ex Benth. sensu Epling & Mathias in Brittonia 8: 298 (1957) **pro parte.**

Woody herb to 2 m. *Stems* unbranched with long, virgate axes, robust; basal parts sparsely pubescent with simple and glandular hairs and setose hairs; upper parts drying dark brown with pale pinkish-brown bloom, terete to slightly angled, glabrous, waxy; internodes fistulose, sometimes slightly swollen. *Leaves* shortly petiolate, erect, persistent, distant; lamina broadly lanceolate, 60 — 150 mm x 15 — 50 mm; apex rounded or acute; base truncate or cordate, clasping stem; margin dentate, regular; upper surface sparsely to moderately pubsecent with long hairs and sparse small subsessile glands; lower surface moderately to densely pubescent with long hairs and sparse small subsessile glands; petiole 4 — 10 mm, sparse setose and short simple hairs. *Inflorescence* a large, open, indeterminate thyrse, floriferous; phyllomes lanceolate, 2 — 7 mm x 1 — 3 mm, caducous; branches erect, 4 per node, glabrous, irregularly developed, waxy; cymes 1 (or 2)-flowered, crowded, 2 — 40 mm apart; peduncle bracts lanceolate, 2 — 3 mm, frequently caducous; peduncle erect and +/- parallel with inflorescence branches, 4 — 10 mm, with minute gland-tipped hairs; paired bracteoles 1 — 2 mm, broadly lanceolate. *Flower* resupinate. *Flowering calyx* yellowish green, erect, narrowly cylindrical, 7 — 9 mm, sparsely to moderately pubescent with minute gland-tipped hairs; teeth symmetrical, broadly deltoid, 1.5 — 2 mm. *Fruiting calyx* erect, 13 — 16 mm; throat glabrous; teeth 2 to 3 mm. *Corolla* tube yellow, cylindrical, straight, sometimes curved, ca. 17 mm; posterior and lateral lobes yellow, ca. 5 mm, spreading; anterior lobe yellow, ca. 4 mm long. *Nutlets* ovoid in section, ca. 4 x 2.5 mm.

DISTRIBUTION. Brazil, Goiás, endemic to Serra Dourada. near Goiás Velha (figure 10-12).
HABITAT. Savanna (cerrado). Altitude: ca. 1000 m.

BRAZIL.

GOIÁS. S. loc., 1839, *Vindol* s.n. (M); loc. incert, between Goyaz and Cujaba, Nov. or Dec. 1844, *Weddell* s.n. (P); Goiás Velha: ca. 15 km S, Serra Dourada, 11 May 1973, *Anderson* 10061 (K, NY); Goiás Velha: Serra Dourada, 16 July 1964, *Duarte & Mattos* 8252 (K); Goiás Velha: 25 km from the town, near Serra Dourada, 8 Sept. 1976, *Gibbs* et al. 2885 (UEC); Serra Dourada, April 1819 *Pohl* 1566 (W); Serra Dourada, 1968, *Rizzo* 4148 (RB); Serra Dourada, 1968, *Rizzo* 4185 (RB); Serra Dourada, 25 Aug.1967, *Sidney* 286 (K).

E. crispata is placed in subsection *Ellipticae* because of its relatively large flowers which are held in a usually floriferous inflorescence and the inversion of the flower which can be observed in herbarium specimens. Observations in the field by Harley (see his photograph on plate I) indicate that this is a resupinate-flowered species.

E. crispata is closely allied to *E. sclerophylla* but can be recognised by the yellow corolla and the large, straw-coloured calyx and the shortly petiolate leaves with cordate bases which clasp the stem. *E. crispata* is unusual in subsection *Hypenia* and in *Eriope* as a whole for its yellow flowers. However, *E. sclerophylla* has variably-coloured flowers which are yellow in bud and change from yellow through shades of apricot and salmon-pink to red. The cymes in *E. crispata* are usually much more clustered in the inflorescence than other species in subsection *Laxiflorae,* although some specimens of *E. sclerophylla* are similar. *E. indaiaensis* has similar calyx and corolla morphology but differs in its more regular inflorescence structure with widely-spaced cymes. Epling (1936) keyed out *E. crispata* on the basis of its single bracteoles. I have been unable to find this character state in any specimens of *E. crispata* I have observed.

E. crispata is restricted to an area of west-central Goiás in the Serra Dourada and has the most westerly distribution of species in subsection *Ellipticae* (figure 10-12).

20. Eriope indaiaensis *R. Atkinson* sp. nov.

Type: Brazil, entre le passage du Rio Indayo et Bocaina, Goyaz, 11 Oct. 1895, *Glaziou* 21957 (holotype RB, isotypes K, G, P, S).

E. niquelandiensi affinis sed caulibus ceraceis et inflorescentiis floridis distincta.

E. indaiaensis is separated from *E. niquelandiensis* by its glabrous inflorescence branches and widely-spaced flowers.

Woody herb to 1 m. *Stems* erect, unbranched with long, virgate axes, moderately robust; basal parts pubescent with simple hairs and lanate hairs; upper parts drying black or pinkish brown, terete, glabrous, waxy; internodes fistulose, not swollen. *Leaves* petiolate, erect, persistent, distant; lamina ovate, 17 — 50 x 8 — 30 mm; base cordate; apex rounded; margin crenate to dentate, regular; upper surface pubescent to tomentose with simple hairs and some glandular hairs; lower surface tomentose with lanate hairs and some glandular hairs; petiole 2 — 5 mm, simple hairs and lanate hairs. *Inflorescence* a large, open, indeterminate thyrse, moderately floriferous; phyllomes narrowly lanceolate, 3 — 5 x 1 — 1.5 mm, frequently caducous; branches 2 per node, regularly developed, erect, straight, glabrous and waxy, pubescent on upper branches with minutely gland-tipped hairs; cymes 1-flowered, regularly spaced or crowded, 5 — 40 mm apart; peduncle erect, 3 — 14 mm, with sparse to moderate minutely gland-tipped hairs; paired bracteoles narrowly lanceolate, 2 — 2.5 mm. *Flowering calyx* vinaceous, erect, gibbous-campanulate, 7 — 9 mm, sparsely pubescent with minutely gland-tipped hairs; teeth broadly deltoid, 2.5 — 3 mm. *Fruiting calyx* straw-coloured, erect, cylindrical, 12 — 14 mm; teeth broadly deltoid -rostrate, 2 — 3 mm; throat with sparse long hairs or glabrous. *Corolla* tube red or reddish-orange to orange, cylindrical, straight, 15 — 24 mm; lateral and posterior lobes red or orangish-red, ca. 4 — 5 mm; anterior lobe red or orangish-red. *Nutlets* not seen.

DISTRIBUTION. Brazil, Minas Gerais, in the northwest of the state, near the River Indaiá (figure 10-12).
HABITAT. Savanna (cerrado) and upland savanna (campo rupestre). Altitude: ca. 720 m.

BRAZIL.
MINAS GERAIS. Montes Claros: Varzea da Palma - Corinto road, km 48, 4 Oct. 1965, *Ferreira & Marques* 93 (SP); Montes Claros: ca. 30 km SE on road to Juramento, 15 May 1977, *Gibbs* et al. 5117 (UEC); Montes Claros: between the passage of the Rio Indayo and Bocaina, 11 Oct. 1895, *Glaziou* 21957 (K, G, P, S); São Gonçalo: BR 040, km 251, 25 July 1984, *Mori* et al. 16992 (K, NY).

E. indaiaensis is placed in subsection *Ellipticae* because of the long corolla tube and clustering of the flowers at the apex of the branches. The leaves of this species are cordate and a similar shape to those of *E. niquelandiensis* but *E. indaiensis* can be distinguished by its glabrous and waxy inflorescence branches compared to those of *E. niquelandiensis* which

are pubescent. *E. crispata* has a similar large, straw-coloured calyx and *Gibbs 5117* has orange corollas also similar to *E. crispata.* The morphology of the inflorescence distinguishes the two species. *E. crispata* has a much more irregular inflorescence with cymes closely spaced along the branches whereas *E. indaiaensis* has a regularly branched inflorescence and the cymes are laxly spaced on the branches and only clustered toward the apex.

The area of distribution of *E. indaiaensis* is the most southerly of subsection *Ellipticae* and is also the most easterly, being the only species from this group endemic to Minas Gerais (figure 10-12). It is named for the River Indaiá, which is cited in the type locality but has now been absorbed by the Trés Marias reservoir in its lower reaches.

21. Eriope macrantha *(St. Hil. ex Benth.) R. Atkinson* comb. nov.

Type: Brazil, Minas Geraes, Paracatú, *St. Hilaire* 591, 1816-1821 (holotype P, isotype UC).

Hyptis macrantha St.Hil. ex Benth., Labiat. Gen. Spec.: 140 (1833); Bentham in DC. Prodr.12: 139 (1848); Schmidt in Fl. Bras. 8(1): 157 (1858); Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 232 (1936); Epling in Revista Mus. La Plata, secc. Bot. 7: 236 (1949); Epling & Mathias in Brittonia 8: 298 (1957).
Mesosphaerum macranthum (St. Hil. ex Benth.) Kuntze, Revis. Gen. Pl. 2: 157 (1891).
Hypenia macrantha (St.Hil. ex Benth.) Harley in Bot. J. Linn. Soc. 98: 92 (1988).

Woody herb to 3 m. *Stems* unbranched with long, virgate axes, moderately robust; basal parts woody, pubescent with long setose hairs (sometimes also with short simple and glandular hairs); upper parts glaucous green drying pinkish or dark brown, terete, glabrous, waxy; internodes fistulose, sometimes swollen. *Leaves* petiolate, erect, frequently caducous, distant; lamina lanceolate or oblong, 40 — 130 x 8 — 50 mm; base cuneate; apex rounded or acute; margin serrate, regular; upper surface sparsely to moderately pubescent with simple hairs, sometimes glandular hairs; lower surface moderately to densely pubsecent with simple hairs, subsessile glands and occasional glandular hairs; petiole 2 — 15 mm with simple hairs or glabrous. *Inflorescence* a large, open, indeterminate thyrse, moderately floriferous; phyllomes caducous; branches erect, straight, 2 (sometimes 4) per node, regularly developed, glabrous and waxy, higher order branches often with moderate to dense glandular hairs; cymes 1-flowered, regularly spaced 4 — 50 mm apart; peduncle erect, 6 — 15 mm,

moderately to densely pubescent with glandular hairs; paired bracteoles ca. 2 mm, broadly lanceolate or ovate, deflexed. *Flower* resupinate. *Flowering calyx* vinaceous, erect, gibbous-campanulate, 6 — 8 mm, densely pubescent with short glandular hairs; teeth 1 — 3 mm, narrowly deltoid, rostrate. *Fruiting calyx* erect, 9 — 12 mm; teeth 2 — 4 mm; throat with sparse long hairs or glabrous. *Corolla* tube red, cylindrical, straight, 20 — 26 mm long; posterior and lateral lobes red ca. 7 mm, spreading; anterior lobe red, sometimes with yellow mark on retracted apex, 5 — 6 mm. *Stamen* filaments pink with white hairs; anthers yellow. *Style* pink. *Nutlets* ovoid in section, 3 — 4 x 1.5 — 2.5 mm, muilaginous.

DISTRIBUTION. Brazil, Distrito Federal and surrounding parts of Goiás and Minas Gerais (figure 10-12).

HABITAT. Grassy savanna (cerrado, campo limpo type). Altitude: ca. 1100 m.

BRAZIL.

DISTRITO FEDERAL. Brasília: Fazenda Água Limpa, UNB experimental station, 19 July 1995, *Atkinson & Giorgio* 144 (UB, K, IBGE); Brasília: ca. 18 km SSW, Reserva Ecologica do IBGE, 2 Sept.1995, *Atkinson & Giorgio* 152 (K, UNB, IBGE); Brasília: Fazenda Água Limpa, 19 July 1989, *França & Mello* 20219 (NY); Brasília: 3 June 1960, *Gomez* 1152 (RB); Brasília: basin of Rio São Bartolomeu, 8 Oct. 1979, *Heringer* et al. 2197 (K); Brasília: 2 March 1978, *Heringer* 16874 (K); Brasília: Fazenda Água Limpa, 3 Oct. 1980, *Kirkbride* 1388 (UB); ; Brasília: Braslândia, 23 July 1961, *Macedo* 11 (RB)Brasília: Cabeça do Veado stream, 21 Sept. 1982, *Matos* 13 (K); Brasília: Cristo Redentor, next to Taquara stream, 25 Aug.1988, *Mendonça* 1052 (K); Brasília: Parque Naçional da Gama, *Onishi* 120 (UB); Brasília: SE of dam, Rio Paranoá, 5 May 1968, *Philcox & Onishi* 4918 (K); Brasília: between Brasília and Fercal, 30 June 1964, *Pires* 58071 (K, UB); Fercal: 13 July 1976, *Davis* 60259 (E, UEC).

GOIÁS. Ipameri: 5 - 10 km N, Rio Corumbá, 11 July 1976, *Hatschbach* 38803 (K, MBM); Luziânia: Lagoa de Prata, 15 Aug. 1980, *Hatschbach* 43150 (MBM, K); Luziânia: Cidade Industrial, 30 June 1975, *Heringer* 14605 (K, NY, UEC).

MINAS GERAIS. Paracatú: Minas Geraes, 1816-1821, *St. Hilaire* 591 (P, UC).

E. macrantha is recognised by the moderately floriferous inflorescence, the large flowers held at the apex of the inflorescence branches, lanceolate to ovate leaves and glandular pubescence on the peduncle and calyx. There is considerable variation in the indumentum of different specimens. The type and specimens from Distrito Federal have leaves, peduncles and calyx lacking conspicuous glandular hairs whereas *Hatschbach 38803* and *43150* from Goiás have conspicuous glandular hairs

The type specimen of *E. macrantha, St.Hilaire 591,* is undoubtedly conspecific with the resupinate-flowered taxon observed in Distrito Federal. There are no more recent collections of *E. macrantha* from Paracatú although this locality is close to Distrito Federal.

Despite being endemic to the cerrados around Brasília, some of the most intensively collected cerrado in Brazil, it is not represented by a large number of specimens indicating that it is not common throughout its area. Its distribution (figure 10-12) overlaps with that of *E. reticulata* which is much more common in the same area. The two species are clearly distinguished by the resupinate corolla and larger flowers of *E. macrantha.*

22. Eriope niquelandiensis *R.Atkinson* sp. nov.

Type: Brazil, near Niquelândia, Goiás, *Atkinson , Walter, Silva Jr., Assis, Silva* 140, (holotype CEN, isotypes K, IBGE, UB).

E. indaiaensis affinis sed caulibus puberulis et inflorescentiis pauciflоris differt.

E. niquelandiensis can be separated from *E. indaiaensis* by the pubescent inflorescence branches and fewer flowers.

Woody herb to 2 m. *Stems* erect, unbranched with long, virgate axes, moderately robust to slender; basal parts woody, pubescent with simple and glandular hairs and setose hairs; upper parts olive green to brown, densely to moderately pubescent with simple and glandular hairs, terete or slightly angled, wax absent; internodes not fistulose, not swollen. *Leaves* petiolate, spreading, persistent, distant; lamina ovate, 35 — 135 mm x 12 — 75 mm; apex acute or rounded; base cordate or truncate; margin coarsely dentate to crenate, irregular; upper surface sparsely pubescent with simple and glandular hairs; lower surface sparsely pubescent with simple hairs on vein margins, subsessile glands in the lumina; petiole 5 — 10 mm, pubescent with glandular hairs. *Inflorescence* a large, open, indeterminate thyrse, few-flowered or moderately floriferous; phyllomes narrowly lanceolate, 5 — 10 mm x 1 — 2 mm; branches erect, straight, 2 per node, regularly developed, pubescent with dense to moderate short glandular and non-glandular hairs; cymes 1-flowered, regularly spaced 4 — 20 mm apart; peduncles erect, 6 — 15 mm, pubescent with short glandular hairs; paired bracteoles ca. 1 mm. *Flower* resupinate. *Flowering calyx* vinaceous, horizontal, gibbous-campanulate, 4 — 8 mm, sparsely pubescent with simple and glandular hairs; teeth 1 — 3 mm, symmetrical,

broadly deltoid. *Fruiting calyx* erect, 7 — 8 mm; throat with sparse long hairs; teeth 2 mm, broadly deltoid. *Corolla* tube red, cylindrical, straight, 15 — 25 mm; posterior and lateral lobes red, spreading, 5 — 10 mm; anterior lobe red with yellow mark on retracted apex, ca. 5 mm. *Stamen* filaments pink with white hairs; anthers yellow. *Style* pink. *Nutlets* ovoid in section, 3 — 4 x 1.5 — 2.5 mm.

DISTRIBUTION. Brazil, northwestern Goiás endemic to a small area near Niquelândia and with a disjunction to serpentine rocks in southern Goiás (figure 10-12).
HABITAT. Savanna (cerrado) and secondary growth in the vicinity of serpentine rocks. Altitude: 400—800m.

