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ROYAL BOTANIC GARDENS KEW

EDITORIAL

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A belated welcome, we are afraid, to this the 6th issue of the Lamiales Newsletter. Apart from perennial excuses due to pressure of work, one of us, who was taking a year in Brazil, until mid-1997, is still trying to catch up, and is currently very busy bringing together and editing contributions for a Lamiales part for Kubitzki's Families and Genera of Vascular Plants. This is proving challenging, but he is very grateful for all the support he has received. In spite of all this, we still hope to be on time for the next issue, and have already received contributions! Please continue to send your valuable contributions to **y.harvey@rbgkew. org.uk.**

The Newsletter is not intended for papers which could be more appropriately published in reviewed scientific journals but is rather intended for preliminary research reports especially on new or interesting topics, reports on the activities of research groups, requests for information, news and comments. We try to vary the content, so that there will be something for everyone, so please do not be down-hearted if your paper does not appear. The present issue starts with a topic that could affect us all: phylogenetic nomenclature, using Labiatae as a case study, by Phil Cantino. This is a highly contentious issue, so we hope you will send us your views. Dick Olmstead points out that in line with molecular evidence, the Lamiales includes not only the Verbenaceae and Labiatae, but also other families such as Scrophulariaceae, Acanthaceae, Bignoniaceae, Myoporaceae etc. Should the Newsletter be more inclusive or should we restrict it to just the Labiatae? Not wishing to either exclude anyone already in our group, nor wishing to take on a lot more work, the editors have taken a unilateral decision to keep things as they are for the present, but your views would be welcome.

Unfortunately, the hoped-for Lamiales meeting in Mexico has not materialized, due to unforeseen difficulties, so we have suggested postponing Mexico for a future meeting. As a lot has happened since our last meeting, we look forward to suggestions for a venue and local support for a meeting around the year 2001. Our directory of research in the Labiatae and Verbenaceae is now showing its age, and needs updating. Please can we ask you to complete the accompanying form?

So far we have been fortunate to have the newsletter production funded by R.B.G. Kew. Due to current financial restraints and to ensure continuity, we would like to ask for a voluntary contribution of £3 Sterling (cash or bank draft) or $5 US (cash only), per issue. This will cover postage and part of production costs and you may find it worthwhile to pay for a few years in advance. ❑

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OXERA, FARADAYA AND HOSEA, SYSTEMATICS AND CONSERVATION

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The genera *Oxera* Labill., *Faradaya* F. Muell. and *Hosea* Ridl. were revised as part of a D.Phil thesis at the Department of Plant Sciences, Oxford (de Kok 1997). A cladistic analysis of the whole group, based on morphological, flavonoid and

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Table 1. Generic differences between *Oxera*, *Faradaya* and *Hosea* and some close relatives

	Number of fully matured stamens	Mature fruit elongated	Nr. of corolla lobes	Flower resupinate	Corolla wall fleshy	Orientation of the vascular bundles in the petiole	Ovule position
Oxera	**2(-4)**	**Yes**	**4**	**No**	**Some**	**a**	**p**
Faradaya	**4**	**Yes**	**4**	**No**	**Yes**	**a**	**p**
Hosea	**4**	**Yes**	**4**	**No**	**No**	**b**	**p**
Clerodendrum s.s.	**4**	**No**	**5**	**Yes**	**No**	**b**	**c+m**
Rotheca	**4**	**No**	**5**	**No**	**No**	**b**	**c+m**
Huxleya	**4**	**?**	**5**	**?**	**No**	**?**	**c+m**
Kalaharia	**4**	**No**	**5**	**Yes**	**No**	**b**	**c**

Key to symbols: **a**. interrupted full circle, **b**. half circle, **c**. basal, **m**. middle position & **p**. pendulous

nrDNA characters, was also part of the project. The revisions, their flavonoid contents (Grayer & de Kok, in press) and the cladistic analyses will be published elsewhere. In this article a brief overview of the genera studied is given, and the conservation status of two rare *Oxera* taxa are discussed.

In the genus *Oxera*, 21 species and six subspecies are recognised. The genus is mainly restricted to New Caledonia, but a new species which is only known from cultivation is recognised from Vanuatu. The genus is subdivided into five informal groups, based on a number of floral and fruit characters. In the genus *Faradaya* only three species are recognised: *Faradaya amicorum* (Seem.) Seem., *F. lehuntei* (Horne ex Baker) A.C. Smith and *F. splendida* F. Muell. The genus is found from Samoa and Tonga to New Guinea, North Queensland and Sabah. The monotypic genus *Hosea* is recognised as distinct with its one species, *Hosea lobbii* (C. B. Clarke) Ridl., confined to Sarawak and Brunei.

Oxera, *Faradaya* and *Hosea* differ from the larger genus *Clerodendrum s.s.* and *Rotheca* and the monotypic genera *Huxleya* and *Kalaharia* by having a tetramerous corolla and an elongated mature fruit (Table 1). This morphological character is supported for *Clerodendrum s. s.* and *Rotheca* by a cladistic analysis of cp- and nrDNA characters (Steane 1995).

Like *Clerodendrum*, *Rotheca* and *Huxleya*, *Hosea* has two pairs of equal stamens and a fragile corolla. It also shares with *Clerodendrum* and *Rotheca* the character of having its vascular bundles in the cross-section of the petiole in a half circle, rather than in an interrupted full circle as in *Oxera* and *Faradaya*. The general shape of the corolla reminds one strongly of *Faradaya*, but it differs in the texture of the wall. *Faradaya* corollas are thick and waxy compared with *Hosea* flowers. The sharply pointed shape of the fruit is one of the main differences between *Hosea* and its close relatives.

Faradaya has two pairs of equal stamens and a large fleshy corolla. It shares with *Oxera* the character of the vascular bundles in the cross-section of the petiole in an interrupted full circle. *Oxera* usually has one pair of stamens, but sometimes may have two pairs. The flowers of *Faradaya* are tetramerous and actinomorphic, unlike those of *Clerodendrum* which have five corolla lobes, and those of *Oxera* and *Hosea*, which although having four corolla lobes, are zygomorphic, as one of the lobes is a well developed lip.

Conservation

Two taxa of *Oxera* can be considered endangered according to the IUCN status categories (Lucas & Synge 1978: 25); both plants are restricted to the sclerophyll forest of New Caledonia which is the vegetation type most at risk on this Island (Bouchet et al. 1995: 418-420).