BRAZIL.
GOIÁS. Morro Feio, 8 June 1990, *Brooks & Reeves* TMEX 425 (K); Niquelândia: ca. 25 km NE in Serra da Mesa, near Codemin, 22 July 1995, *Atkinson* et al. 140 (CEN, IBGE, UB, K); Niquelândia: southernmost ultramafic hill of Tocantine complex, 29 April 1988, *Brooks* et al. BRASPEX 230 (NY); Niquelândia: nickel mine, 15 Aug. 1996, *Mendonça* et al. 2594 (K, IBGE); Niquelândia: 24 July 1952, *Macêdo* 3645 (UC, S).

E. niquelandiensis can be recognised by the usually cordate leaves with irregular dentate margins and the long, straight-tubed corollas, exserted from the calyx by more than three quarters of their length. The inflorescence branches are short and pubescent and the inflorescence is few-flowered. It occurs in the same area as *E. aristulata,* a non-resupinate member of subsection *Laxiflorae,* and may overlap in its distribution with *E. calycina* in subsection *Ellipticae.* It can be easily separated from *E. calycina* by the lack of broad, overlapping bracteoles, by the pubescence on the inflorescence branches and by the long, straight and red corolla tube.

E. niquelandiensis seems to be restricted in its habitat preferences and has a limited distribution (figure 10-12). Most collections come from the area of Niquelândia in northern Goiás but there is a collection from ultramafic rocks further south (*Brooks & Reeves, TMEX 425*). This specimen differs from those from Niquelândia in its larger leaves and smaller corolla. It has the same regularly, shortly-branched, pubescent inflorescence of *E. niquelandiensis* and appears to represent a disjunction associated with geology.

23. Eriope sclerophylla (*Epling*) *R. Atkinson* comb. nov.

Type: Brazil, entre Megaponte e Caïsara, Goyaz, *Burchell* 6325 (holotype K).

Hyptis durifolia Epling in Revista Mus. La Plata, secc. Bot. 7: 235 (1949).
Hypenia durifolia (Epling) Harley in Bot. J. Linn. Soc. 98: 92 (1988).
Hyptis sclerophyllum Epling in Repert. Spec. Nov. Regni Veg. Beih. 85: 234 (1936). **nom illeg. non** *Hyptis sclerophyllum* Briq. in Die Nat. Pflanz. IV, 3a: 340 (1896).
Hyptis macrantha St. Hil. ex Benth. sensu Epling & Mathias in Brittonia 8: 298 (1957) **pro parte**.

Woody herb to 3 m. *Stems* erect, unbranched with long, virgate axes, moderate to very robust; basal parts sparsely pubescent with short simple hairs and setose hairs; upper parts drying purplish brown, terete, glabrous, waxy; internodes fistulose, sometimes swollen. *Leaves* petiolate, erect, distant; lamina, ovate or oblong-lanceolate, 65 — 200 x 25 — 110 mm; apex rounded or acute; base cuneate or truncate; margin crenate (sometimes serrate), regular; upper surface sparsely pubescent with short simple hairs and subsessile glands; lower surface sparsely to moderately pubescent with short simple hairs along the veins and subsessile glands in the lumina; petiole 10 — 75 mm, glabrous or with sparse long setose hairs and short simple hairs. *Inflorescence* terminal, a large open, indeterminate thyrse, floriferous; phyllomes narrowly lanceolate, ca. 5 x 1.5 mm, frequently caducous; branches erect or spreading, straight, 2 (rarely 4) per node, irregularly developed, glabrous or pubescent on upper branches; peduncle 3 — 12 mm, glabrous or pubescent with short hairs, sometimes with branches developing in the axils; cyme bracts 1.5 — 3 mm, narrowly lanceolate; cymes 1-flowered, crowded, 2 — 25 mm apart; bracteoles 1.5 — 3 mm, narrowly lanceolate. *Flowering calyx* vinaceous, erect, gibbous-campanulate, 7 — 11 mm, pubescent with short simple hairs subsessile glands; teeth narrowly deltoid, 2.5 — 4 mm. *Fruiting calyx* erect, 12 — 14 mm, papery; teeth ca. 4 mm; throat with very sparse long hairs or glabrous. *Corolla* tube deep pink, apricot or red, yellow in bud, 16 — 18 mm, cylindrical; posterior and lateral lobes deep pink, apricot or red, ca. 6 mm; anterior lobe ca. 5 mm. *Nutlets* ovoid in section, 3 — 4 x 2 — 3 mm.

DISTRIBUTION. Brazil, Goiás, restricted to the area around the town of Anápolis, central Goiás.

HABITAT. Savanna (cerrado). Altitude: ca. 1000 m.

BRAZIL.

GOIÁS. S. loc., Goyaz, 1896, *Glaziou* 21921 (G, P); s. loc., 1896, *Glaziou* 21922 (K, G); loc. incert., Fazenda da Taboquinha, *Glaziou* 21924*(K, G); Anápolis: 2 April 1958, *Lima* 3010 (K); Anápolis: 9 July 1951, *Macêdo* 3284 (UC); Anápolis: 21 July 1952, *Macêdo* 3573 (NY, S); Belle Valle de Chico Costa, 10 Sept. 1895, *Glaziou* 21923* (K, G); Caiçara: between Meiaponte and Caïsara, 23 Oct. 1827, *Burchell* 6325 (K); Pirenópolis: 25 May 1976, *Heringer* 15834 (UB); Pirenópolis: ca. 20 km E, Serra dos Pirineus, 16 Jan. 1972, *Irwin* et al. 34263 (K, NY).

* see note under *E. calycina*.

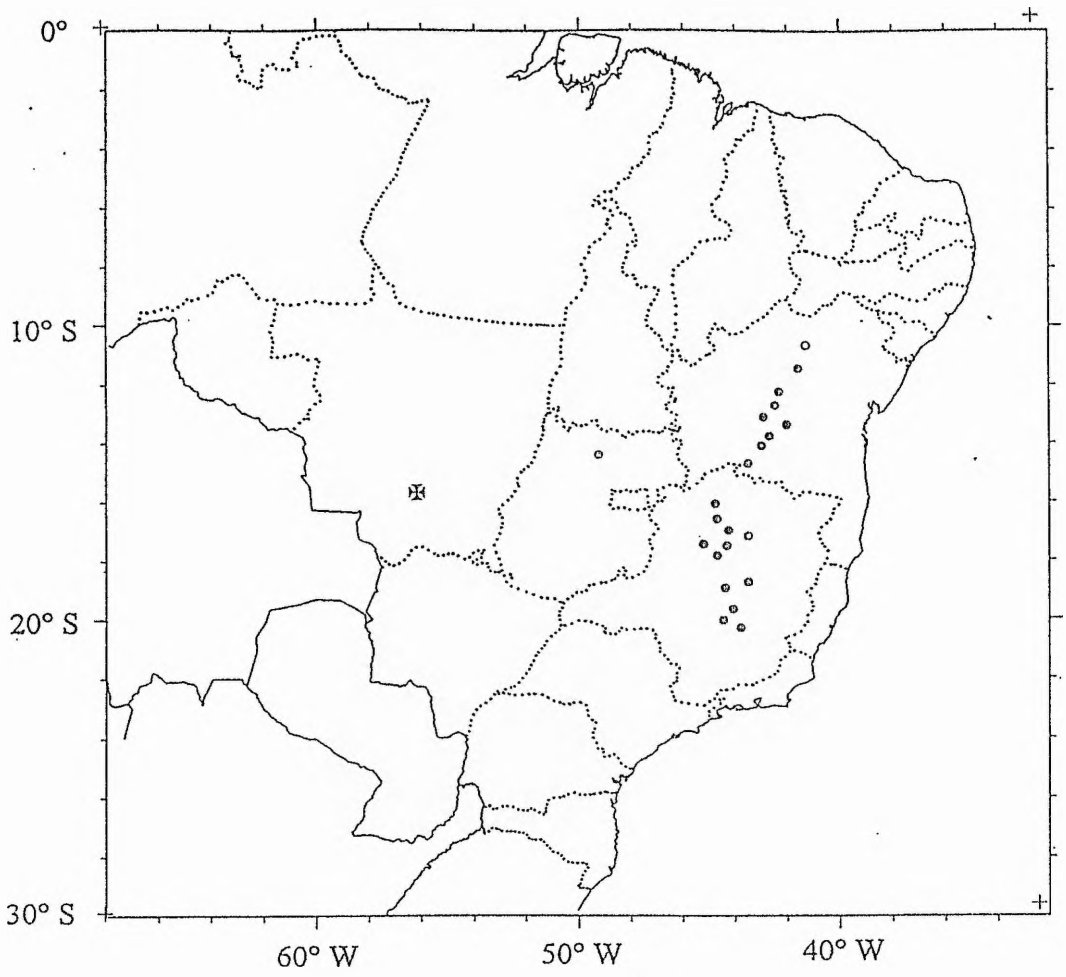
E. sclerophylla, as the name implies, has conspicuous leaves which are very thick and amongst the largest in the genus. The flowers appear to be resupinate on herbarium sheets and this has been confirimed in the field by Harley (pers. comm.). The arrangement of cymes in the inflorescence is very similar to *E. crispata* but *E. sclerophylla* is readily distinguished by its large, distinctly petiolate leaves and smaller, red flowers. One replicate of *Lima 3010* has very few flowers in the inflorescence and is superficially similar to non-resupinate species of subsection *Laxiflorae*. Other replicates of the same collection are more typical of *E. sclerophylla*. This illustrates the variability between individuals of species formerly included in *Hypenia*, particularly in the development of the inflorescence.

The distribution of *E. sclerophylla*, like all the species in subsection *Ellipticae*, is very restricted. It occurs in a small area of central Goiás, intermediate between localities of *E. macrantha* and *E. crispata* (figure 10-12).

G. Taxonomic index

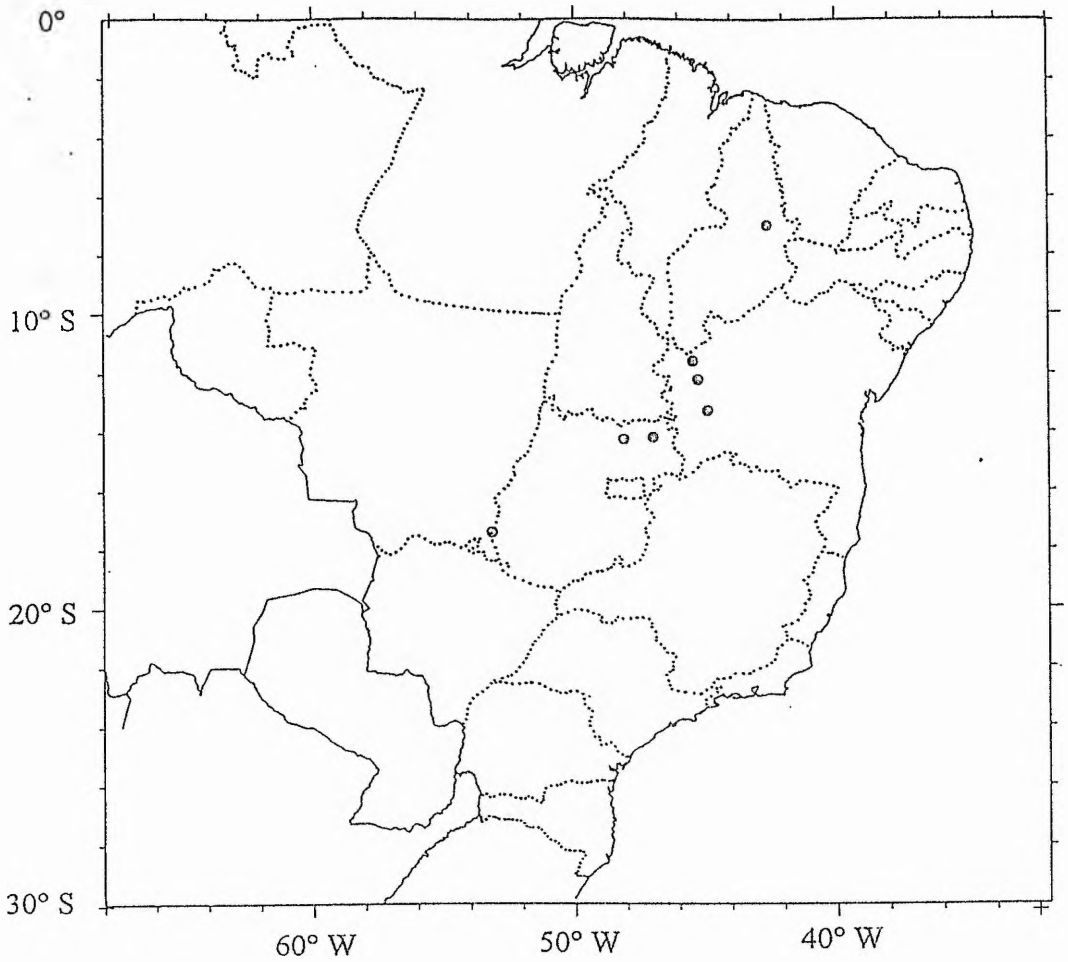
Accepted names are given in bold, synonyms in italics. Numbers in bold after synonyms refer to the currently accepted name.