Oxera rugosa is known from only a few localities in the south of the island. There is one population (which I have not seen) which grows in the protected area of the Parc Territorial de la Thy. The other localities are along the southern slopes of the Mourange mountains. These areas are unprotected and are in constant danger of being burned (Bouchet *et al.* 1995). In 1995, one of the best known populations was accidentally set on fire by the French army during an exercise with live ammunition on a nearby shooting range.

Oxera pulchella subsp. *grandiflora* is restricted to the Nouméa peninsula, and is threatened by the destruction of its habitat for further development of Nouméa City. The last remaining undamaged forest (Point Lascalle) on the peninsula is earmarked for development as a housing estate (Gramon 1995). A road was pushed through the forest in 1995, and has already severely damaged the main population of the plant in this forest. The taxon is also found in two smaller protected areas on the peninsula: Parc Forestier M. Corbasson (35 ha) and Parc Provincial du Ouen-Toro (44 ha). Both parks are city parks rather than biodiversity reserves (Bouchet *et al.* 1995), and no special measures are taken to safeguard the plants.

Specimens of both *O. rugosa* and *O. pulchella* subsp. *grandiflora* should be taken into cultivation to safeguard the survival of the taxa. Plants of *O. pulchella* which are already in cultivation should be properly identified as to which subspecies they belong and maintained in case reintroduction of *O. pulchella* subsp. *grandiflora* into New Caledonia becomes necessary. One way to ensure the survival of *O. pulchella* subsp. *grandiflora* is to promote its use as a garden plant in New Caledonia itself (Gramon 1995), but efforts will have to be made to prevent hybridisation with the northern subspecies.

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LABIATAE AS A CASE STUDY IN PHYLOGENETIC NOMENCLATURE

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Because the journal *Systematic Biology* (formerly *Systematic Zoology*) is not widely read in botanical circles, many readers of this newsletter may not be aware of a recent paper (Cantino *et al*., 1997) in which the classification of Labiatae is used to compare the current system of nomenclature (as represented in the ICBN) to an alternative phylogenetic system proposed by de Queiroz and Gauthier (1992, 1994, 1996). This case study suggests that phylogenetic nomenclature functions better than the current system when one's objective is to provide unambiguous and stable names for clades.

For example, Teucrioideae (Cantino *et al*., 1992) is a novel and well supported clade of 23 genera that transcends the traditional boundary between Labiatae and Verbenaceae. One would like a newly discovered clade to have a name that is both unique (not previously applied to another group) and stable. Teucrioideae, the name that had priority under the ICBN when the clade was discovered, is not unique but at least had not been used in recent times and therefore was unlikely to be associated with another grouping in anyone's mind. However, the subsequent finding that *Ajuga* is a member of this clade (Wagstaff and Olmstead, 1997; Wagstaff *et al*., 1998) requires that the taxon be renamed Ajugoideae, which has priority under the ICBN, an undesirable change because this name has been applied already to at least three other groupings in well known or recent classifications. Under the phylogenetic system of nomenclature, this would not have occurred; once a clade is named, it retains that name permanently. The greatest difficulties with the current system were encountered with infrafamilial taxon names based on the type of the family. For example, the ICBN has required me to apply the name Lamioideae to three different groups in 11 years (Cantino and Sanders, 1986; Cantino *et al*., 1992; Cantino *et al*., 1997), since any subfamily-level taxon that includes *Lamium* must bear this name.

The Phylogenetic System of Nomenclature

Phylogenetic nomenclature differs fundamentally from the current system in lacking mandatory ranks above the species level and in the way that taxon names are defined. In the current system, plant names

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at or below the family level are operationally defined as the taxon assigned to rank X that contains type Y (de Queiroz and Gauthier, 1994). For example, Verbenaceae is the taxon of family rank that contains the type of the genus *Verbena*. This kind of definition is also frequently used above the family level, but this is not required by the ICBN. Phylogenetic nomenclature is entirely different in that names are defined in terms of phylogenetic relationships (de Queiroz and Gauthier, 1990; Schander and Thollesson, 1995). For example, Lamiaceae might be defined as the least inclusive clade that contains *Lamium purpureum*, *Glechoma hederacea*, and *Vitex agnus-castus* (although many more species were cited in my published definition [Cantino *et al*., 1997] to reduce the likelihood of subsequent changes in taxon membership due to future phylogenetic findings).

Phylogenetic nomenclature is the logical culmination of a revolution that began with Darwin and was advanced by Hennig. De Queiroz (1997) dubbed this the "evolutionization of taxonomy." As the principle of common descent has assumed a progressively more prominent position in the philosophical framework of systematics, taxon names have come to have implicit phylogenetic meanings. The final step in this process is replacement of our current system of nomenclature, which is based on preDarwinian principles, with one that defines taxon names in explicit phylogenetic terms.

A particularly desirable feature of phylogenetic nomenclature is that it makes it possible to name newly discovered clades without changing the names of other taxa. A disadvantage of the current system is that the ranks of taxa, and hence their names, are dependent on their position relative to other taxa. Thus naming a newly discovered clade can cause a cascade of name changes elsewhere in the hierarchy as taxa shift in rank (Kron, 1997; Hibbett and Donoghue, 1998). By analogy, if chemists were operating under a system that required the names of some elements to change when a new one is discovered, surely the scientific community would rebel! This serious drawback of the current system discourages systematists from naming clades as they are discovered (Hibbett and Donoghue, 1998). As a result, our classifications are falling farther and farther behind our knowledge of relationships.

Although phylogenetic nomenclature is "rankless" in that it lacks mandatory ranks, there is no prohibition against using ranks if one wishes to (de Queiroz, 1997). The important thing is that the taxon name is not dependent on rank and thus does not change if its position in the hierarchy changes. This is one way in which the phylogenetic system promotes nomenclatural stability.

Species and Clades

In the absence of mandatory ranks, the principal kinds of taxa in the phylogenetic system are clades and species. The acceptance of the species category in a rankless system may seem like a logical contradiction, but it is not if one views "species" not as a rank but as a fundamentally different kind of entity than a clade. If systematics is a science (and I hope this is no longer at issue), named taxa should correspond to objective natural entities, i.e., products of evolution that exist outside the human mind and whose existence is hypothesized based on evidence. Such entities are discovered rather than created. Species and clades qualify as objective entities for different reasons. Clades, being monophyletic, are complete systems of common descent. Species are not necessarily monophyletic (Rieseberg and Brouillet, 1994; Olmstead, 1995), and it has been argued that the concepts of monophyly and paraphyly don't even apply to species because the relationships among the individuals composing a species are tokogenetic (reticulating) rather than phylogenetic (Nixon and Wheeler, 1990). However, species are still objective entities if they are conceived of as segments of population level evolutionary lineages (de Queiroz, in press) whose cohesiveness is due to natural processes such as interbreeding (de Queiroz and Donoghue, 1988). In contrast, a paraphyletic supraspecific taxon is subjective in that it has no existence outside the mind of the taxonomist. The claim that species are sometimes paraphyletic is not a valid argument for formal recognition of paraphyletic taxa above the species level. Species need not be monophyletic to be objective natural entities, but monophyly is necessary above the species level unless one can demonstrate another natural process that conveys objective existence to supraspecific entities.

The Binominal System

Because phylogenetic taxonomy lacks formal ranks, there is no such thing as a genus, thus binominal nomenclature is incompatible with phylogenetic taxonomy. Although people may object to abandoning a convention that has been in use for over 200 years, a critical examination of binominal nomenclature reveals serious drawbacks (Cantino, 1998). One problem is that the binominal system cannot accommodate uncertainty about generic relationships. This can lead to a taxonomic dilemma when a genus ("X") is found to be para- or polyphyletic as currently delimited, but phylogenetic resolution is too poor to refer all of its species to smaller monophyletic genera. The most satisfactory approach would be to name the well supported clades within former genus X as genera but designate the species that lie outside these clades as "*incertae sedis*" with regard to genus. However, this cannot be done within the binominal system. A second problem with the binominal system is that it is a major cause of nomenclatural instability at the species level, because changes in generic circumscription necessitate species-level name changes. This is not true at any other rank (e.g., splitting a family into two families does not require that genus names change).

A viable alternative to binominal nomenclature is a system of hyphenated uninomials (Michener, 1964). Thus, for example, *Lamium purpureum* would become known as *Lamium-purpureum* and would retain this name even if subsequent research demonstrated that it is not a member of a clade called *Lamium*. The transition to such a system would be relatively painless if the hyphenated uninomials were based on currently accepted binomials and retained the same type. As Michener (1964) noted, decoupling species names from genus names would have two advantages: It would greatly increase nomenclatural stability at the species level, where stability is most critical, by preventing changes in species names due to generic recircumscription. Secondly, it would facilitate reclassification above the species level in response to new research, as systematists would no longer have to weigh the implications for species-level nomenclature when deciding whether to translate their findings into classification.

The Future of Phylogenetic Nomenclature

Any major change in the naming of organisms is bound to involve a difficult transitional period, thus it should only be undertaken if the end product will be a substantial improvement. For this reason, I question the BioCode concept (Greuter *et al*., 1996) which I view as having a poor cost/benefit ratio. Why impose a new system of rules on the systematic community when it suffers the same fundamental drawbacks as the current system? On the other hand, the phylogenetic system has significant advantages. It would facilitate the naming of newly discovered clades without forcing name changes elsewhere in the classification, and it would substantially improve nomenclatural stability - at least at the species level and perhaps elsewhere. The jury is still out on the latter point. My case study using the Labiatae suggests that nomenclatural stability will improve above the species level as well, but other taxa should be examined.

What is needed now is a draft code of phylogenetic nomenclature, which could be applied experimentally to a wide variety of organisms with different levels of phylogenetic resolution. There is no reason why the two nomenclatural systems cannot coexist, so long as names bear some kind of designation to show which system they pertain to. For example, if names in the phylogenetic system were to end with a standard symbol such as the Greek letter ϕ (phi, for phylogenetic), they would be immediately recognizable. Users would know that a taxon called Scutellarioideaeϕ represented a hypothesized clade, whereas no such assumption could be made about a taxon called Scutellarioideae. Alternatively, a uniform ending might be used for all names in the phylogenetic system (e.g., -*ina*; Kron, 1997), but this would require the abandonment of many unambiguous names such as Asteraceae and Poaceae that already apply (although not explicitly) to clades.

The incompatibility of the traditional "Linnaean" system of classification (and nomenclature) with the goals of modern phylogenetic systematics has been noted by both opponents (e.g., Brummitt, 1996, 1997; Sosef, 1997) and proponents of the view that all taxa above the species level should be monophyletic. Even if one accepts paraphyletic supraspecific taxa, phylogenetic nomenclature appears to improve nomenclatural stability and facilitate the naming of clades. If these advantages are corroborated by additional case studies, the systematic community should give serious consideration to adopting phylogenetic nomenclature. Classification is the principal way in which knowledge of relationships is imparted to other biologists and the general public. Tradition alone is not sufficient reason to retain a set of conventions that impedes the communication of scientific progress.

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STUDIES ON COLOMBIAN LABIATAE LECTINS

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During the last decade some genera from Labiatae in Colombia, have been reviewed by Wood (1988), Wood and Harley (1989) and Fernández-Alonso (1990, 1995). In the course of this work many endemic species have been described.

Recently the Laboratory of Biochemistry at the Chemistry Department and the Instituto de Ciencias Naturales, both at the National University, Bogotá, Colombia, have joined forces to work on lectins from Labiatae. Due to their many interesting properties, two of them being the specific recognition of carbohydrates and the agglutination of cells, the lectins have attracted much attention. Leguminosae and some Gramineae are the commonest source of lectins; in Labiatae only the *Salvia* species from temperate zones have been investigated in this respect (Bird & Wingham, 1974, 1976, 1977, 1982) and in *Salvia sclarea* L. a lectin with very interesting properties and potential medical applications, has been characterised (Piller *et al*. 1986).

Our first purpose is to screen the numerous (62 taxa, including subspecies) neotropical Colombian *Salvias* as well as some *Lepechinia*, *Hyptis* and *Minthostachys* species for seed lectins. Our preliminary results show that a lectin is present in *Salvia bogotensis* Benth., *S. palifolia* Kunth, *S. pauciserrata* Benth., *S. rubescens* Kunth, *S. amethystina* J. E. Smith, and *Lepechinia conferta* (Benth.) Epling. In *Salvia scutellarioides* Kunth, *S. sordida* Benth., *S. uribei* Wood & Harley, *S. gachantivana* Fernández-Alonso and *Lepechinia salvifolia* (Kunth) Epling, no lectin has been detected so far. Some species in which lectins have been detected, such as *Salvia bogotensis* (section *Angulatae*), *S. pauciserrata* (section *Flexuosae*) and *S. rubescens* (section *Rubescentes*), are very diverse in Colombia with several subspecies.