H. Distribution maps



- *E. vitifolia*
- *E. longicaulis*
- *E. micrantha*

Figure 10-6: Distribution of sections *Micranthae* and *Vitifoliae*



- *E. irregularis*
- *E. gracilis*

Figure 10-7: Distribution of section *Irregulares*

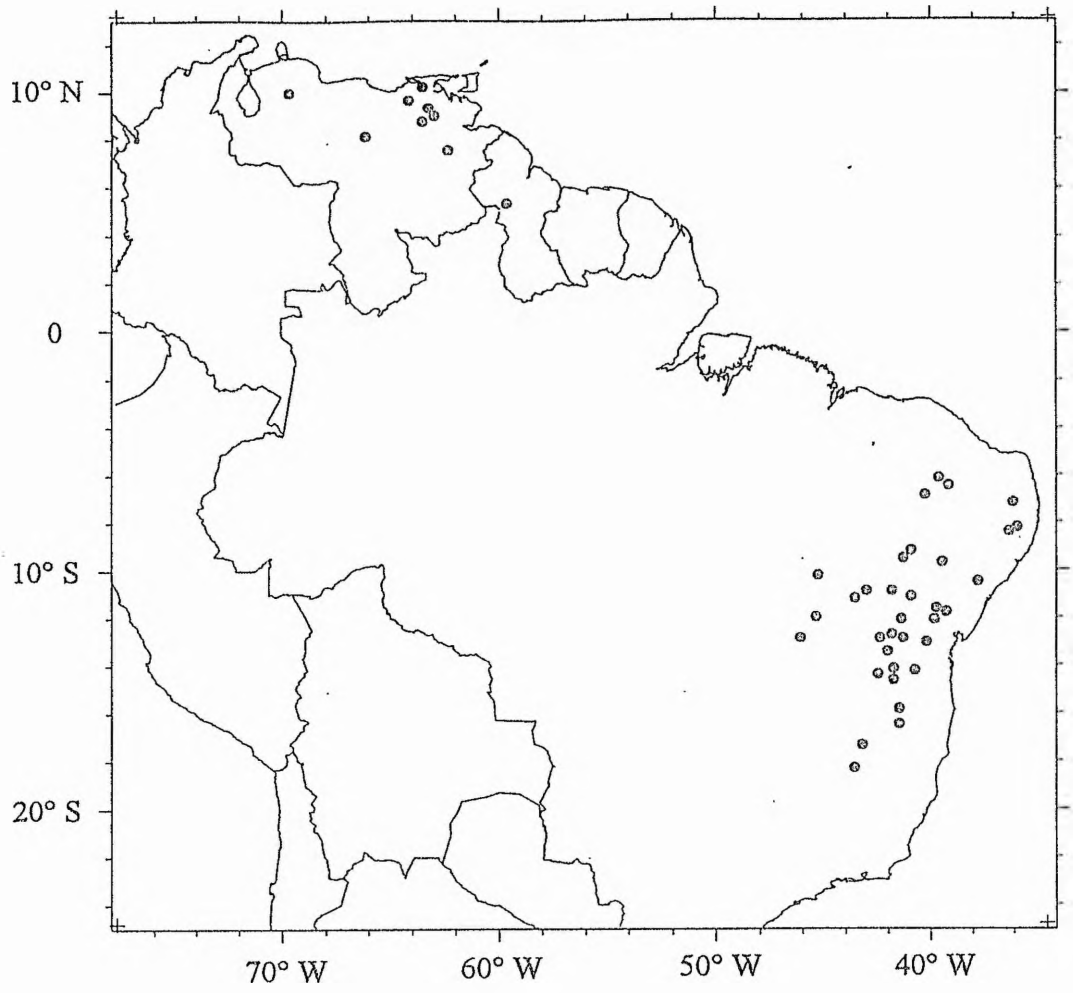
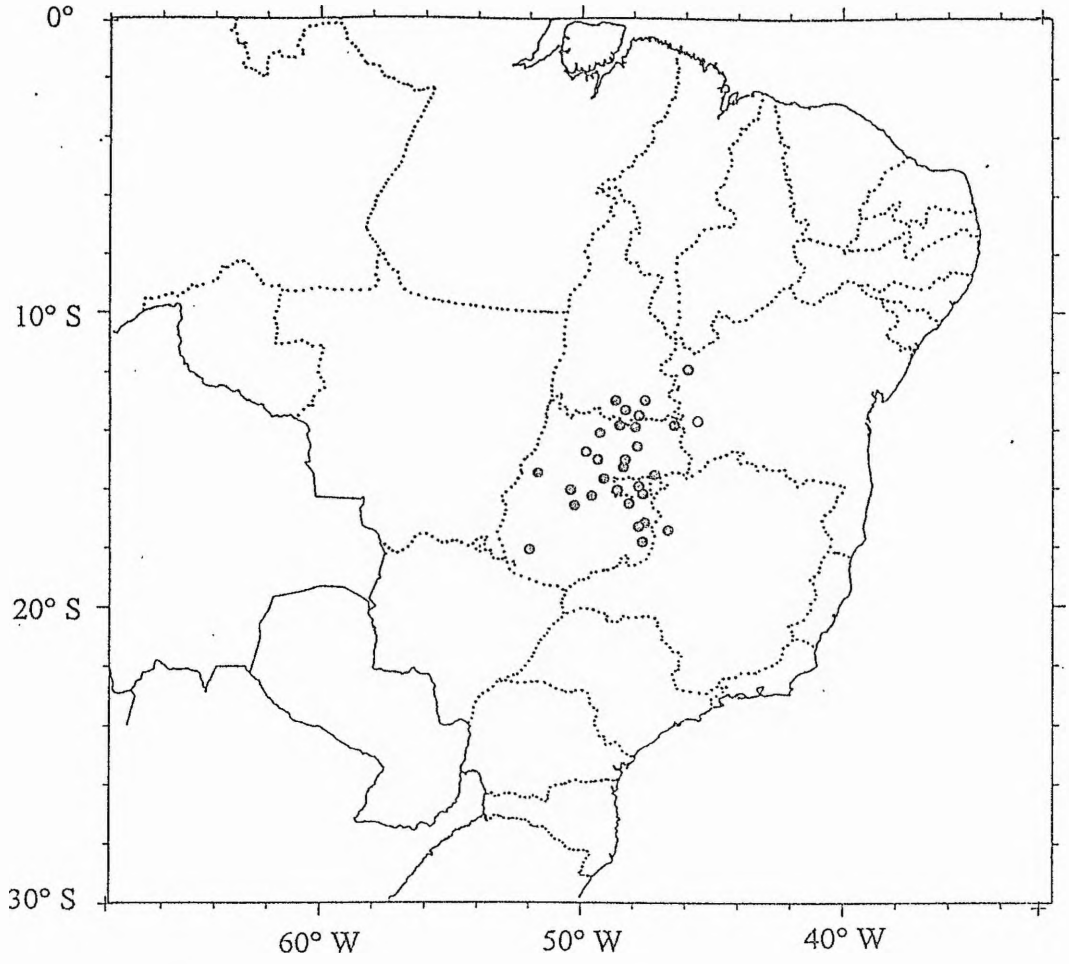
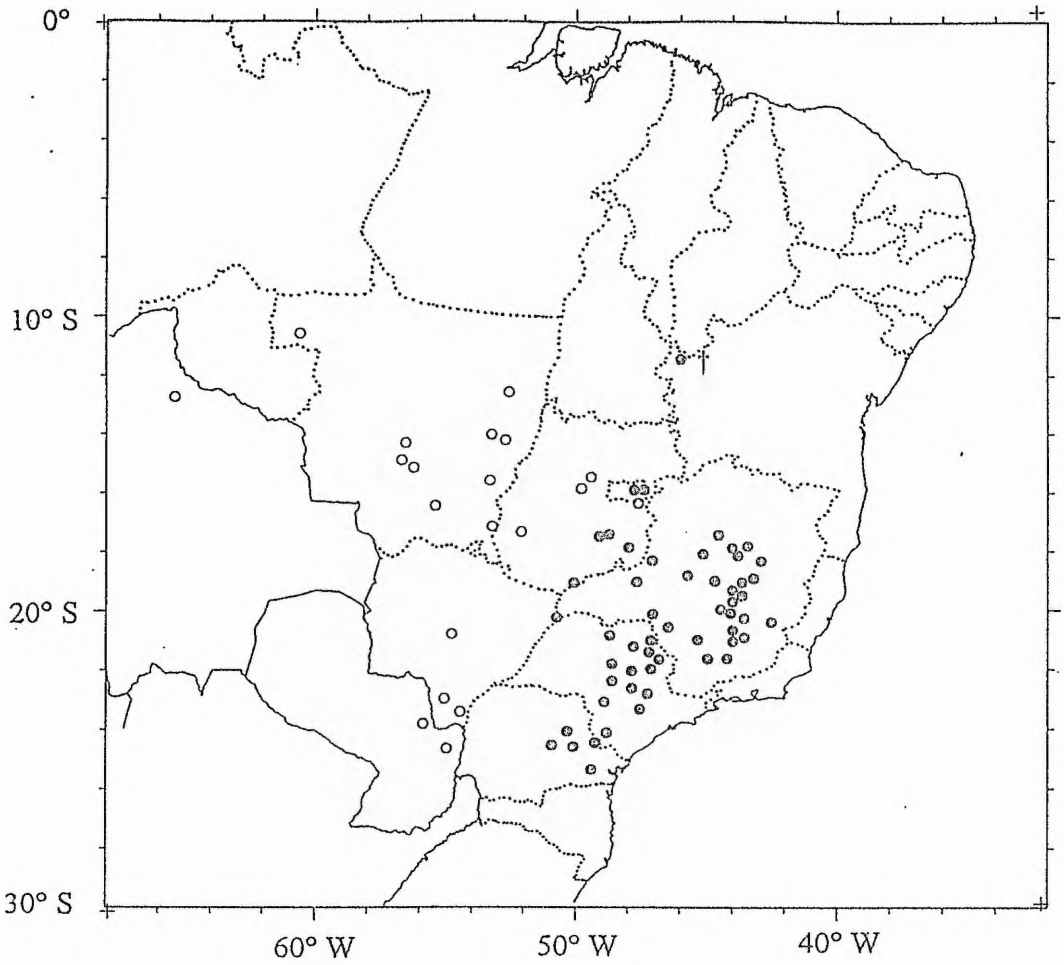


Figure 10-8: Distribution of section *Salzmanniae*



- *E. brachystachys*
- *E. concinna*
- *E. densiflora*

Figure 10-9: Distribution of section *Densiflorae*

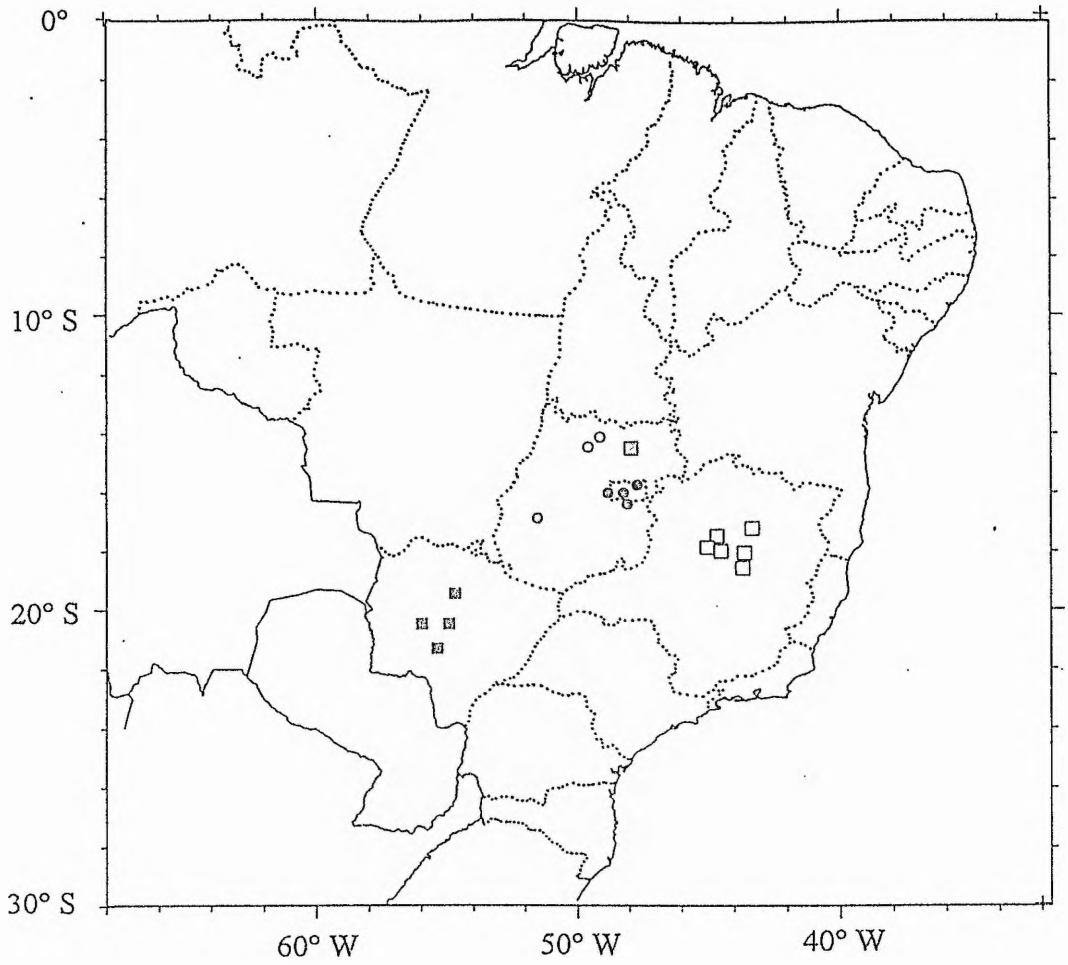


● *E. reticulata*

◉ † Gardner 3391

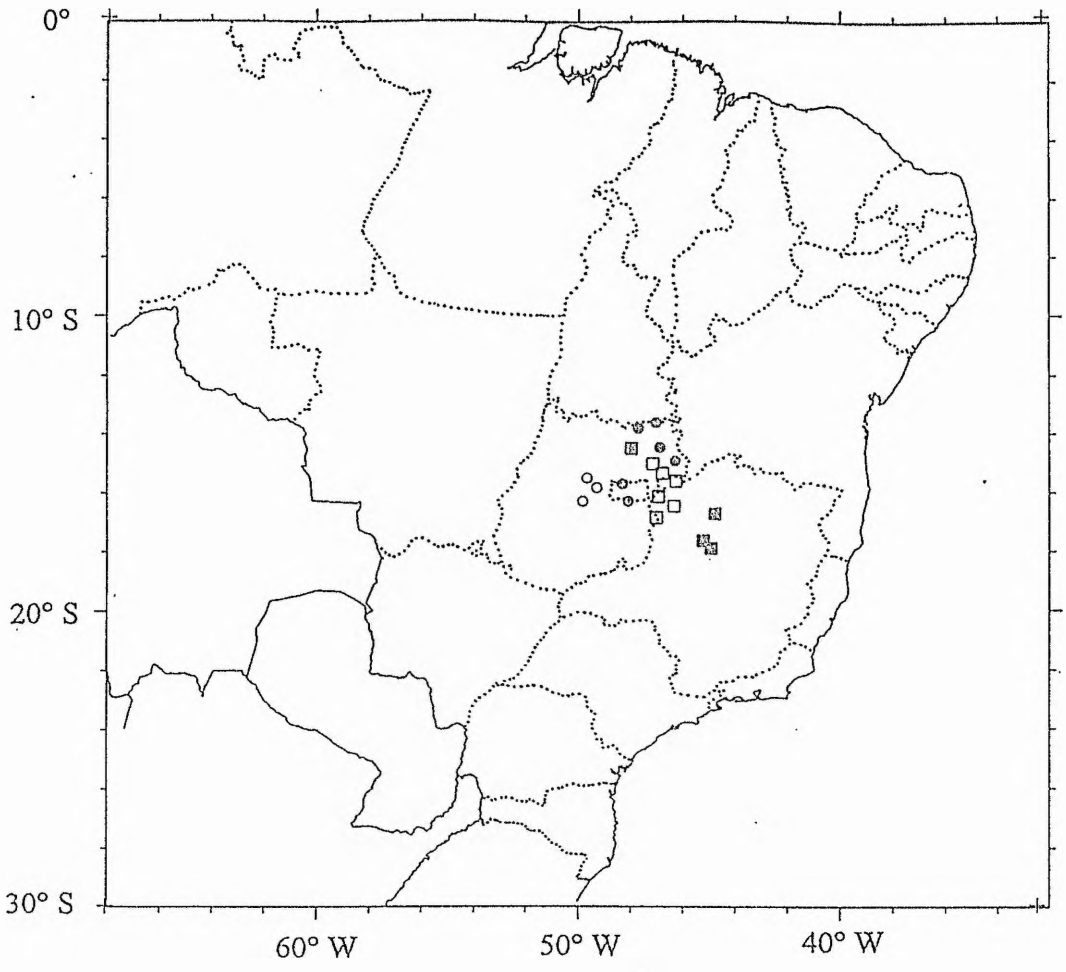
○ *E. macrosiphon*

Figure 10-10: Distribution of *E. macrosiphon* and *E. reticulata*



- *E. recoria*
- *E. aristulata*
- *E. caiaponiensis*
- *E. hatschbachii*
- *E. paniculata*
- *E. subrosea*

Figure 10-11: Distribution of six species in section *Hypenia* subsection *Laxiflorae*



- *E. calycina*
- *E. crispata*
- *E. sclerophylla*
- *E. indaiensis*
- *E. macrantha*
- *E. niquelandiensis*

Figure 10-12: Distribution of section *Hypenia* subsection *Ellipticae*

Chapter 11: Biogeography of *Eriope*

A. Introduction

This chapter is concerned with the biogeography of species of *Eriope* which were formerly included in *Hypenia* sensu Harley (1988a). The geographical distributions of these species are closely linked to the distribution of savanna vegetation in central South America and the first part of the chapter describes the savannas of central Brazil. The geographical distribution of the relevant species of *Eriope* is discussed in the second part of the chapter.

B. The vegetation of central Brazil

Eriope species are largely confined to savanna or dry woodland. Most species of *Hypenia* sensu Harley (1988a) are found in the cerrados of central Brazil but some species of *Eriope,* including the majority of section *Eriope* are found in a closely related vegetation, campo rupestre. These two vegetation types are described below before the distributions of individual species are discussed.

i) Cerrado

Cerrado is the dominant vegetation of central Brazil and extends into neighbouring parts of Paraguay and Bolivia. It consists of a well-developed herbaceous layer, dominated by grasses, with varying degrees of woody cover. Several classes of cerrado can be recognised depending on the density of the woody vegetation. Campo limpo ('clean field') is dominated by grasses and other herbs with scattered bushes. Cerrado *sensu stricto* retains a similar ground layer but the density of woody vegetation makes it difficult to ride a horse through it (hence 'cerrado' which means closed in Portuguese). Cerradão has a reduced herbaceous ground layer and a more or less closed canopy of woody species. Tree species are typically sclerophyllous, evergreen or deciduous and often have contorted trunks and branches. Many show adaptations to fire such as corky bark and the development of underground xylopodia. There is considerable heterogeneity in the species composition of cerrado in different regions and there are few woody species which are found throughout the whole cerrado region. Some of the most widespread species include *Curatella americana* L. (Dilleniaceae), species of *Qualea* Aubl. (Vochysiaceae), *Caryocar brasiliense* Cambess. (Caryocaraceae) and *Dimorphandra mollis* Benth. (Leguminosae: Caesalpinoideae) (Ratter et el. 1996).

Cerrado extends across the Central Brazilian Plateau and occupies an altitudinal range from just above sea level to 1800 metres. The region is dissected by river systems which flow westward into the Rio Paraná, eastward into the Rio São Francisco or northward into the Amazon basin. The climate of the region is distinctly seasonal with a dry season from April to September and average rainfall of 800 - 2000 mm. Temperatures average between 18 - 28 °C. Cerrado occurs on dystrophic soils which have a low levels of calcium and magnesium and high levels of aluminium (Furley & Ratter 1988). The soil conditions vary within the cerrado and there is a marked association of some species with the patches of mesotrophic soils which occur within the cerrado region (Furley & Ratter 1988).