Our second goal is to purify and characterise the lectin from *S. bogotensis* subsp. *bogotensis*, a taxon which is quite abundant near Bogotá. We have devised an extraction scheme and are currently working on the isolation and characterisation of the lectin. We are interested in establishing contacts with groups working on Labiatae lectins/proteins so we can discuss results and eventually collaborate.

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CONFIRMATION OF A MONOPHYLETIC CHLOANTHOIDEAE (LAMIACEAE) COMPRISING TRIBES CHLOANTHEAE AND PROSTANTHEREAE

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As part of our ongoing molecular systematic research into the phylogeny of the Lamiales, we have conducted an investigation of the hypothesis put forward by Cantino (1992) that the Labiate tribe Prostanthereae and the Verbenaceous tribe Chloantheae (*sensu* Bentham 1876; Labiatae subfam. Prostantheroideae and Verbenaceae subfam. Chloanthoideae *sensu* Briquet 1895) together form a monophyletic group, which also included *Tectona* (Viticoideae *sensu* Briquet). The evidence for this postulated relationship comes from a cladistic analysis of morphological and anatomical characters, in which members of these two groups come out adjacent to each other in an unrooted tree (Cantino 1992). However, alternative prospective rootings of the tree differed in whether the groups formed a clade, or a paraphyletic group with the root attaching among them. Prior to Cantino's (1992) suggestion, these groups generally had been assigned to separate families. However, Junell (1934) included Chloantheae and much of the rest of the Verbenaceae, excluding subfamily Verbenoideae, in the Lamiaceae. In contrast, Hutchinson (1959) elevated the Chloantheae to Chloanthaceae, a move accepted in a series of papers on the group by Munir (Munir 1976, 1977, 1978a, 1978b, 1978c, 1979). Based on the study cited above, Cantino *et al*. (1992) suggested a provisional classification for the Labiatae that included Prostanthereae and *Tectona* in the subfamily Chloanthoideae, but excluded *Spartothamnella* from the subfamily and excluded *Acharitea* and *Nesogenes* from the Labiatae. The latter three genera were included in Chloanthoideae by Briquet (1895) and *Nesogenes* and *Spartothamnella* were included in Chloantheae by Bentham (1876). This study builds upon previous studies using chloroplast DNA (cpDNA) sequences of the gene *ndh*F for phylogenetic inference in the Lamiales (Olmstead & Reeves 1995; Scotland *et al*. 1995; Steane *et al*. 1997; Wagstaff *et al*. 1998).

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Materials and Methods

DNA of most ingroup species was obtained from either silica-gel-dried material (collected by BJL) or from herbarium specimens. Sequences of *ndh*F were determined by PCR-amplification of the region encompassing the first 2135 nucleotides (nt) of tobacco *ndh*F (2086 nt excluding PCR primers at each end), followed by either manual or automated dideoxy sequencing (Olmstead & Reeves 1995). A total of 45 sequences were analysed, including 23 previously unpublished sequences mostly representing Chloantheae and Prostanthereae and 22 previously published sequences representing species from throughout the Lamiaceae and related families. Sequences were aligned by eye with gaps inserted to accommodate variation in length and to optimize positional homology among species throughout the sequence. The phylogeny was inferred by parsimony with all changes equally weighted using the program PAUP* (test version 4.0d61 kindly provided by D. Swofford). Analyses were run with 100 replicate random-order entries of the taxa using a heuristic search strategy (TBR swapping; MULPARS). Bootstrap analysis was done with 500 replicates, each with a single random-order entry of the taxa and a heuristic search with TBR swapping, but with MULPARS off.

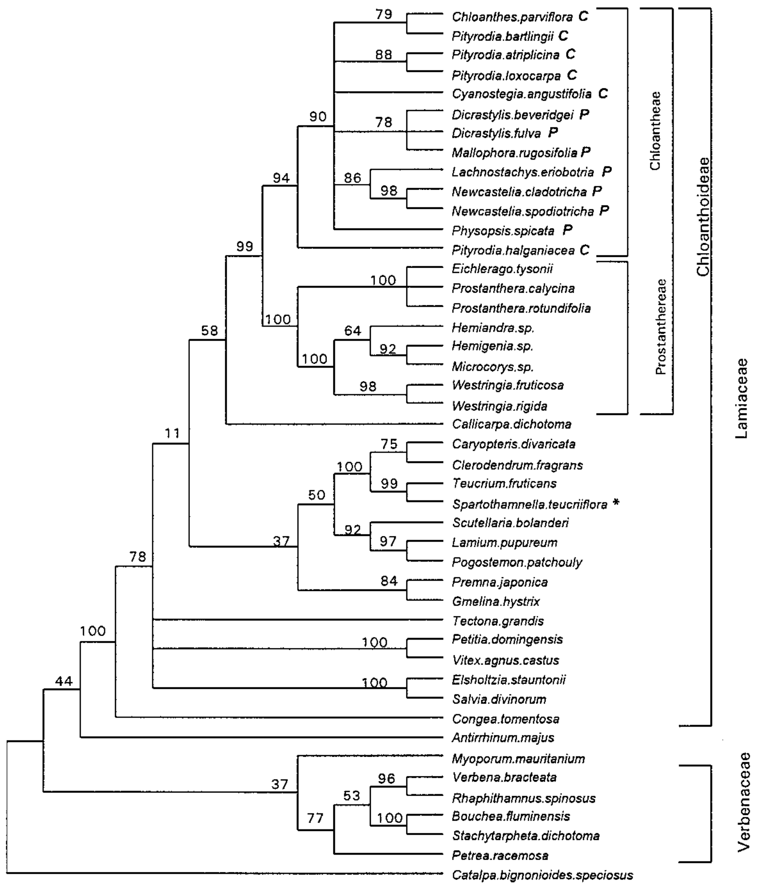

Fig. 1: Strict consensus tree of 72 equally parsimonious trees. The classification of the Chloanthoideae into tribes Chloantheae and Prostanthereae is indicated by brackets. "C" and "P" indicate Junell's (1934) tribes Chloantheae and Physopsideae. An asterisk highlights the placement of *Spartothamnella*, assigned to Chloanthoideae by Bentham (1876), Briquet (1895) and Munir (1979).