The rivers and streams which cross the cerrado region are bordered by gallery forests which are floristically allied to the Amazon and Atlantic forest regions but also contain significant numbers of endemics (Oliveira-Filho & Ratter 1995). The southern edge of cerrado borders the open savanna 'chacos' in Argentina and Paraguay and the northeastern edge abuts the dry, thorny 'caatinga' scrub. Central regions of South America are therefore dominated by a series of vegetation types characterised by a marked dry season. The distribution of plant species indicates that seasonal woodland was formerly distributed in a more or less continuous arc from northeastern Brazil into northwestern Argentina but largely skirting the dystrophic cerrado soils (Prado & Gibbs 1993). However, there are also patches of deciduous and semideciduous forest on the more fertile soils within the cerrado region which currently connect the caatingas of northeastern Brazil and the chacos of northern Argentina (Oliveira-Filho & Ratter 1995). The northern and western fringes of cerrado are bordered by the *hylaea,* or Amazon forest, which under current climatic conditions is advancing onto cerrado in many places (Ratter 1992). The eastern edge of the Central Brazilian Plateau is bordered by the Serra do Espinhaço range of mountains which form a chain through the states of Minas Gerais and Bahia. The vegetation of the higher altitude regions of the Serra do Espinhaço and other mountain ranges in the cerrado region is known as campo rupestre and is discussed in more detail below. Some savanna regions are scattered throughout the Amazon basin and contain a few cerrado species although they are generally much more species-poor than cerrado in central Brazil.

With the addition of fertilisers cerrado soils are easily converted to agricultural production. As a result large tracts of cerrado have been destroyed and over 40% of the original cerrado vegetation is estimated to have been lost (Ratter et al. 1997).

ii) Campo rupestre

Campo rupestre is a term used to refer to a mosaic of different vegetation types found at high altitude in the Serra do Espinhaço range (Giulietti & Pirani 1988) and in mountainous regions of central Brazil. The high altitude, over 900 metres, and the extensive rocky outcrops with shallow soils are the principal defining characteristics of campo rupestre. The soils are sandy or stony, nutrient poor and acidic. In the areas where soils accumulate herbaceous communities dominated by Gramineae, Cyperaceae, Eriocaulaceae and Xyridaceae are found with small trees, shrubs and subshrubs scattered throughout. The trees and shrubs are evergreen and sclerophyllous. The rock outcrops, where soils are thin or non-existent, support members of the Velloziaceae, Orchidaceae and Bromeliaceae. Areas of impeded drainage occur between the rock outcrops and bog communities occur here dominated by members of the Xyridaceae, Cyperaceae, Eriocaulaceae, Gentianaceae and Lentibulariaceae. Cerrado occurs in lower altitude areas adjoining campo rupestre and forests may be found along watercourses and on valley sides. The northern end of the Serra do Espinhaço drops in altitude and as it does so gives way to caatinga in northern Bahia. With reducing altitude on the western side of the mountain range cerrado becomes the dominant vegetation type.

The climate in the Serra do Espinhaço is seasonal and the dry season occurs in the same months as in the cerrado region. However, rain and dew are important sources of moisture during the dry season. Rainfall averages 1500 mm but it is very variable and the southern end of the range is much wetter than the north. The caatinga region in the north of Bahia can experience extreme droughts which may affect neighbouring campo rupestre regions. Temperatures can fall below 18°C in the dry season and rarely exceed 22°C in the wet season.

Plant species in campo rupestre show a number of distribution patterns. Some species belong to genera which are otherwise more species-rich in the Andes, e.g. *Drimys brasiliensis* Miers (Winteraceae) but there are a number of species which are widely distributed in cerrados and campos throughout South America, e.g. *Tapirira guianensis* Aubl. (Anacardiaceae). Other distribution patterns show connections between the restingas (coastal sands) and the mountain ranges (serras) of Goiás in the cerrado region. In addition there are a large number of species which are endemic to the Serra do Espinhaço itself. Some families have

particularly high numbers of endemics, especially the Eriocaulaceae, Velloziaceae and Compositae (Stannard 1995). The Labiatae also have many endemic species in the region (Harley 1995a).

Because of the mountainous topography and poor soils campo rupestre has not been subject to major disturbance for agriculture. However, many of the forests which reach into campo rupestre regions have been devastated and considerable damage has been caused to the vegetation by burning and mining. Some endemic species may also be threatened by over-exploitation, e.g. species of *Syngonanthus* Ruhland (Eriocaulaceae) which are harvested for the dried flower trade and *Vellozia sincorana* L. B. Sm. & Ayensu (Velloziaceae) which is locally prized as a firelighter (Harley 1995b).

iii) Historical change in cerrado and campo rupestre

Historically cerrado has existed in a dynamic equilibrium with the Amazonian forest and this interaction is still continuing (Ratter 1992). It has been proposed, from evidence accrued from the distribution of plant and animal species, that during drier, cooler periods in the Pleistocene the evergreen Amazonian forest was restricted to isolated refugia in the Amazon basin surrounded by a mosaic of dry forest and savanna formations (Prance 1982, Prado & Gibbs 1993). The Refuge Theory has been used as an explanation for the high levels of species diversity in the neotropics and, although other mechanisms undoubtedly play an important part, for example ecology, dispersal ability and stochastic events, it highlights the dynamic nature of neotropical vegetation and the importance of historical processes in explaining current distribution patterns.

The drier, cooler conditions of the Pleistocene would also be expected to favour the expansion of campo rupestre. The occurrence of typical campo rupestre species in coastal restingas and on isolated mountains indicates that this could indeed have occurred (Harley 1995b).

iv) Plant distribution patterns in cerrado

The tree and large shrub flora of the cerrado is comparatively well known and about 800 species are thought to occur here. The diversity of the ground layer is much less well known however, but it is estimated that there are four to seven times the number of ground layer species compared to trees and large shrubs (Ratter et al. 1997). From the list of trees and

large shrubs published in Ratter et al. (1996) it is interesting to note that there are very few large genera. Some of the most speciose genera, e.g. *Miconia* Ruíz & Pav. (Melastomataceae) and *Eugenia* L. (Myrtaceae) are in need of revision and are consist mostly of widely distributed shrubs which occur in a range of habitat types. *Byrsonima* Rich. ex Kunth (Malpighiaceae) was the most species-rich genus of trees in cerrado with 18 species. By contrast, the ground flora contains many more species in the same genera. *Banisteriopsis* C. Rob. ex Small also in the Malpighiaceae is a genus of about 90 species of subshrubs and climbers, two thirds of which occur in cerrado (Gates 1982). *Mimosa* L. is an extremely large genus which is particularly speciose in cerrado where there are several species complexes, e.g. Barneby (1991) recognises 10 varieties of *M. clausenii* Benth., all of which are confined to central cerrado regions in Goiás. The taxonomy of many central Brazilian genera is poorly known which makes it difficult to draw conclusions about the distribution of species in the cerrado. However, it appears that the cerrado region has been the centre of diversification in a number of herbaceous or shrubby plant genera. This is undoubtedly the case for *Eriope,* especially section *Hypenia,* and is discussed further below.

An analysis of the woody vegetation from 98 areas in cerrado indicated the heterogeneity of the cerrado flora (Ratter et al. 1996). Of the 534 species included in the analysis only 28 were present at 50% or more of the 98 areas compared to 158 species which were present at one site only. The sites with the most similar species lists were strongly correlated with geography and it was possible to identify the following clusters: southern (São Paulo and neighbouring parts of Minas Gerais); south-eastern (Minas Gerais); central (central and southern Goiás and the 'Triângulo Mineiro'); central-western (western Goiás, Mato Grosso and Mato Grosso do Sul); northern (Tocantins, Maranhão, Piauí and Ceará); and Amazonian (Pará and Amazonas).

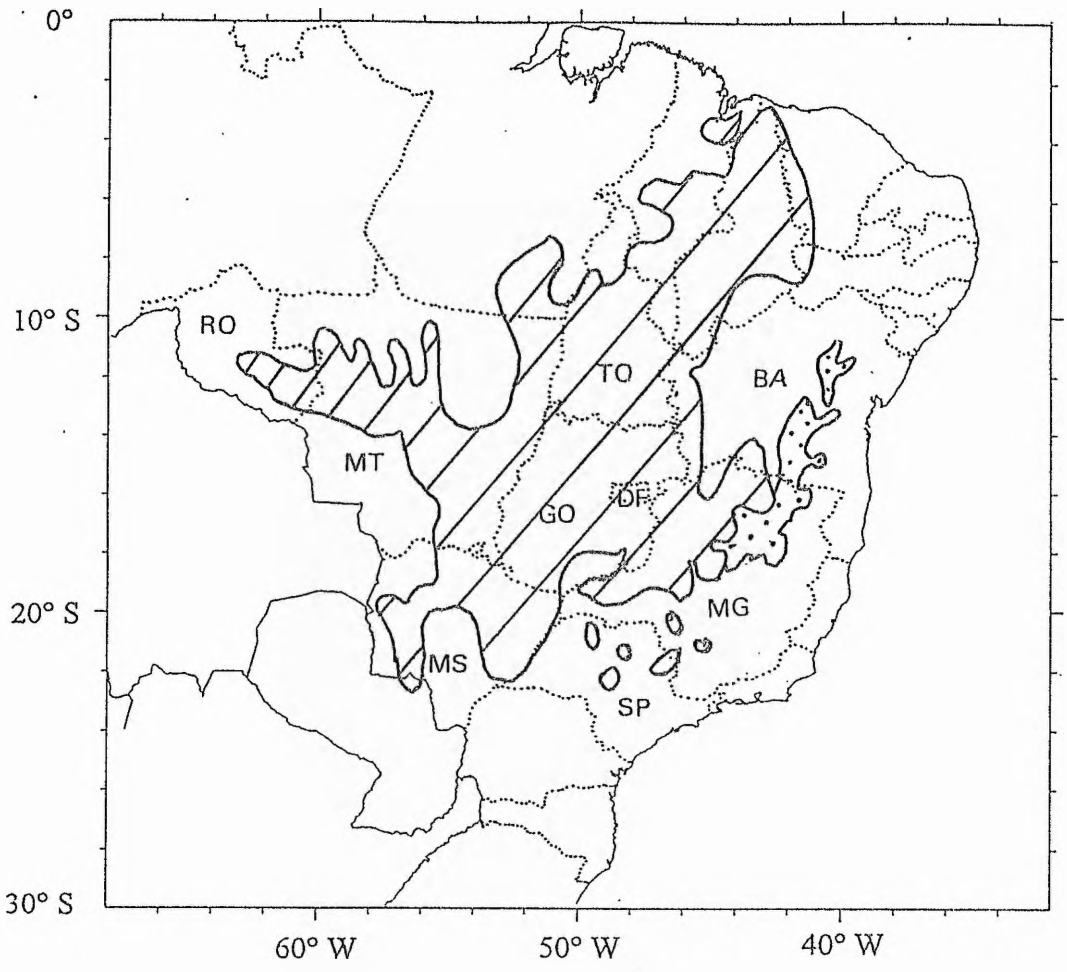


Figure 11-1: Approximate distribution of cerrado (diagonal lines) and campo rupestre (stippled) in Brazil

Letters refer to the Brazilian states mentioned in the text: BA = Bahia; TO = Tocantins; DF = Distrito Federal; MG = Minas Gerais; SP = São Paulo; GO = Goiás; MT = Mato Grosso; MS = Mato Grosso do Sul; RO = Rondônia. The isolated patches marked in SP and MG are cerrado.

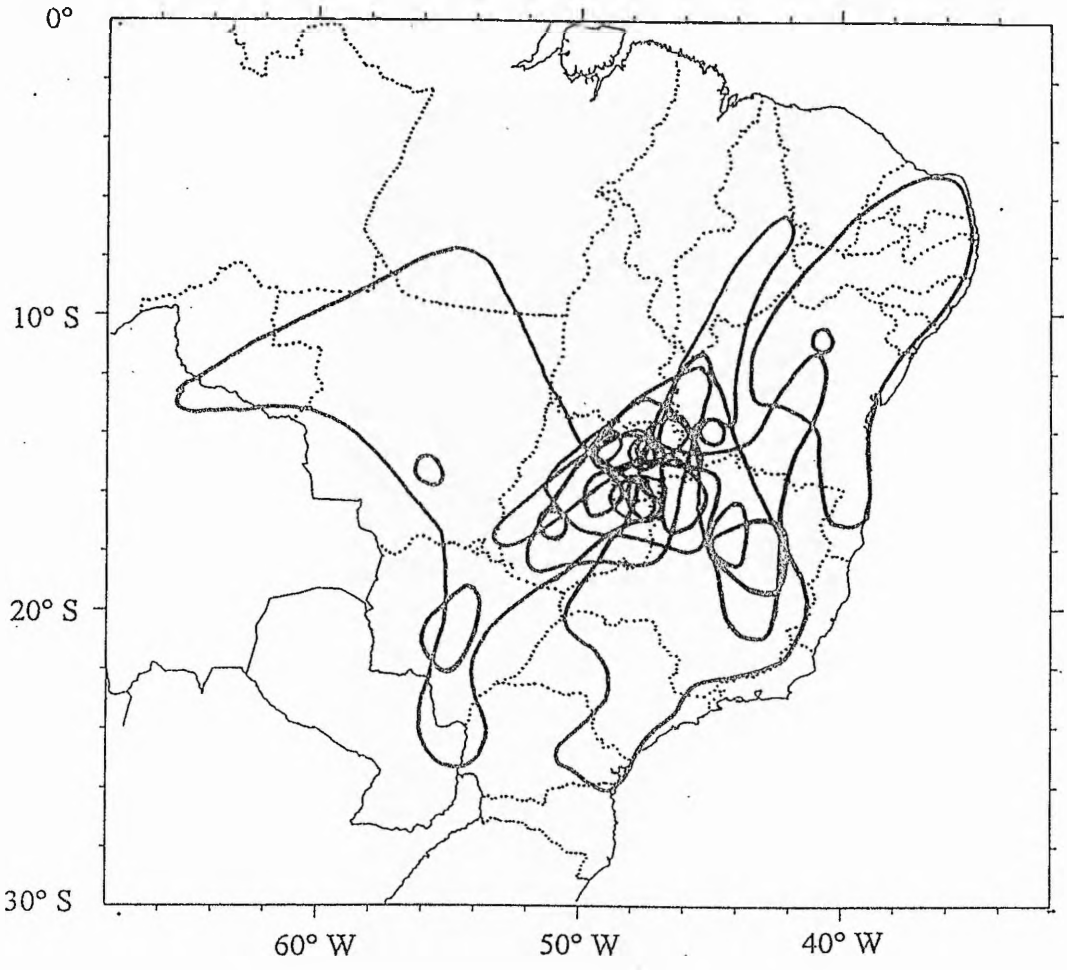


Figure 11-2: Ranges of *Eriope* species covered in this account in South America

Approximate ranges of *Hypenia* species are superimposed to demonstrate areas of maximum diversity in the group.

c. Distribution of *Eriope* species

The distribution of individual species of *Eriope* covered in this account is discussed briefly below. The distribution of section *Eriope* is also considered

i) Section *Eriope*

Section *Eriope* contains about 36 species, two thirds of which are endemic to the campo rupestre in the Serra do Espinhaço (Harley appendix I and pers. comm.). Harley (1988b) analysed the distribution of 25 *Eriope* taxa from 13 localities scattered throughout the region. Many taxa had very restricted distributions and most of the sites had endemics which did not occur elsewhere. A marked disjunction was also noted between the Chapada Diamantina region in Minas Gerais and the northern end of the Serra do Espinhaço in Bahia. Only one species, *E. macrostachya* was found in both areas. This indicated a past barrier to migration roughly following the line of the present boundary between the states of Minas Gerais and Bahia. Some species were more widespread in campo rupestre, for example *E. hypenioides* was distributed throughout the campo rupestre and 'gerais' (treeless cerrado) areas of the Chapada Diamantina area. One species, formerly included in *Hyptis* section *Hypenia* subsection *Densiflorae, E. blanchetii,* was restricted to coastal restinga in Bahia.

Eriope sensu stricto was not speciose in cerrado. Only three species were described as 'typical' members of cerrado flora, *E. complicata* Mart. ex Benth., *E. xavantium* Harley *E. crassipes* (Harley 1988b).

ii) *Eriope* species formerly included in *Hypenia* sensu Harley (1988a)

The distribution of *Eriope* species which were formerly included in *Hypenia* sensu Harley (1988a) is from 5° N to 24° S in cerrado, campo rupestre and caatinga. Most species show restricted distributions but *E. reticulata, E. macrosiphon, E. salzmannii* are all widespread. *E. vitifolia* has a relatively wide distribution in the Serra do Espinhaço in eastern Brazil.

The distributions of species in section *Hypenia,* are closely correlated to the distribution of cerrado and most areas of cerrado support at least one species. They are conspicuously absent from northern Tocantins, although this area is very poorly explored botanically and it would not be surprising if species from section *Hypenia* were to be collected here.