Results and Discussion

The sequences ranged in length from 2062 nt in *Pityrodia atriplicina* to 2113 nt in *Stachytarpheta dichotoma*, with a typical length of 2101 nt. Several of the sequences that were derived from herbarium specimens needed to be amplified in smaller fragments than typically used and in a few cases some regions as long as ca. 500 nt were not able to be sequenced. More commonly this only resulted in scoring the PCR primer sites as missing (typically ca. 25 nt each for 2-3 primer sites). The insertion of 24 gaps were required for the alignment. Most gaps were unique to a single taxon and were not used for phylogenetic inference.

The phylogenetic analysis yielded 72 equally parsimonious trees (length = 2258; Consistency Index = 0.56; Retention Index = 0.60). The strict consensus of these trees is shown in Fig. 1 along with bootstrap values for clades and a suggested classification based on our results.

The results concur with previous studies that indicate that the Verbenaceae *s.s.* do not form a clade with the Lamiaceae *s.l.* (Olmstead *et al.* 1993; Wagstaff & Olmstead 1997) and in finding a monophyletic Lamiaceae *sensu* Cantino *et al.* (1992) with the inclusion of *Congea* (Symphorematoideae) (Wagstaff *et al.* 1998). In this study five species of Verbenaceae *s.s.*, representing the four remaining tribes, are included and together form a clade. This result contrasts with the results from a previous *rbc*L-based study (Wagstaff & Olmstead 1997), in which *Petrea* did not form a clade with the other four genera. These results provide strong evidence that tribe Chloantheae does not belong with the Verbenaceae.

Sampling here includes fewer representatives of each of the other subfamilies of Lamiaceae than in a previous study of the family (Wagstaff *et al.* 1998), but includes all genera of Chloantheae, except the monotypic *Hemiphora*, and all genera of Prostanthereae, except the monotypic *Wrixonia*. In a previous study, combined analysis of *rbc*L and *ndh*F sequences provided weak indication that *Tectona* did not belong with the Australian Prostanthereae (represented by one species of *Prostanthera* only; tribe Chloantheae was not represented), but *ndh*F sequences were unable, by themselves, to resolve the question of whether *Tectona* belonged with *Prostanthera* (Wagstaff *et al.* 1998). Sampling was insufficient to address the greater question of chloanthoid monophyly. The results presented here provide strong evidence (99% bootstrap support) of a monophyletic Chloanthoideae, excluding *Tectona*. In addition, these results find strong support for monophyletic tribes Chloantheae (94%) and Prostanthereae (100%). There is modest support (58%) for a relationship of this group with *Callicarpa* (subfamily Viticoideae). *Tectona* does not appear closely related to any other group and fits the pattern of belonging to a viticoid grade that is more or less basal to the other subfamilies of Lamiaceae (except Symphorematoideae, represented here by *Congea*, which may be basal to the rest of the family, Wagstaff *et al.* 1998).

Relationships within tribes Chloantheae and Prostanthereae are incompletely resolved, but some preliminary conclusions seem warranted. Briquet (1895) elevated Bentham's (1876) tribe Chloantheae to subfamily Chloanthoideae and split it into three tribes, Achariteae (*Acharitea*, *Nesogenes*, *Pityrodia* and *Spartothamnella*), Chloantheae (*Chloanthes*, *Denisonia*, *Cyanostegia* and *Hemiphora*) and Physopsideae (*Dicrastylis*, *Lachnostachys*, *Mallophora*, *Newcastelia* and *Physopsis*). Subsequently, on the basis of gynoecial morphology, Junell (1934) suggested the transfer Chloanthoideae to the Labiatae at the tribal level, with two subtribes, Chloanthinae and Physopsidinae, and dispersed the elements of tribe Achariteae to other groups in and out of the Labiatae. He transferred *Pityrodia* to Chloanthinae, *Spartothamnella* to tribe Viticeae (Labiatae), subtribe Ajuginae, near *Teucridium*, and *Acharitea* and *Nesogenes* to Stilbaceae (later segregated into Nesogenaceae by Marais, 1981). Munir (1978b, 1979) maintained two tribes within the Chloanthaceae, Chloantheae and Physopsideae, similar to Junell's (1934) circumscriptions, but maintained the Australian endemic *Spartothamnella* in tribe Chloantheae. Additional evidence for the relationship of *Spartothamnella* with the Ajugeae (Teucrioideae sensu Cantino *et al.* 1992), close to *Teucrium*, comes from pollen morphology (Abu-Asab and Cantino 1992; Cantino 1992) and is confirmed by our results.

Neither of the tribes Chloantheae and Physopsideae, as described by Briquet (1895) and modified by Junell (1934), is monophyletic in any of the most parsimonious trees. Subsequent searches in which these groups were constrained to monophyly individually and simultaneously resulted in trees four steps longer for a monophyletic Physopsideae, 11 steps longer for Chloantheae (*sensu* Junell 1934), and 13 steps longer for both to be monophyletic on the same tree. These results concur with Cantino (1992) in finding *Pityrodia* to be polyphyletic and having *P. halganiacea* as sister to the rest of the group. These results also concur with Cantino in finding a clade comprising *Dicrastylis* and *Mallophora* and in finding a close relationship between *Lachnostachys* and *Newcastelia* (a clade here, but unresolved in Cantino 1992).

Within tribe Prostanthereae, Cantino (1992) and Conn (1992a) concurred in recognizing two lineages, one comprising *Prostanthera*, *Eichlerago* and *Wrixonia*, and the other comprising *Hemiandra*, *Hemigenia*, *Microcorys* and *Westringia*. Our results are congruent with their findings. Cantino (1992) and Conn (1992a) also agreed that *Eichlerago* was derived from within *Prostanthera* and should belong to that genus (Conn 1992b), a conclusion that is consistent with our results. Both authors also concurred on the close relationship between *Microcorys* and *Westringia* based on the presence of a reduced number of fertile anthers. However, our results suggest a closer relationship between *Microcorys* and *Hemigenia*, suggesting an independent loss of one pair of fertile stamens in *Microcorys* and *Westringia*. A reduction in fertile stamen number also occurs in *Wrixonia*.

Before this project is completed, it is hoped that *Hemiphora* and *Wrixonia* can be added to the analysis to complete sampling of recognized genera in the subfamily. Some preliminary conclusions that can be drawn include 1) neither *Spartothamnella* nor *Tectona* belong in subfamily Chloanthoideae, 2) two tribes can be recognized within Chloanthoideae, Chloantheae and Prostanthereae (*sensu* Bentham 1876), and 3) Briquet's (1895) tribes Achariteae, Chloantheae, and Phypsopsideae, and Junell's (1934) tribes Chloantheae and Physopsideae should not be recognized.

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Note added in proof: *Hemiandra* sp. = *H. pungens*; *Microcorys* sp = *M. exserta*; *Hemigenia* sp. = *M. obovata*.

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ETHNOBOTANY AND ANTICONVULSANT PROPERTIES OF LAMIACEAE FROM RIO GRANDE DO SUL (BRAZIL).

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Introduction

The Lamiaceae is one of the most diverse and widespread plant families in terms of ethnomedicine. The medicinal value of Lamiaceae species is largely based on their volatile oils (Moerman, 1991). Regarding its effects on the central nervous system (SNC), studies proved anticonvulsant activities in at least four genera, among which *Aeollanthus* (Elisabetsky *et al.*, 1995a), *Leonurus* (Chauhan, 1988), and *Salvia* (González, 1990) are represented in the state of Rio Grande do Sul (RS), Brazil (Fig. 1). According to Ab'Sáber (1971) there are three vegetation domains in this region, Undulating grasslands, Atlantic Forest and *Araucaria* Mixed Forest. In RS the genera *Ocimum*, *Hyptis*, *Cunila* and *Salvia* are the best represented. The genus *Ocimum* with about 65 species, six endemic to South America, has its center of diversity in Tropical Africa (Pereira, 1979). *Hyptis* has some 350 neotropical species between the south of USA and Argentina, with 17 species in RS (Bordignon, 1990). *Cunila* has two centers of diversity, Mexico and Subtropical South America with 11 species in RS. These are often very aromatic and are frequently used in traditional medicine (Bordignon, 1997). *Salvia* is the best represented genus in RS (Epling & Toledo, 1943) with 23 species (Coelho de Souza, 1997).

Fig. 1 - Localization of Rio Grande do Sul (27°- 34° S latitude; 49°40′- 57° 30′ W longitude).

This present paper contributes to the ethnobotanical knowledge of Lamiaceae from RS. Following an ethnopharmacological study of *Aeollanthus suaveolens*, which is used by Amazonian "caboclos" against epilepsy, and which led to the identification of two volatile substances (linalool and g-decanolactone) active as anticonvulsants (Elisabetsky *et al.*, 1995a; Coelho de Souza *et al.*, 1997), the present study especially focused on species from the Subfamily Nepetoideae used as sedatives/anticonvulsants.

Methods

Ethnobotany: An inventory of Lamiaceae species in RS was made in the herbaria of Porto Alegre Colégio Anchieta - PACA, Instituto de Ciências Naturais - ICN of the Universidade Federal do Rio Grande do Sul and Universidade Federal de Pelotas - PEL. Ethnobotanical information, as well as chemical and pharmacological data available for the species listed in the inventory were collected from scientific and lay literature. The analysis of these data guided the selection of species for pharmacological tests. The criteria for selection were: 1) belonging to the Nepetoideae, 2) being easily available and showing broad distribution in RS, 3) being traditionally used as a sedative, 4) known to have classes of chemical compounds with recognized sedative properties. Samples were collected and voucher specimens were deposited at the ICN herbarium.

Phytochemistry: Methods for extraction of essential oils and pharmacological analysis have been detailed elsewhere (Elisabetsky *et al.*, 1995a, Coelho de Souza, 1997). In short, essential oils were extracted from 50-100g samples (leaves, inflorescence and terminal buds) by hydrodistillation (Clevenger apparatus). Essential oils were extracted from *Cunila galioides*, *Cunila menthoides*, *Cunila microcephala*, *Cunila spicata*, *Glechon thymoides*, *Hyptis mutabilis*, *Leonurus sibiricus* [Editorial note: This almost certainly refers to the species *Leonurus japonicus* Houtt.], *Nepeta cataria*, *Ocimum selloi*, *Rosmarinus officinalis*, *Salvia officinalis*. The pharmacological method was based on that of Gladding *et al.* (1985), focussing on pentylenetetrazol (PTZ, 90 mg/kg, sc.)-induced convulsions (in mice, thirty minutes after i.p. treatment, with doses ranging from 100 -700 mg/kg (i.p.) of essential oils). Diazepam (1.0 mg/kg i.p.) and phenobarbital (30 mg/kg i.p.) were used as reference drugs. Animals were observed for 60 min. for the presence of clonic convulsions lasting more than 3 seconds. Results were analysed by means of Fisher exact test.

Results

Through the herbarium survey, 125 species were listed, 49 being exotic to and 76 native in RS. Native species are distributed in 17 genera including *Aegiphila* (4 spp.), *Cunila* (10 spp.), *Glechon* (5 spp.), *Hedeoma* (3 spp.), *Hesperozygis* (2 spp.), *Hoehnea* (2 spp.), *Hyptis* (17 spp.), *Marsypianthes* (1 sp.), *Ocimum* (4 spp.), *Peltodon* (1 sp .), *Rhabdocaulon* (5 spp.), *Salvia* (14 spp.), *Satureja* (1 spp.), *Scutellaria* (2

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spp.), *Stachys* (2 spp.), *Teucrium* (2 spp.), and *Vitex* (1 spp.). The ethnobotanical survey shows that 53.4% of the species do not have recorded traditional uses, while 36.4% are considered of medicinal value. The medicinal uses are diverse, mainly focussing on digestive (14.2%), respiratory (11.9%), central nervous system ailments (16.2%) and topically applied for wound healing (11.5%). Lamiaceae species are also used as seasonings (15.3%), ornamentals (20.8%), species used by bees for honey production (13.9%) or for other industrial purposes (13.9%), mainly as cosmetics and personal care products. Among native species, 28% are utilized as medicines.

The exotic species are mostly cultivated and show a larger number of ethnobotanical uses, reflecting the influx of European immigrants and culture to the State. It is likely that some of the medicinal uses of native species are related to their morphological and/or aromatic resemblance to exotic species with medicinal uses diffused throughout the world. Of the exotic species, 86.7% (39) are cultivated while 13.3% (7) are adventives; among the cultivated species 84.6% (33) belong to the subfamily Nepetoideae.