The two subsections of section *Hypenia, Laxiflorae* and *Ellipticae,* contain groups of species which are largely allopatric in central Brazil. Only *E. reticulata* is partially sympatric with other species in subsection *Laxiflorae.* However, the ranges of the two subsections overlap. Field observations confirmed that *E. macrantha* and *E. reticulata* occupied the same localities in the Distrito Federal area, *E. niquelandiensis* and *E. aristulata* occurred together near Niquelândia and *E. calycina* and *E. aristulata* were sympatric in western parts of the Chapada dos Veadeiros. The different pollination biology of the two subsections offers an explanation for the occurence of sympatric pairs of species. The resupination of the corolla of species of subsection *Ellipticae* indicates hummingbird pollination in this group whilst the species in subsection *Laxiflorae* are likely to be bee pollinated (see appendix II). This may provide an
effective barrier to pollen transfer between sympatric species.

E. reticulata and *E. macrosiphon*

E. reticulata is primarily a species of eastern and central Brazil but is also found in southern Brazil and Paraguay. It is the most widespread and taxonomically problematic species in section *Hypenia.*The existence of closely related species, e.g. *E. paniculata,* within its range and the prevalence of geographically localised variants indicates that *E. reticulata* may be a paraphyletic species. In this case it appears that tokogenetic relationships are largely restricted within populations. However, the process of isolation does not appear to be complete, hence the high degree of overlapping variation within the species. *E. macrosiphon* is a similarly wide ranging species but it is confined to western Brazil and extends into Bolivia. Although it is a variable taxon and geographical variants can be identified, the correlation between variation and geography is not as well-defined as it is for *E. reticulata.* This may reflect the lack of on the more continuous nature of cerrado in central and western Brazil. It seems to be a relatively uncommon species of cerrado and is found in campo rupestre in Chapada dos Guimarães, Mato Grosso.

E. reticulata occurs both in cerrado and sometimes in campo rupestre. However, habitat of either type is relatively fragmented in southeastern Brazil, both as a result of human activity and because of the mountainous topography of the region. This habitat fragmentation may help to explain the pattern of geographical variation in *E. reticulata.* Human activity in the region is relatively recent and may not have allowed sufficient time for population differentiation. However, Harley's (1995b) suggestion that campo rupestre may have been more widespread in the Pleistocene could offer an explanation for the wide geographical

distribution, and current fragmentation of populations corresponding to the fragmentation of campo rupestre in southeastern Brazil. Cerrado on the central plateau of Brazil has also been fragmented by the presence of deciduous and semideciduos forest on better soils in the cerrado region (Oliveira-Filho & Ratter 1995) and by the presence of gallery forests where the water table is high. This habitat fragmentation could also offer an explanation for the geographically correlated variation observed in *E. reticulata* in the cerrados of central Brazil. The isolation is by no means complete however, hence the lack of clearly defined discontinuities.

E. vitifolia

According to the morphological and molecular analyses, *E. vitifolia* is sister taxon to most species of *Eriope,* including section *Eriope* and section *Hypenia.* This suggests that the latter two sections evolved from a *E. vitifolia*-like ancestor. In this context it is interesting to note that the current distribution of *E. vitifolia,* in campo rupestre throughout the Serra do Espinhaço range of mountains, is sympatric with the distribution of the majority of species of section *Eriope* but is not sympatric with any species of section *Hypenia.* This could either indicate that *E. vitifolia* (or something like it) gave rise the ancestor of section *Eriope* and neither taxa have dispersed far since that event, or simply that it shares a similar habitat requirement. Nevertheless, the latter possibility does imply the possibility of shared ecological adaptation which may be the result of a close evolutionary relationship between *E. vitifolia* and section *Eriope*.

Habitat specificity and distribution of *Eriope* species

There is clearly a strong correlation between habitat and the distribution of *Eriope* species. However, in most cases the correlation can only be described in broad terms, e.g. section *Eriope* is most diverse in campo rupestre and section *Hypenia* in cerrado. As a result it is difficult to explain how the distribution of individual species can be correlated with habitat specificity under current conditions and explanations based on historical fragmentation of habitat are required (see the discussion under *E. reticulata*). There is little information available on detailed habitat specificity for most of the species formerly included in *Hypenia.* However, some information is available. *E. salzmannii* has a ruderal habit and is found in caatinga and restinga (coastal sand-dune vegetation) as well as campo rupestre. *E. niquelandiensis* has been collected from serpentine rocks and *E. densiflora* appears to be restricted to the narrow band of cerrado on the margin of gallery forest. *E. densiflora* occurs in the same localities as *E. brachystachys* and habitat differentiation may explain the

sympatric distribution of these species. However, *E. densiflora* and *E. brachystachys* may not be as closely related as some aspects of their morphology (notably inflorescence structure) suggest. Neverthess, these observations suggest that, in these species at least, contemporary ecological requirements are important in determining geographical distribution.

Dispersal in *Eriope*

Dispersal mechanisms in *Eriope,* would only rarely allow for long-distance transport. The nutlets are relatively heavy and the myxocarpic nutlets, which are produced just as the rains start, would adhere very effectively to wet soil. It therefore seems unlikely that *Eriope* species would be able to disperse rapidly over a large area (but see *E. salzmannii* below). This perhaps helps to explain the restricted distribution of most of the species covered in this account although the picture is complicated by the widespread distribution of *E. reticulata* and *E. macrosiphon.* However, the effects of isolation in maintaining variation in *E. reticulata* may be accentuated by relatively poor dispersal ability. *E. salzmannii* is the most widespread species in *Eriope* and its long-distance dispersal into Venezuela suggests it has better dispersal abilities than other related species. It is notable in this context that *E. salzmannii* has small and light nutlets.

Endemism in Goiás and Distrito Federal

Figure 11-2 shows the approximate distributions of all the species covered in this account. A striking feature of the distributions mapped is their concentration in a relatively small area. 18 of the 23 species covered in this account occur in Goiás, Distrito Federal and neighbouring parts of Bahia and Minas Gerais. All three species of section *Densiflorae,* four species of section *Hypenia* subsection *Laxiflorae* and all six species of section *Hypenia* subsection *Ellipticae* are confined to the area. In addition *E. reticulata, E. macrosiphon, E. vitifolia, E. irregularis* and *E. gracilis* all occur in the region. Section *Eriope* is not so well represented in here but *Hyptis* also has high levels of endemism, particularly in the Chapada dos Veadeiros, about 200 km north of Brasília, e.g. *Hyptis pachyphylla* Epling and *H. cruciformis* Epling, both in section *Pachyphyllae* (Epling) Harley, are restricted to this area.

It is not clear why there should be such high levels of endemism in this group in such a small area of central Brazl. However, there does seem to be an association with the distribution of tree species since northern Goiás and neighbouring areas roughly correlates with the Central Area identified by the vegetation analysis of Ratter et al. (1996). It may be that this represents the central core of cerrado and the vegetation has remained relatively stable for

much longer than areas adjoining over vegetation types such as *hylaea* or caatinga. If this is the case it suggests that those *Eriope* species which are found here are strongly associated with cerrado.

Cerrado has characteristically poor soils with mineral contents which are not amenable to plant growth (Furley & Ratter 1988) and soil may be a strong factor in determining the distribution of these species. However, *E. macrantha, E. reticulata, E. brachystachys* and *E. densiflora* were all grown in the Jodrell glasshouses at Kew and did not appear to have unusual nutrient or substrate requirements. They were apparently most sensitive to humidity and could not tolerate prolonged moist atmospheric conditions. This indicates that they could not tolerate forest conditions but clearly does not provide a complete explanation for the preference for cerrado. Further ecological investigation is required to clarify this. Historical processes of habitat fragmentation and poor dispersal ability may account for the high numbers of taxa, both at the species and sectional level, in this area of central Brazil.

D. Biogeographic conclusions

The distribution of *Eriope* species formerly included in *Hypenia* is strongly correlated with the distribution of cerrado and to a lesser extent with campo rupestre. It is possible to postulate some mechanisms to explain the existence of sympatric species in cerrado, for example pollination biology in section *Hypenia* has apparently provided an important means of maintaining sympatric groups of species. In addition, habitat specificity of some species appears to maintain differentiation of sympatric taxa, e.g. *E. densiflora* and *E. brachystachys*. In other cases, historical processes of habitat fragmentation coupled with relatively poor dispersal abilities appear to offer the most plausible explanation for species distributions in the species discussed. However, this does not explain why some species have widespread distributions and others are much more restricted.

Biogeographical patterns in the cerrado region are poorly known and further accounts of taxa with high levels of endemism in the area are required to help understand the influence of geography on biodiversity.

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Appendix I: Harley's provisional classification of the Hyptidinae

***Hyptidendron* Harley** (18 species)

1. Section *Umbellaria* (Benth.) Harley
(syn. *Hyptis* sect. *Umbellaria* Benth.)

glutinosum (Benth. in DC.) Harley
unilaterale (Epling) Harley
torrendii Harley ined. (N. E. Brazil)
amethystoides (Benth. in DC.) Harley
caudatum (Epling & Jativa) Harley
vauthieri (Briq.) Harley
vepretorum (Mart. ex Benth.) Harley
rhabdocalyx (Mart. ex Benth.) Harley
arbusculum (Epling) Harley
dictiocalyx (Benth. in DC.) Harley
clausennii (Benth. in DC.) Harley
rondonicum (Harley) Harley
sp. nov. A (Mato Grosso)

2. Sect. *Hyptidendron* Harley
(syn. *Hyptis* sect. *Buddleoides* Benth.)

asperrimum (Sprengel) Harley
arboreum (Benth. in DC.) Harley
canum (Pohl ex Benth.) Harley
conspersum (Benth. in DC.) Harley
leucophyllum (Pohl ex Benth.) Harley

***Eriope* Kunth** (36 species)

1. Sect. *Nudicalyx* sect. nov. ined.

latifolia (Mart. ex Benth.) Harley
ssp. *latifolia*
ssp. *heterantha* (Benth. in DC.) Harley ined.
orbignyi Harley sp. nov. ined.
hypoleuca (Benth. in DC.) Harley
exaltata Harley
salviifolia (Pohl ex Benth.) Harley
sincorana Harley
blanchetii (Benth.) Harley
confusa Harley
luetzelbergii Harley
machrisae (Epling) Harley
simplex (St. Hil. ex Benth.) Harley

2. Sect. *Eriope*

macrostachya Mart. ex Benth.
var. *macrostachya*
var. *hypoleuca* Benth.
var. *grandiflora* Epling
var. *platyantha* (Epling) Harley
velutina Epling
alpestris Mart. ex Benth.
foetida Pohl ex Benth.
parvifolia Mart. ex Benth.
glandulosa (Harley) Harley
meninae ined.
filifolia Benth.
angustifolia Epling
hypenioides Mart. ex Benth.
? 3 subspp.
polyphylla Mart. ex Benth.
crassifolia Mart. ex Benth.
tumidicaulis Harley
xavantium Harley
crassipes Benth.
subsp. *crassipes*

subsp. *trichopoda* (Briq.) Harley

subsp. *cristalinae* Harley

'*pseudocrassipes*' from Bahia

obovata Epling

var. *obovata*

var. *gracilis* Harley

arenaria Harley

complicata Mart. ex Benth.

anamariae Harley

montana Harley

+ 9 additional species (unpublished).

***Eriopidion* Harley** (1 species)

strictum (Benth.) Harley

***Hypenia* (Mart. ex Benth.) Harley** (25 species)

(syn. *Hyptis* sect. *Hypenia* Mart. ex Benth.)

1. Sect. *Hypenia*

(syn. *Hyptis* sect. *Hypenia* subsect. *Densiflorae* (Benth. in DC.) Epling)

densiflora (Pohl ex Benth.) Harley

inelegans (Epling) Harley

pruinosa (Pohl ex Benth.) Harley

irregularis (Benth. in DC.) Harley

brachystachys (Pohl ex Benth.) Harley

paradisi (Harley) Harley

marifolia (Benth. in DC.) Harley

concinna (Benth. in DC.) Harley

2. Sect. *Laxiflorae* (Benth. in DC.) Harley

(syn. *Hyptis* sect. *Hypenia* subsect. *Laxiflorae* (Benth. in DC.) Epling)

aristulata (Epling) Harley

perplexa (Epling) Harley

reticulata (Mart. ex Benth.) Harley

glauca (St. Hil. ex Benth.) Harley

calycina (Pohl ex Benth.) Harley

crispata (Pohl ex Benth.) Harley

durifolia (Epling) Harley

macrantha (St. Hil. ex Benth.) Harley

gardneriana (Epling) Harley

macrosiphon (Briq.) Harley

pauliana (Epling) Harley

paniculata (Benth.) Harley

subrosea (Harley) Harley

3. Section *Vitifoliae* Harley sect. nov. ined.

vitifolia (Pohl ex Benth.) Harley

longicaulis Harley ined.

micrantha (Benth. in DC.) Harley

salzmannii (Benth.) Harley

***Hyptis* Jacq.** (277 spp.)

1. Section *Subumbellaria* Epling

asperifolia Standl.

hagei Harley

2. Section *Rhytidea* Epling

rhytidea Benth.

pseudolanata Epling

3. Section *Umbellatae* (Epling) Epling

tafallae Benth.

subtilis Epling
iodantha Epling

4. Section *Laniflorae* Epling
tomentosa Poit.
decipiens M. E. Jones
tephrodes Gray
albida Kunth
emoryi Torrey
anitae Epling & Jativa
laniflora Benth.

5. Section *Minthidium* Benth.
trichodes Epling
elegans (Briq.) Briq.
fasciculata Benth.
thrysiflora Epling
sp. nov. (Rio de Janeiro)
scandens Epling
mixta Epling
chyliantha Urb. & Ekman
cubensis Urb.
verticillata Jacq.
domingensis Urb.
rivularis Britton
escobilia Urb.
scoparioides Urb.
americana Briq.

6. Secion *Latiflorae* Epling
eximia Epling

7. Section *Mesosphaeria* Benth.

7a. *Spicaria* (Benth.) Epling
spicigera Lam.
stricta Benth.

7b. *Plectranthodon* Epling
plectranthoides Benth.
7c. *Pectinaria* (Benth.) Epling
suaveolens (L.) Poit.
argutifolia Epling
oblongifolia Benth. in DC.
arborescens Epling
heterodon Epling
collina Brandegee
macrotera Briq.
racemulosa Mart. ex Benth.
propinqua Epling
multiseta Benth. in DC.
pectinata (L.) Poit.
septentrionalis Epling
urticoides Kunth
diffusa Epling

7d. *Ocimoideae* Epling
cymulosa Benth. in DC.

7e. *Eriocephalae* (Epling) Epling
umbrosa Salzm. ex Benth.
gymnocaulos Epling
chacapoyensis Briq.
pilosa Benth.
pseudoglauca Epling
purdiaei Benth. in DC.
eriocephala Benth. in DC.
irwinii Harley
obtusata Benth.
marrubifolia Epling & Mathias

diversifolia Benth.
melissoides Kunth

8. Section *Trichosphaeria* Benth.

8a. *Plumosae* Epling (2 species)
plumosa Benth. in DC.
mollissima Benth.

8b. *Crinitae* Epling
eriophylla Pohl ex Benth.
carvalhoi Harley
simulans Epling
martiusii Benth.
multiflora Pohl ex Benth.
crinita Benth.

9. Section *Hilaria* Epling
lobata St. Hil. ex Benth.

10. Section *Polydesmia* Benth.

10a *Oocephalus* Epling
crassifolia Mart. ex Benth.
piranii Harley
lacunosa Pohl ex Benth.
nubicola Harley
argyrophylla Harley
halimifolia Mart. ex Benth.

10b. *Tubulosae* Briq.
macrostachys Benth. in DC.
pinheiro Harley
calida Mart. ex Benth.
siphonantha Harley
leptostachys Epling

10c. *Glomeratae* Benth.
lythroides Pohl ex Benth.
subrotunda Pohl ex Benth.
nivea Epling
indivisa Pilger
petraea St. Hil. ex Benth.
foliosa St. Hil. ex Benth.
silvinae Harley
glomerata Mart. ex Schrank

10d. *Rigidae* Benth. in DC.
carpinifolia Benth.
violacea Pohl ex Benth.

10e. *Malvastra* Epling
althaeifolia Pohl ex Benth.
duplicato-dentata Pohl ex Benth.

10f. *Vulgares* Benth. in DC.
impar Epling
pinetorum Epling
similis Epling
erythrostachys Epling
villicaulis Epling
rubicunda Pohl ex Benth.
dubia Pohl ex Benth.
colombiana Epling
muricata Schott ex Benth.
mutabilis (L .Rich.) Briq.

11. Section *Fruticosae* Sect. nov.

fruticosa Salzm. ex Benth.
cuniloides Epling
brightonii Harley ined.

12. Section *Myriocephala* Benth. in DC.
odorata Benth.

13. Section *Leucocephala* Epling
leucocephala Mart. ex Benth.
stachydifolia Epling
elongata Benth.

14. Section *Plagiotis* Benth.
laciniata Benth.
uliginosa St. Hil. ex Benth.
eriocauloides Rich.

15. Section *Cyrta* Benth.

15a *Tomentosae* Benth. in DC.
lavandulacea Pohl ex Benth.
divaricata Pohl ex Benth.

15b *Paludosae* Epling
microphylla Pohl ex Benth.
paludosa St. Hil. ex Benth.
microsphaeria Epling

15c. *Tetragonae* Benth.
recurvata Poit.
lagenaria St. Hil. ex Benth.
dumetorum Morong

16. Section *Cyanocephalus* Pohl ex Benth.
peduncularis Benth.
nitidula Benth. in DC.
tenuifolia Epling
selaginifolia Mart. ex Benth.
capriarifolia Pohl ex Benth.
digitata Harley
tagetifolia Harley
tripartita Briq.
delicatula Harley
viatica Harley
rugosa Benth.
desertorum Pohl ex Benth.
apertiflora Epling
pedalipes Griseb.
bombycina Epling
polioides Briq.
albicoma Epling
cretata Epling
coriacea Benth. in DC.
lippioides Pohl ex Benth.
lanata Pohl ex Benth.
cardiophylla Pohl ex Benth.
adpressa St. Hil. ex Benth.
quadrangularis Glaz. ined.
cuneata Pohl ex Benth.

17. Section *Gymneia* Benth.
virgata Benth.
sp. nov. (Veadeiros)
ovalifolia Benth. in DC.
interrupta Pohl ex Benth.
platanifolia Mart. ex Benth.
malacophylla Benth. in DC.
ampelophylla Epling

18. Section *Muellerohyptis* Briq.

pulchella Briq.

muelleri Briq.

pachyartha Briq.

huberi Harley

19. Section *Hyptis* Jacq.

19a. *Lavandulaceae* Benth.

linarioides Pohl ex Benth.

caespitosa St. Hil. ex Benth.

19b. *Marrubiastrae* Benth.

colubrimontis Epling & Jativa

pyriformis Epling & Jativa

ssp. *pyriformis*

ssp. *yutajeensis* Harley ined.

mollis Pohl ex Benth.

sinuata Pohl ex Benth.

pseudosinuata Epling

balansae Briq.

angulosa Schott ex Benth.

lappulacea Mart. ex Benth.

lappacea Benth.

longifolia Pohl ex Benth.

ramosa Pohl ex Benth.

involucrata Benth.

lanceolata Poir.

lanceifolia Schum.

brevipes Poit.

vilis Kunth & Bouche

guanchezii Harley

inodora Schrank

atlantica Harley ined.

personata Epling

intermedia Epling

lacustris St. Hil. ex Benth.

obtusiflora Presl. ex Benth.

brachypoda Epling

pachycephala Epling

parkeri Benth.

19c. *Hyptis*

lantanifolia Poit.

paupercula Epling

bahiana Harley ined.

minutifolia Griseb.

hygrobia Briq. (?)

conferta Pohl ex Benth.

var. *angustata* (Briq.) Pool & Harley

alata Raf.

ssp. *alata*

ssp. *rugosula* (Briq.) Harley

actinocephala Griseb.

savannarum Briq.

florida Benth.

macrocephala Mart. & Gal.

rhomboidea Mart. & Gal.

capitata Jacq.

mariannarum Briq.

petiolaris Pohl ex Benth.

lorentziana O. Hoffm.

luticola Epling

havanensis Britton ex Epling

armillata Epling

shaferi Britton

20. Section *Ammophila* Harley sect. nov. ined.

ammotropha Wright ex Griseb.

leptoclada Benth. in DC.
lanuginosa Glaz. ex Epling
passerina Mart. ex Benth.
gardneri Briq.

Subgenus *Stylopodiferae* (stylopodium present)

21. Section *Eriosphaeria* Benth.

21a. *Sessilifoliae* Benth. in DC.
pycnocephala Benth. in DC.
asteroides St. Hil. ex Benth.
ovata Pohl ex Benth.
crenata Pohl ex Benth.
dilitata Benth. in DC.
lanicephala Epling & Jativa
turnerifolia Mart. ex Benth.
hamatidens Epling & Jativa
alpestris St. Hil. ex Benth.

21b. *Velutinae* Benth. in DC.
origanoides Pohl ex Benth.
velutina Pohl ex Benth.
adamantium St. Hil. ex Benth.
hilarii Benth.
colligata Epling & Jativa
rhypidiophylla Briq.
saxatilis St. Hil. ex Benth.
angustifolia Pohl ex Benth.

21c. *Graciles* Epling
arenaria Benth. in DC.

21d. *Passerinae* Benth. in DC.

21e. *Obtectae* Epling
obtecta Benth. in DC.

21f. *Gnidifoliae* Benth.
ditassoides Mart. ex Benth.
deltifolia Epling & Jativa
imbricata Pohl ex Benth.

21g. *Heterophyllae* Epling
dictyodea Pohl ex Benth.
heterophylla Benth. in DC.

22. Section *Pachyphyllae* (Epling) Harley
cruciformis Epling
penaeoides Taub.
pachyphylla Epling
imbricatiformis Harley

23. Section *Induratae* Epling
monticola Mart. ex Benth.
corymbosa Benth. in DC.
rotundiolia Benth.
tricephala St. Hil. ex Benth.
complicata St. Hil. ex Benth.
lucida Pohl ex Benth.
proteoides St. Hil. ex Benth.
viminea Epling
xanthiocephala Mart. ex Benth.

24. Section *Xylodontes* (Benth.) Epling

24a. *Paniculatae* Epling
nigrescens Pohl ex Benth.
salicina Schmidt in Mart.
rubiginosa Benth.
amaurocaulos Briq.
lutescens Pohl ex Benth.
brachiata Briq.
fulva Epling
remota Pohl ex Benth.
ferruginosa Pohl ex Benth.
villosa Pohl ex Benth.
frondosa S. Moore
orbiculata Pohl ex Benth.
alutacea Pohl ex Benth.
subviolacea Briq.
fallax "confertoides" or ? sp. nov.

24b. *Macilenta* Epling
rondonii Epling

24c. *Axillares* Benth. in DC.
crassipes Epling
argentea Epling & Mathias
marrubioides Epling
tumidicalyx Epling & Jativa ? ined.
homalophylla Pohl ex Benth.
hirsuta Kunth
uncinata Benth.

25. Section *Apodotes* Benth.
multibracteata Benth.
australis Epling
pulegioides Pohl ex Benth.
loseneriana Pilg.
tetragona Pohl ex Benth.
sericea Benth.
hassleri Briq.
nudicaulis Benth.

26. Section *Pusillae* Epling
atrorubens Poit.
hispida Benth. in DC.
humilis Benth. in DC.
caduca Epling

***Marsypianthes* Mart. ex Benth.** (ca. 7 species)
chamaedrys (Vahl) Kuntze
montana Benth.
hassleri Briq. (dubious)
burchellii Epling
foliolosa Benth. in DC.
formosa sp. nov.

***Peltodon* Pohl** (5 species)
reptans Pohl (= *repens* (Vell.) Kuntze)
tomentosus Pohl
rugosus Tolm.
pusillus Pohl
longipes St. Hil. ex Benth.

***Raphiodon* Schau** (1 species)
echinus (Nees & Mart.) Schau.

Affinity uncertain:
***Asterohyptis* Epling** (3 species)

Appendix II: Observations on pollination in *Hypenia*

The species names used in the following account use *Hypenia* as the generic epithet. Field work for studies on floral biology was conducted during two visits to Brazil. The first was to Distrito Federal in Central Brazil, with field work at the Reserva Ecológica do IBGE just outside Brasília, where *Hypenia macrantha* sensu stricto and *H. reticulata* were studied. The work was conducted over three weeks in late August to early September 1995.
The second trip was to southern Bahia, with field work based in Rio de Contas, working with *H. salzmannii* and *H. vitifolia* over a three week period in late February to early March 1996.

General notes on flowering time, floral and inflorescence morphology in relation to pollination, phenology and flower visitors are given below for each species studied in the field and figure II-1 is a graphical summary of phenology in *H. salzmannii.* Nectar concentration and volume were also measured for some of these species and the results are presented in table II-1.

General observations on phenology in *Hypenia*

Hypenia species produce their flowers seasonally. Observations from herbarium specimens indicate that species from the Central Brazilian cerrado region, *H. macrantha, H. reticulata, H. brachystachys* and *H. densiflora,* flower and fruit in the latter half of the dry season which occurs between June and October in Central Brazil. *H. vitifolia* and *H. salzmannii* flower in Northeastern Brazil during the early part of the dry season in February to April.

All *Hypenia* species observed displayed clearly defined protandry with the explosive male stage preceding the female stage by several hours. Prior to anthesis the anthers are held under tension and are explosively released if the corolla mouth is subject to physical disturbance. On triggering of the anterior lobe the anthers spring up, releasing a small cloud of pollen against the visitor. This corresponds to the male stage. The filaments then extend to hold the anthers out of the mouth of the corolla. The flower is now in its neutral stage which lasts as the style elongates down the corolla tube until the stigma is exserted out of the corolla mouth. When the bifid stigma appears the flower enters the female phase, after which, depending on the success of fertilisation, the corolla together with the style and anthers absciss leaving the ovary inside the calyx to mature into ripe nutlets or to absciss at a later stage. This general

pattern is found in all species of *Hypenia* studied, differing in details of timing and with constraints imposed by developments of the corolla.

Phenology and flower visitors for individual species

H. reticulata

This species was widespread and common throughout the IBGE reserve in all types of cerrado, from open grassland to closed canopy woodland. The corolla tubes were 11 to 13 mm long and red to pink. Flowers were regularly distributed in a lax inflorescence and the corolla throat usually pointed downwards. The inflorescence was 1 - 2 m tall and borne on virgate, waxy stems.

Phenology

Each plant of *H. reticulata* had between two and 30 open flowers at any one time and all stages of floral development were present on each individual. According to herbarium specimens the flowering period of populations extends for about three months and by early September the majority of individuals were coming to the end of their flowering period.

130 flowers were tagged as buds before 10:00 h and 14 buds tagged at 16:00 h. For buds tagged in the morning the corolla lobes were open within 2 hours of tagging and within 6 hours most were triggered and a few had already dropped. Buds tagged in the afternoon were all open and triggered by 10:00 h the following morning. The style was exserted from the corolla within 6 hours of triggering of the anterior lobe and the corolla persisted with the style exserted for about 24 hours before dropping. Extension of the style was observed to occur independently of corolla lobe triggering, in which case the style projected through the anterior lobe. The style rarely persisted after the corolla had dropped. All the corollas had abscissed within 48 hours of opening and about half had abscissed within 24 hours.

The corolla of this species went through distinct colour changes from red in bud to pink or apricot in the female phase.

Flower visitors

H. reticulata was seen to be visited on several occasions by large, xylocopid bees and was occasionally visited by the hummingbird *Amazilia fimbriata.*

H. macrantha

H. macrantha occurred in a localised population of about 50 individuals in the IBGE reserve in open grassland with few trees or shrubs. It was observed in the neighbouring Água Limpa experimental farm in a similar habitat but was uncommon in the Distrito Federal area. The corolla tubes were 23 to 27 mm long and bright red. The inverted flowers were held towards the end of the inflorescence branches and were held horizontally or pointed upwards. Inflorescences were one to three metres tall and borne on virgate, waxy stems.

Phenology

Each plant of *H. macrantha* had between two and 10 open flowers at any one time and all stages of floral development were present on each individual. According to herbarium records the flowering period of this species extends for about 3 months beginning in June to. In early September the majority of individuals were still flowering but all had maturing fruit present.

83 buds were tagged, at 7:30 h, 9:30 h and 17:00 h. The corollas usually opened within 2 or 3 hours of being marked regardless of the time and persisted in the untriggered state until they were visited. This time could vary from a few minutes to over a day depending on visitor activity. Buds tagged in the evening were mostly triggered by 9:45 am the following day indicating that visitation is highest in the early morning and evening. This was confirmed by direct observation of hummingbirds visiting this species. Elongation of the style and opening of the stigmatic lobes took from three to eight hours. Once the stigma was exposed the corolla could persist for about a further 24 hours before it dropped, sometimes leaving the withered style behind although this frequently dropped with the corolla. Corollas occasionally persisted in the untriggered state for up to two days before dropping. The corolla of this species remains the same bright red colour throughout its life.

Flower visitors

H. macrantha occurred in a localised population on the IBGE reserve which was regularly patrolled by a species of hummingbird common in cerrados of the Distrito Federal region, *Amazilia fimbriata*. This is a small hummingbird, weighing approximately 4 g and with a beak length of ca. 35 mm. One individual regularly visited flowers of *H. macrantha* at the studied population. This individual was highly territorial and vigorously defended its territory from intruders. The flowers were visited throughout the day but activity was usually highest in the early morning and evening, possibly associated with a drop in the wind. On

several windy days there was a decrease in hummingbird visits to the flowers. The long, slender stems swayed violently in the wind which presumably made foraging difficult.

One xylocopid bee was caught visiting this species. The mouthparts of this bee are too small to be able to extract nectar from the throat of the corolla It may have been responsible for causing the holes observed through the calyx and corolla near the nectary of some flowers of *H. macrantha.*

Hypenia densiflora and *H. brachystachys*

The corolla of both species was tubular, 5 to 12 mm long and white to lilac. The flowers were held in densely clustered heads in inflorescences 1.5 - 2.5 m tall. Both species had virgate, waxy stems.

Phenology

The phenology of *H. brachystachys* and *H. densiflora* was not studied in detail but a few observations can be made. Like the other species of *Hypenia* in Central Brazil they flower over a period of two or three months at the end of the dry season. There were from three to 50 flowers which were open and in the male, neutral or female phase at any one time in each individual.

Flower visitors

Two large xylocopid bees were caught on *H. brachystachys* and similar bees were observed on *H. densiflora.*

H. vitifolia

H. vitifolia was studied in one population of ca. 40 individuals in disturbed cerrado, Rio de Contas, Bahia. It was observed in other localities in Bahia in populations of ca.100 individuals. The large inflorescence was at 1 - 1.5 m and was borne on virgate, waxy stems with large swellings. The corolla was about 4 mm long, yellowish brown in bud and purple when open, the tube was flared. The style was exserted ca. 5 mm out of the corolla before the very small (< 1 mm) stigmatic lobes were exposed.

Phenology

Individual plants had from two to more than 100 open flowers at any one time. The middle bud in a cyme was followed as individual flowers were difficult to mark. This made

assessment of fruit set from tagged flowers difficult as there were often several mature calices per cyme and individual flowers were not identified.

30 cymes were tagged at 17:30 h and most of them were still in bud by 9:00 h the following morning. After this they followed much the same pattern as that for *H. salzmannii* (see below) and by 17:40 h, 24 hours after tagging, 19 had dropped and 8 were in the female phase. The remainder were triggered. By the next morning all the corollas had dropped. Colour differences between the bud and corolla occurred as the bud went from brownish yellow to purple as the corolla lobes opened.

Flower visitors

H. vitifolia was constantly visited throughout the day by a range of small bees, flies and, on one occasion, a butterfly was observed visiting it (plate V b) and *H. salzmannii*. Occasional observations were made of a small wasp feeding on the untriggered lobe of some flowers. This species was also a very common visitor to open flowers of *H. vitifolia* but like the butterfly may have been too fragile to be an effective pollinator.

Three species of bee feeding on *H. vitifolia* were identified from photographs (O'Toole, Hope Entomological Collections, University Museum, Oxford)
Paratetrapedia sp. female.
Solitary ground-nesting, oil-collecting bees, family Anthophoridae. Plate V d.
Trigona sp., worker.
Stingless bees, family Apidae. (not shown)
Apis mellifera, worker.
Honeybee, family Apidae. Plate V f.

H. salzmannii

H. salzmannii was common in the Rio de Contas area and tended to be weedy in open disturbed areas with large, widespread populations. The inflorescence was large and borne on virgate, waxy stems 0.5 - 1.5 m tall. The corollas were yellow in bud changing to sky blue as the lobes opened.

Phenology

Each plant had between two and more than 100 open flowers. Individuals flowered throughout the observation period of 3 weeks and had been in flower for some time before. It

seems likely that they flower continuously for two to three months in the period after the rains finish. Early 1996 was exceptionally dry but flowering did not seem to be adversely affected.

64 buds were tagged at 9:00 h, 12:00 h and 17:00 h. Buds tagged at 17:00 h were all triggered by 9:00 h the following morning. The exact time taken to be triggered depended on when the flower was first visited but they had almost all been visited 1 to 4 hours after opening. The style was exserted two to three hours after triggering and the corolla dropped four to six hours later.