Regarding phytochemical and pharmacological data, we found that 71.6% of species had not been the subject of previous studies. Phytochemical and/or pharmacological data are available for 20.7% of the exotic and only 7.8% of native species.

Cunila spicata essential oil was effective at 400 and 500mg/kg producing 66.7% of protection. *Glechon thymoides* essential oil was effective at 500mg/kg producing 60% of protection while the *Nepeta cataria* essential oil was effective at 700mg/kg producing 66.7% of protection. *Salvia officinalis* essential oil was found to be toxic at 500mg/kg, leading to convulsions and death. *Cunila galioides* essential oil induced a dose related protection, reaching a maximum at 300mg/kg. The ED50 (95% confidence limit) was estimated to be 217.7 (14.0-367.6) mg/kg and the LD50 (95%) was 398 (280.2-3057.2) mg/kg. Oils from *Cunila menthoides*, *Cunila microcephala*, *Hyptis mutabilis*, *Leonurus sibiricus* [Editorial note: This almost certainly refers to the species *Leonurus japonicus* Houtt.], *Ocimum selloi*, *Rosmarinus officinalis*, were ineffective up to 700mg/kg (data not shown).

Cunila galioides is called “poejinho” due to its likeness with *Mentha pulegium* and *C. microcephala* (known as “poejos”), being used traditionally for the same purposes (expectorant, tranquilizer and laxative) (Lopes *et al*., 1988). Few studies are available for *C. galioides*, which is restricted to southern Brazil. Preliminary phytochemical studies of its essential oil shows the presence of neral, geranial and linalool (Bordignon, 1997). Linalool proved to be active as an anticonvulsant in mice (Elisabetsky *et al*., 1995a) with relevant neurochemical properties (Elisabetsky *et al*., 1995b). Citral has sedative activity (Duke, 1992) while neral and geranial are citral isomers with no reported pharmacological studies. Therefore, the abundant presence of linalool, neral and geranial is likely to be related to *C. galioides* essential oil anticonvulsant activity.

Salvia officinalis is considered as a “cure-all” and, accordingly, indicated for a myriad of uses (Font Quer, 1992; De Feo & Senatore, 1993). When administered i.p. to mice *S. officinalis* essential oil induced lethal convulsions; this pattern of neurotoxicity is also found with opioids and local anesthetics. Although phytochemical and pharmacological studies of this species are abundant (Ribeiro *et al*., 1986, Masterova *et al*., 1989, Rutherford *et al*., 1992, Asanov *et al*., 1994, Tada *et al*., 1994), the toxic effect revealed by this study needs further investigation.

Conclusion

Lamiaceae species have been used all over the world for seasoning, perfumes and/or medicines (Tomas-Barberán, 1986; Heinrich, 1992; Lawrence, 1992). Recently, aromatic species extracts and distillates have become important raw material for the cosmetic, food and personal care industries (Lawrence, 1992). This study of Lamiaceae of RS showed the importance of the family for local communities and revealed the paucity of phytochemical and pharmacological studies of native species. Analysis of data gives a rational basis for some traditional uses of Lamiaceae species and suggests that this family is a source of yet unknown medicinal resources. It is hoped that these conclusions can add value to the flora of Rio Grande do Sul and its associated traditional culture, and therefore contribute to its conservation.

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Note: A list of the labiate species (both native and introduced) occurring in RS is available from Elaine Elisabetsky (elisasky@vortex.ufrgs. br) on request.

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PHLOMIC ACID IN LAMIOIDEAE SEED OILS

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Products with an allene structure (C=C=C) are very rare in nature because their synthesis and biosynthesis is rather difficult. The only major natural allene known to be widely occurring is neoxanthin, one of the yellow pigments in green leaves. On the other hand, allenes

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were known to occur in a number of seed oils since the early investigations of Bagby, Hagemann and coworkers (Bagby *et al.* 1965, Hagemann *et al.*, 1967). The allenes which were found by infrared spectroscopy were later characterized as a fatty acid with 18 carbon atoms and a Δ5,6*allene* structure (Fig. 1).

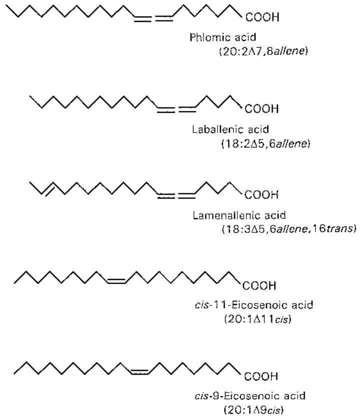

Fig. 1: Structures of laballenic, phlomic and lamenallenic acids and of the more significant 20:1 fatty acids, 20:1D9*cis* and 20:1D11*cis*.

This fatty acid was thoroughly investigated and characterized, and was called laballenic acid (Bagby *et al.*, 1965), because its occurrence was known only from plants belonging to the family Labiatae. With very few exceptions, the early work by Hagemann *et al.* (1967) showed that laballenic acid occurred only in members of what is now considered as the subfamily Lamioideae by Cantino *et al.* (1992).

In addition to laballenic acid, another allenic fatty acid was later discovered which had an additional double-bond in position Δ16 (Mikolajczak *et al.*, 1967). This fatty acid was called lamenallenic acid, because it was found only in the seed oils of species belonging to the genus *Lamium* (Mikolajczak *et al.*, 1967). This fatty acid so far has never been observed in any other Labiate seed oil outside genus *Lamium*. We too found it only in *Lamium* (*L. maculatum* being the only species investigated here), but not in other genera of Labiatae - except possibly for traces (see Table 1) requiring confirmation.

Our own work (Aitzetmüller, 1997; Tsevegüren *et al.*, 1997; Aitzetmüller *et al.*, 1997) on seed oil fatty acid fingerprints (Aitzetmüller, 1993) so far confirmed the occurrence of laballenic acid in seed oils of a number of species belonging to subfamily Lamioideae as defined by Cantino *et al.* (1992). With the exception of only one sample, obtained from Bulgaria and labelled as *Stachys alpina*, laballenic acid was present in all members of the Lamioideae investigated by us (about 20 spp.), and at the same time it was absent in all members of other Labiatae subfamilies. So we currently believe that the presence of allenic fatty acids is a highly characteristic feature of this one subfamily. (The single sample of *S. alpina* which proved the exception, may perhaps be due to a misidentification.)