Flower visitors

H. salzmannii was visited by a number of insects including a small butterfly, but most commonly by a range of small bee species. The butterfly perched on the upper side of the corolla tube and inserted its proboscis above the anthers so it seems unlikely that it would trigger the lower lip of the corolla and thus was probably visiting the plant for nectar without pollinating the flowers. Fewer visitors were observed visiting *H. salzmannii* than were seen on the neighbouring *H. vitifolia.*

Distribution of floral stages in *H. salzmannii* and *H. vitifolia*

The total number of flowers and fruit at different stages were counted between 8:00 h and 12:00 h on 10 stems of *H. salzmannii* and 10 stems of *H. vitifolia* and the results summarised in figure II-1.

Figure II-1: Floral stages in *H. salzmannii* and *H. vitifolia*

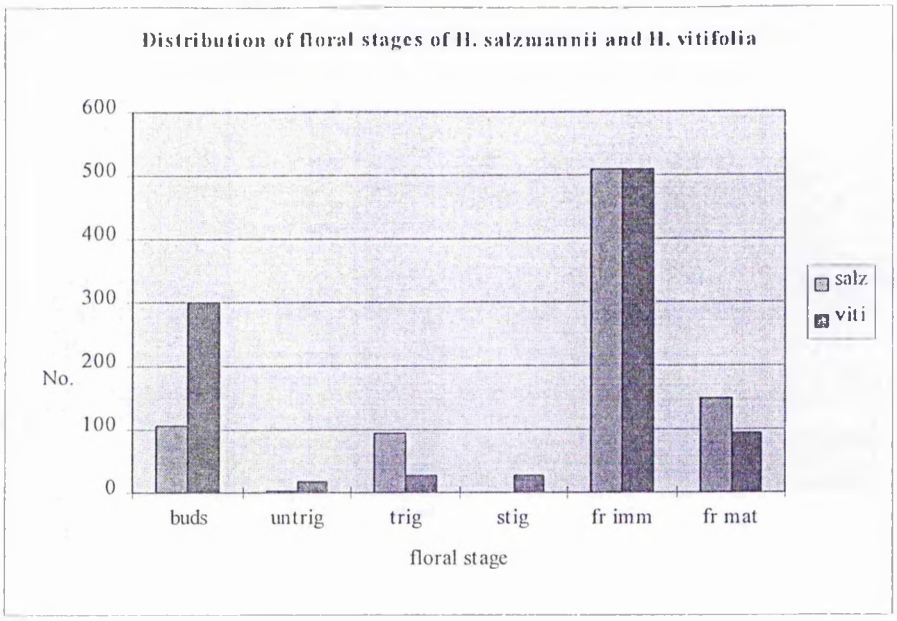

Figure II-1 shows the distribution of floral stages in *H. salzmannii* and *H. vitifolia* and illustrates the small number of open flowers (i.e. those which are untriggered, triggered or with the stigmatic lobes showing, corresponding to the pre-male, neutral and female stages) relative to those still in bud or forming immature fruit.

Nectar volume and composition

Nectar volume and sucrose concentration were assessed for *H. reticulata* and *H. macrantha* and limited measurements of nectar production were made for *H. brachystachys*. All measurements were from unbagged flowers and therefore reflect the standing crop. Nectar was collected in 10 µl and 5 µl micropipettes and sucrose concentration was assessed with a hand-held refractometer. Nectar volume and concentration for *H. salzmannii* and *H. vitifolia* was not measured because of difficulties in extracting nectar from their small flowers.

Table II-1: Volume and sucrose concentration of nectar from untriggered and triggered flowers of *H.macranha, H. reticulata* and *H. brachystachys*

Species	Mean nectar volume (µl)		Sucrose concentration (%)	
	Untriggered flowers	Triggered flowers	Untriggered flowers	Triggered flowers
H. macrantha	3.09 (n = 57) (0 - 10)	1.22 (n=46) (0 - 9)	19 (n = 26) (16 - 23)	18 (n = 10) (14 - 23)
H. reticulata	1.22 (n = 49) (0 - 4.8)	0.5 (n=4) (0 - 0.5)	23.4 (n = 27) (21 - 26)	-/-
H. brachystachys	0.11 (n = 21)*	-/-	-/-	-/-

* This figure is calculated from a single cumulative measurement taken from 21 flowers.
-/- Volume too small to measure concentration.

The volume and viscosity of nectar produced by flowers has been correlated with the requirements of floral visitors (Stiles 1981). Energy content is derived from total sugar content and is dependent on the total volume of nectar. According to Stiles (1981) high nectar viscosity is associated with high sucrose concentrations. Typically, hummingbird pollinated flowers produce large volumes of nectar with a relatively low sucrose content and bee pollinated flowers produce smaller volumes of higher viscosity nectar.

The results in table II-1 show that both untriggered and triggered flowers of *H. macrantha* produced higher volumes of nectar with a lower sucrose content than the nectar produced by flowers of *H. reticulata.* The volume and sucrose concentration produced by flowers of *H. macrantha* suggest that hummingbirds feed from these flowers. *H. reticulata* has more viscous nectar which offers a more appropriate reward for bees. *H. brachystachys* produces small volumes of nectar and the densely clustered flowers provide a platform for bees to crawl on so they can feed from several flowers without having to expend energy flying between them. Field observations of animals visiting these three species support the scenario that *H. reticulata* and *H. brachystachys* are pollinated by large, xylocopid bees and that *H. macrantha* is pollinated by hummingbirds.

Appendix III: Morphological data matrix

Hypenia

1		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31
		Virgi	Uppi	Wax	Fist	Petil	Seto	Bran	Inde	Phyl	1st	Cinc	Brac	Anth	Int.	Flow	Flow	Flow	Cont	K lol	K lat	Post	Post	Post	Ante	C tu	C tul	C cc	C tul	C. in	Anth	Styl
1	Hypenia micrantha	1	0	0	0	1	0/1	1	0	0	1	0	1	1	0	0	1	0	0	0	0	0	0	0	0	0	1	1	0	0	1	0
2	Hypenia irregularis	1	0	0	0	1	1	0	0	1	1	0	1	1	0	0	0	0	1	0	0	0	0	0	0	1	0	1	0	0	1	0
3	Hypenia vitifolia	1	1	1	1	0	1	0	0	1	1	0	0	2	0	0	1	0	0	0	0	0	0	0	0	1	1	0	0	0	1	0
4	Hypenia salzmannii	1	1	1	1	0	1	0	0	1	1	0	1	1	0	0	1	0	0	0	0	0	0	0	1	0	1	0	0	0	1	0
5	Hypenia densiflora	1	1	1	1	0	1	0	1	1	0/1	0	1	1	0/2	0	0	1	0	1	0	1	0	0	1	0	0	1	0	1	0	0
6	Hypenia brachystachys	0/1	1	0/1	1	1	1	0	1	1	0/1	0	1	1	0/2	0	0	1	0	1	0	1	0	0	1	0	1	0	0	0	1	0
7	Hypenia reticulata	1	1	1	0/1	0	1	0	0	1	1	0	1	1	0	0	1	0	0	1	0	0	0	0	1	0	2	2	1	1	1	0
8	Hypenia macrantha	1	1	1	0/1	0	1	0	0	1	1	0	1	1	0	1	1	0	0	1	0	0	0	0	1	0	2	1	1	1	1	0
9	Hypenia crispata	1	1	1	0/1	0	1	0	0	1	1	0	1	1	0	1	1	0	0	1	0	0	0	0	1	0	2	1	1	1	1	0
10	Eriope hypenioides	1	1	1	0/1	0	1	0	0	1	1	0	1	1	0	0	1	0	1	0	0	1	1	1	0	1	1	1	0	0	1	1
11	Eriope glandulosa	1	0	0	0	1	1	0	0	1	1	0	1	1	0	0	1	0	1	0	0	1	1	1	0	1	1	1	0	0	1	1
12	Eriope salviifolia	0	0	0	0	1	1	0	1	1	1	0	1	1	0	0	1	0	0	1	0	1	0	0	0	1	1	1	0	0	1	1
13	Eriope sincorana	1	1	0	0	0	1	0	0	1	1	0	1	1	0	0	0	0	0	0	0	1	0	0	0	1	1	1	0	0	1	1
14	Hyptidendron canum	0	0	0	0	0	1	0	0	1	1	0	0	0	1	0	0	0	0	1	0	1	0	1	1	0	0	1	0	0	1	1
15	Hyptidendron vepretorum	0	0	0	0	0	1	0	0	1	1	0	0	1	0	0	0	0	0	1	0	0	0	1	1	0	1	1	0	0	1	1
16	Hyptis asperifolia	0	0	0	0	0	1	1	0	1	1	0	0	1	0	0	0	0	1	1	0	0	0	1	1	0	0	1	0	0	1	0
17	Hyptis hagei	0	0	0	0	0	1	0	0	0	0	0	1	2	0	0	0	0	1	0	0	0	0	0	0	0	0	1	0	0	1	0
18	Hyptis rugosa	0	0	0	0	0	0	0	0	1	0	0	1	2	0	0	0	1	0	1	1	0	0	0	0	0	0	1	0	0	1	0
19	Hypenia subrosea	1	1	1	1	1	0	0	1	1	0	1	1	0	0	0	0	0	0	1	0	0	0	0	1	0	0	1	1	0	1	0
20	Hypenia gracilis	1	0/1	0	1	1	0	0	1	1	0	1	1	0	0	0	0	0	1	0	0	0	0	0	1	0	0/1	0	0	0	1	0
21	Hypenia aristulata	1	0/1	0/1	0	1	1	0	1	1	1	0	1	1	0	0	0	0	0	1	1	0	0	0	0	1	0	2	1	1	1	0

Appendix IV: Molecular protocols

a) Isolation of total cellular DNA, CTAB procedure

Modified from Doyle and Doyle (1987).

1. Preheat 20 ml of 2x CTAB isolation buffer in Oakridge tubes at 65°C. Add 80µl of beta mercaptoethanol. Preheat pestles and mortar to 65°C. Grind fresh or dried material in the preheated mortars adding a small amount of the CTAB mixture to produce a slurry. Pour slurry back into the tube and incubate at 60-65°C for 15-20 minutes with occasional gentle mixing.

2. Under the fume hood add equal volumes of SEVAG (24:1 chloroform:isoamyl alcohol) and gently mix for up to 30 minutes.

3. Spin at 8000 r.p.m. for 20 minutes at 20°C.

4. Remove aqueous top phase with a disposable pipette and transfer to an orange cap tube. Dispose of SEVAG and plant debris in SEVAG waste container.

5. Add 2.5 volume of cold (-20°C) absolute ethanol or 2/3 volume isopropanol and mix gently. Put in -20°C freezer for up to several weeks to precipitate the DNA.

6. Spin in centrifuge at 2000 r.p.m. for 2 to 5 minutes to collect precipitate. Pour off liquid after spin and add 70% ethanol to wash the pellet for 5 to 10 minutes.

7. Spin down DNA at 2000 r.p.m. for 5 to 10 minutes, pour off the alcohol and allow to evaporate for 5 to 10 minutes.

8. Resuspend the pellet in 3 ml of caesium chloride/ethidium bromide* mixture and store in the dark.

*Ethidium bromide is highly mutagenic and should be handled with care.

b) Caesium Chloride Gradient

1. Place DNA / CsCl suspension in ultra-centrifuge tube. Add more CsCl/Etbr mixture until the tube weighs between 7.9 and 8.1g.

2. Transfer tubes to ultra-centrifuge rotor, place plastic plugs over tube and screw on caps tightly.

3. Place rotor in ultra-centrifuge and set to run at hold (ie. until stop is pressed), at 20°C and 45 0000 r.p.m. overnight.

4. Stop the ultra-centrifuge, remove the rotor and tubes.

5. View tubes under ultra-violet light to check size and position of band, mark approximate position if band is faint. Record size of bands.

6. Using a 1000 µl micropipette, with tips trimmed to reduce chances of shearing the DNA, remove about ½ supernatant and discard. Remove the band in the ca. 1.5 ml of liquid, more may be necessary if the band is very faint, and place in a capped tube.

7. To each sample add 3 volumes of n-butanol suspended over a saturated sodium citrate buffer. Cap firmly and shake to mix solution. Lie on side and settle for 20-30 minutes.

8. While butanol is extracting the ethidium bromide remove dialysis tubing from 50% ethanol stored in the fridge and cut tubing into ca. 8 cm lengths. Clamp one end of the tubing.

9. Using a disposable pipette remove the colourless phase from the n-butanol mix and place in the prepared dialysis tubing. Discard n-butanol/ethidium bromide mix in waste bottle. Clamp top of tube.

10. If the DNA band was very faint concentrate the solution by placing the sample in sucrose for ca. 15 minutes, wash it and place in 4 litres of double distilled water with the other samples.

11. Leave in water for 4 hours, then transfer to 10X TE buffer for a further 4 hours. Change to fresh TE buffer for a further 4 hours.

12. Transfer sample to labelled 1.5 ml eppendorf tube. Run 5µl of sample on agarose gel to check DNA concentration.

13. Store DNA in eppendorf tube in the fridge at 4°C for immediate use, at -20°C for short term storage or -80°C for long term.

c) PCR reactions

1. Make a master mix containing per sample:

Sterile, double distilled water	73.5µl
Magnesium-free buffer	10
Magnesium	6
dNTPs	2
Taq	0.5
BSA	1
ITS 4 primer (400 ng/µl)	1
ITS 5 primer (400 ng/µl)	1
Total	**95µl**

2. Add the Taq last, vortex to mix ingredients and keep on ice. Aliquot 95µl of master mix into 0.5 ml eppendorf tube. Add 5 µl of template DNA.

3. Place the samples (including positive and negative controls) in the PCR machine run according to the parameters outlined in section 2b.

4. Run out the PCR products on an agarose gel with a standard to check size of the amplification product, photograph or note intensity of band.

d) Cleaning PCR products using QIAquick PCR purification kit (also used for cleaning tDNA)

1. Add 5 volumes of buffer PB to 1 volume of PCR reaction and mix, for 100 µl PCR reaction add 500 µl buffer PB and for 150 µl tDNA add 750 µl buffer PB. It is not necessary to remove mineral oil.

2. Place a QIAquick spin column in a 2 ml collection tube.

3. Apply the sample to the column and centrifuge for 30 to 60 seconds to bind the DNA in the column.

4. Discard flow through. Place column back in the same tube.

5. Add 0.75 ml of buffer PE to wash and centrifuge for 30 to 60 seconds.

6. Discard flow-through and place column back in the same tube. Centrifuge for 60 seconds at maximum speed.

7. Place QIAquick column in a clean 1.5 ml microfuge tube.

8. To elute DNA add 50 µl 10 mM Tris-HCl, pH 8.5 or water to the centre of the column and centrifuge for one minute. For increased concentration add 30 µl elution buffer to the centre of the column, stand for one minute and then centrifuge. Elution efficiency is dependent on pH and maximum efficiency is acheived between pH 7.0 and 8.5, if using water make sure the pH value is within this range.

9. Run ca. 3 µl of product on gel to check for intensity of band as a rough estimate of the DNA concentration.

e) Cycle sequencing

Separate reactions are required for each primer, one reation for ITS 4 and one for ITS 5, so double the number of cycle sequencing samples for each PCR product.

1. Make a master mix in the same way as for PCR reactions using the following ingredients per sample:

Terminator Ready Reaction mix (contains dNTPs and Taq)	2µl
Template PCR product	x
Primer diluted to 20 ng/µl	0.5
Sterile double distilled water	2.5 - x
Total reaction mix per sample	**5 µl**

The template volume (x) depends on concentration of PCR product and can be varied from 1 to 2.5 µl. If 2.5µl of template are added no water is required.

2. Add 5-x µl of master mix to tubes and add x µl of template.

3. Place tubes in cycle sequencing machine set to run 26 cycles of 10 seconds at 96°C to denature, 50°C for five seconds to anneal and 60°C for four minutes for sequence extension. Temporarily store products at 4°C.

f) Cleaning sequencing products

1. For each sample prepare a 1.5 ml eppendorf tube containg 2 µl 3M sodium acetate (pH 4.6 or 5.2) and 50 µl absolute ethanol.
2. 5µl reactions can be difficult to remove from the cycle sequencing reaction tube so their volume is made up to 20 µl with sterile double distilled water and the entire volume of the sequencing reaction plus water is transferred into the 1.5 ml tube containing the sodium acetate and alcohol.
3. Leave at room temperature for 5 minutes then vortex and place on ice for 10 minutes.
4. Spin at 13 000 r.p.m. for 25 minutes.
5. Drain off the solution
6. Wash pellet with 300 µl 70% ethanol and spin at 13 000 r.p.m. for 15 minutes.
7. Drain off the solution, add a further 300 µl 70 % ethanol and repeat spin for 15 minutes.
8. Drain of the solution and dry in oven for ca. 30 minutes.
9. Store the dry tubes at -20°C.

The following steps are performed by the technician operating the cycle sequencing machine immediately prior to loading the samples:

10. Add EDTA / formamide mix and vortex to mix, spin briefly.
11. Heat for 5 minutes at 90°C and place on ice immediately prior to loading on the gel.

g) Recipes

2x CTAB buffer: 100mM Tris HCl, pH 8.0
1.4M NaCl
20mM EDTA
2 % CTAB (hexadecyltrimethylammonium bromide)

CsCl/EtBr (CsCl density = 1.55 g / 1.0 litre solution)

CsCl	753.0g (heat on low)
Tris base 50mM	50.0ml of 1.0M Tris base
EDTA 10mM	40.0ml of 0.25M EDTA
EtBr 100mg/l	10.0ml of 10.0 mg/ml solution
double distilled water	900 ml

Appendix V: ITS data matrix of all species of Hyptidinae sequenced

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Hyp.aln.combine.nexus                                          Page 1
Tuesday, June 23, 1998  3:08 pm

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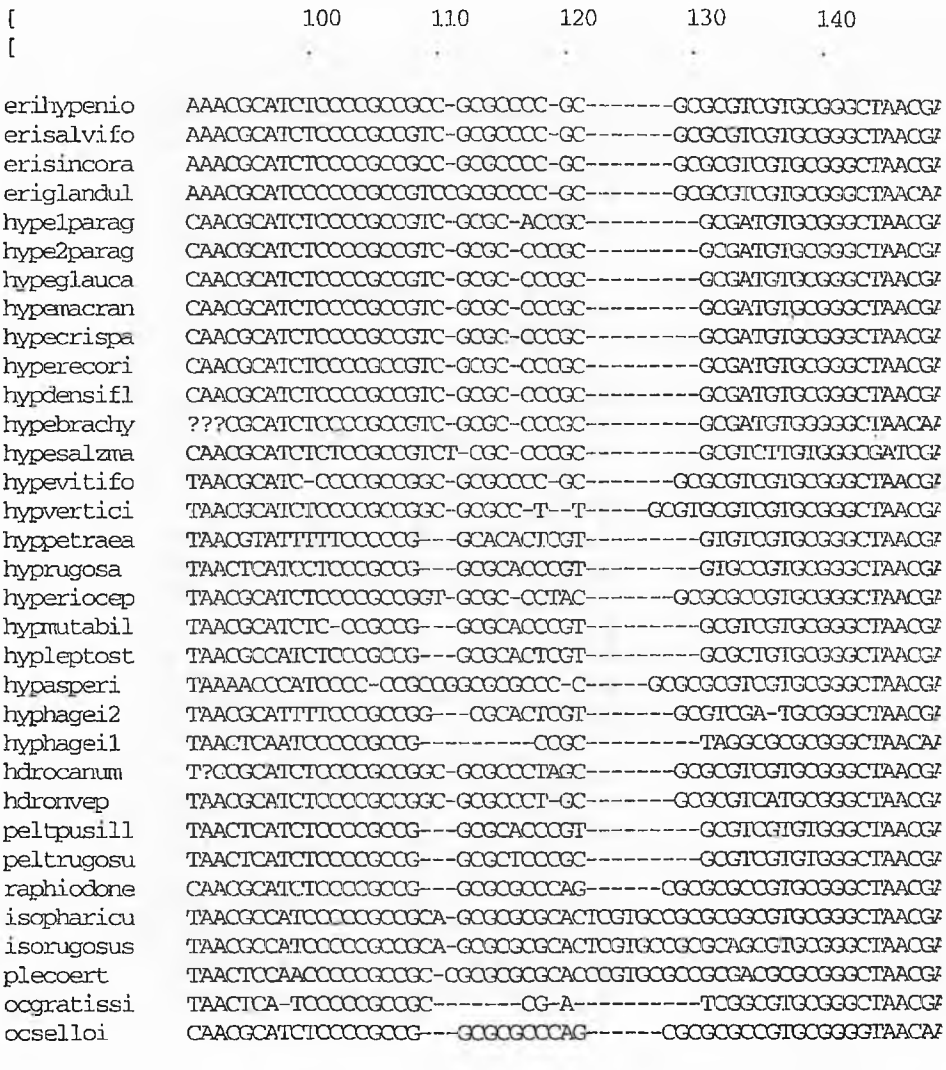
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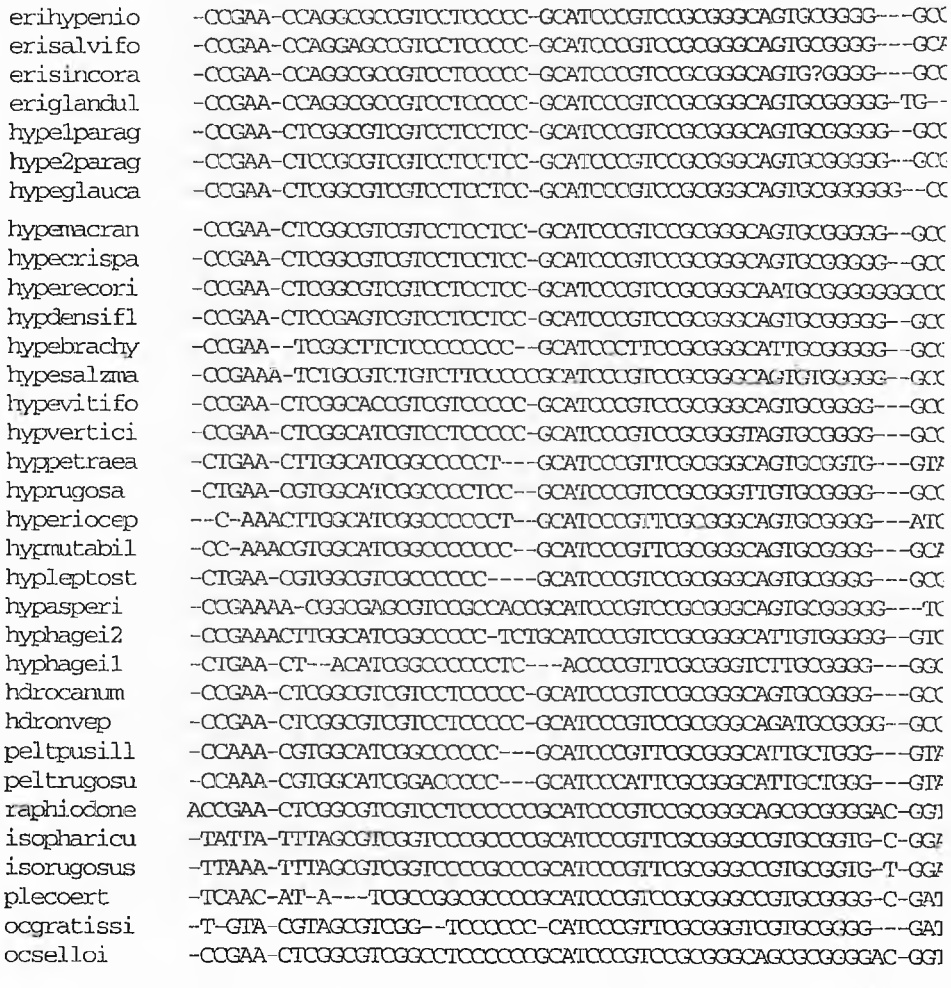
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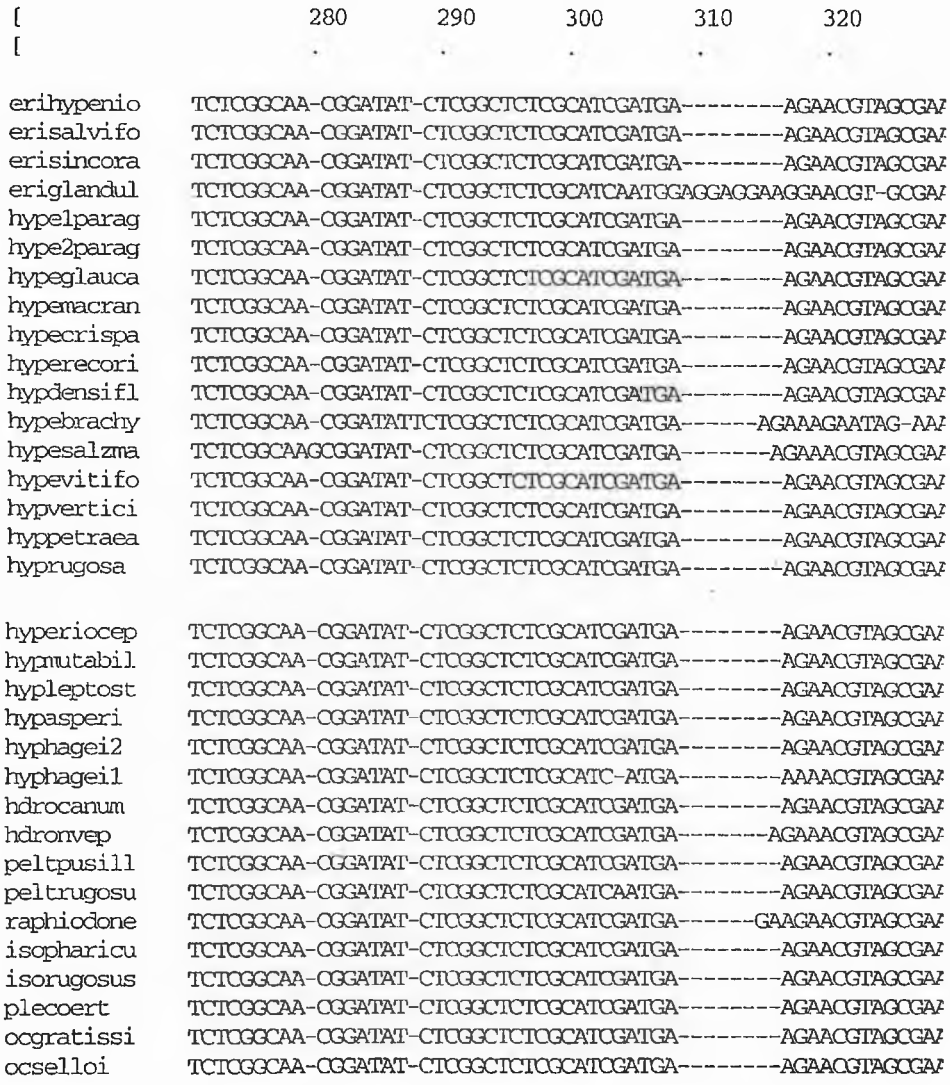
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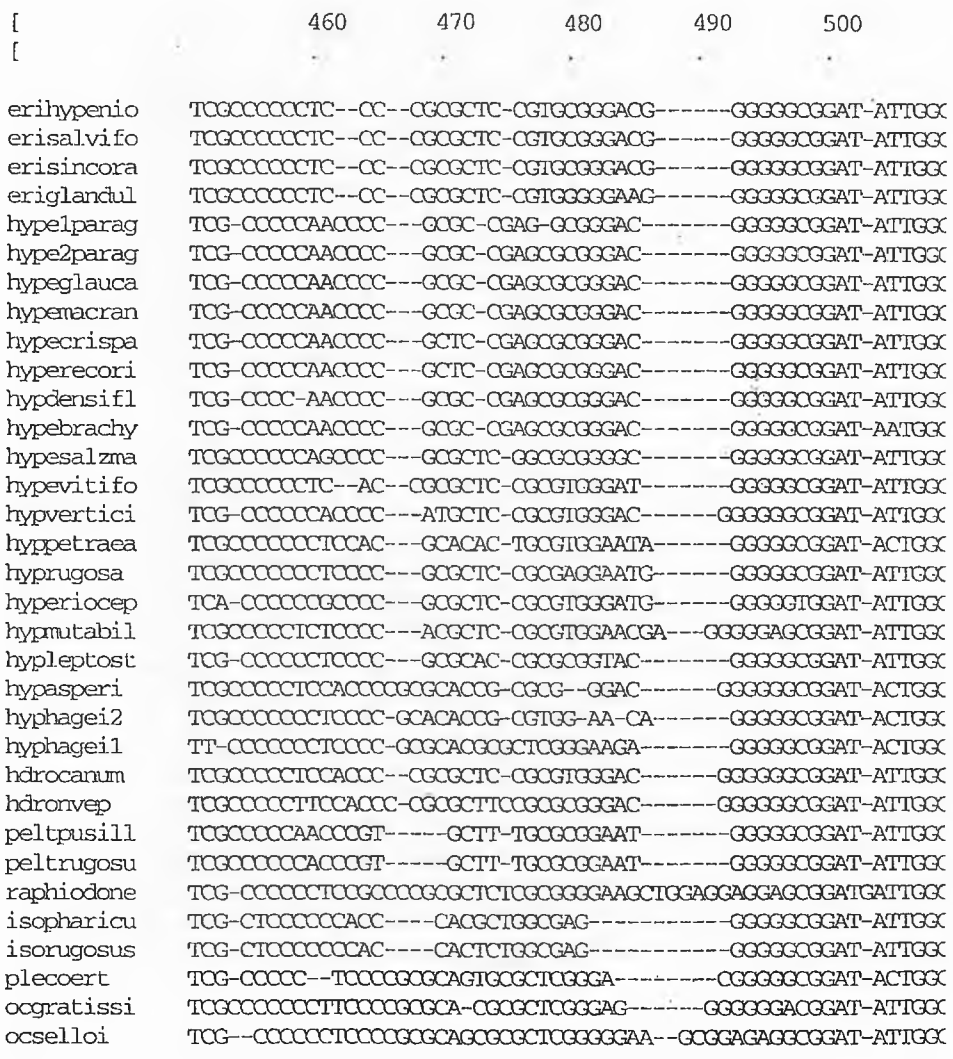
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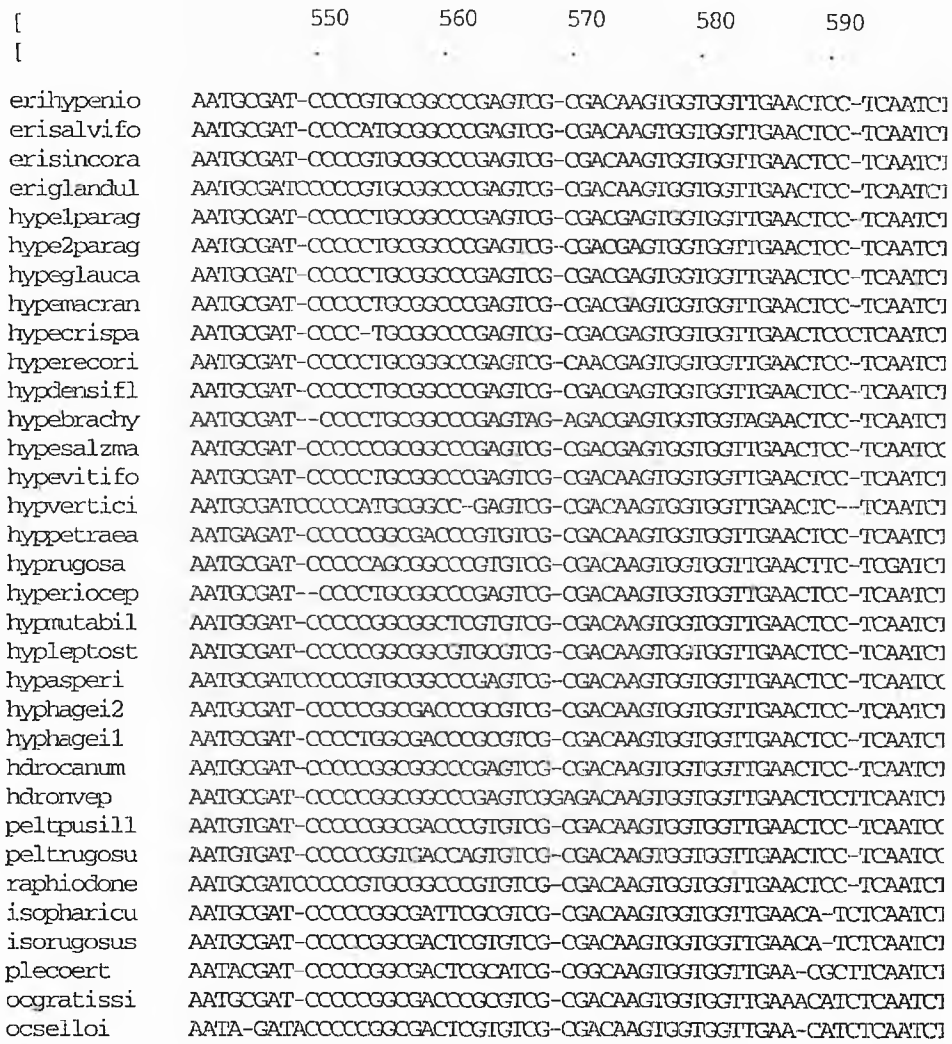
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END;
;

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