Figure 2 shows a number of typical fatty acid fingerprints (Aitzetmüller, 1993) as obtained from a few members of the Labiatae plant family. As usual we have labelled only those fatty acids in the gas chromatograms of Figure 2, which are "unusual" in the sense that they do not occur in every normal seed oil. We believe that these "fatty acid fingerprints" are of chemotaxonomic value (Aitzetmüller, 1993; Aitzetmüller, 1995) and can be used in much the same way as one would use "leaf shape" or "pollen surface structure" in taxonomic work in botany. The presence or absence of these unusual fatty acids, at levels above a few tenths of a percent at least, is genetically determined (or, to be more exact, the presence or absence of the enzymes necessary for the biosynthesis of this particular fatty acid depends on the presence and expression of the appropriate genes coding for them). The presence of fatty acids with unusual structures in the seed oils of closely related genera can therefore also be considered to be an important indicator of phylogenetic evolution (Aitzetmüller, 1996).

Recently, phlomic acid was discovered and described (Aitzetmüller *et al.*, 1997) as a minor additional allenic fatty acid occurring in certain representatives of the subfamily Lamioideae. Phlomic acid (20:2Δ7,8*allene*) seems to be the chain-elongation product of laballenic acid, and it may have been synthesized by the addition of a C_2-unit to the carboxyl end of laballenic acid - *i.e.*, as usual in the chain-elongation of normal fatty acids. However, the chain-elongation of fatty acids which contain double-bonds in position Δ5 or Δ6 of the fatty chain so far was not known in plants (Aitzetmüller and Tsevegüren, 1994; Aitzetmüller *et al.*, 1997). The finding is also significant for another reason: The presence of small amounts of phlomic acid seems to be correlated with the occurrence of an unusual 20:1 fatty acid, 20:1Δ9*cis* or 20:1n-11. In most other seed oils, including those of most Labiatae, 20:1Δ11*cis* (or 20:1n-9) is the usual, and often the only, 20:1 fatty acid present. Only in those species, where small amounts of phlomic acid occur, the gas chromatographic peak representing 20:1Δ9*cis* is larger than the neighbouring peak of 20:1Δ11*cis*. In species where phlomic acid is absent, the peak size ratio of the two 20:1 fatty acids is always the other way round, or 20:1Δ9*cis* is not present at all.

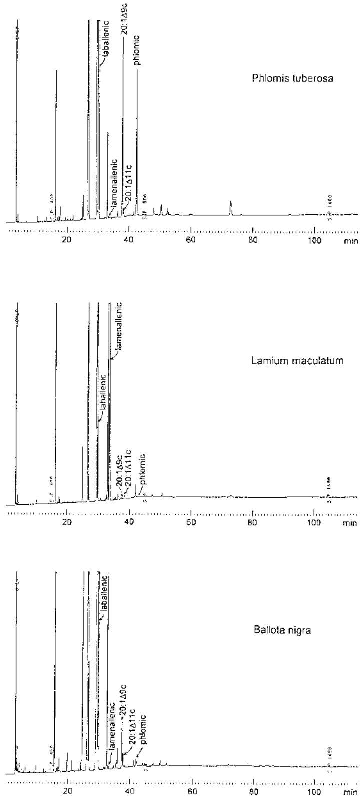

Fig. 2: Capillary gas chromatographic fatty acid fingerprints showing the presence of phlomic acid next to laballenic acid and 20:1D9*cis* in *Phlomis* and *Ballota*, and of lamenallenic acid in *Lamium*. (Unusual fatty acids only are labelled).

Table 1: Occurrence of laballenic acid (18:2Δ5,6*allene*), phlomic acid (20:2Δ7,8*allene*) and related fatty acids (in % of total fatty acids) in seed oils of selected members of Labiatae: Lamioideae (this investigation).

Plant species	fat in seed %	laballenic acid	lamen-allenic acid	20:1Δ9c	phlomic acid
Phlomis tuberosa	11.8	25.1	-	3.3	2.9
**Leonurus sibiricus*	28.5	18.1	[0.2][1]	1.3	0.3
Phlomis fruticosa	12.5	13.0	-	0.7	0.5
Marrubium vulgare	32.1	12.0	-	0.4	[0.2][1]
Sideritis hyssopifolia	34.0	11.8	-	0.2	0.1
Leonurus cardiaca	34.5	11.7	-	0.7	0.2
Panzerina canescens	16.0	10.3	-	1.4	0.2
Ballota nigra	30.4	10.1	-	0.6	0.1
Molucella laevis	32.9	9.1	[0.2][1]	0.3	[0.1][1]
Physostegia virginiana var. *alba*	41.1	8.6	-	0.6	0.3
Stachys recta	22.7	7.6	-	0.3	0.2
Stachys palustris	31.6	7.2	-	0.4	[tr.][1]
Lamiastrum galeobdolon	33.1	6.1	-	0.7	0.4
Stachys byzantina	32.0	4.5	-	0.2	[tr.][1]
Phomis samia hort.[2]	24.3	3.9	-	0.2	0.3
Lamium maculatum	35.0	2.7	8.8	-	-
Galeopsis tetrahit	30.8	2.7	-	0.2	[tr.][1]
Galeopsis speciosa	-	2.6	-	0.2	[0.1][1]

[1]tentative identification
[2]presumably *P. russelliana*

[*Editorial note: *Leonurus sibiricus*, mentioned above, almost certainly refers to *Leonurus japonicus* Houtt.]

The chemotaxonomic significance of the prescence or absence of phlomic acid in the Lamioideae is not yet known. This fatty acid was present - at very low levels - in several species examined (Table 1). It has not been found in *Lamium* and in a large number of other genera, where the seed oils contain laballenic acid. It was found, however, in one sample of *Lamiastrum galeobdolon* (Aitzetmüller *et al.*, 1997).

Cantino *et al.* (1992), in their revision of the Labiatae made no attempt to subdivide their subfamily Lamioideae into tribes or other subunits. The seed oil fatty acid patterns, however, may suggest this.

Acknowledgements

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Note added: If one of our readers could supply us with authentic *Lamium galeobdolon, L. amplexicaule* and *Stachys alpina* seed from an independent source, and/or with seeds of very close other relatives of *Lamium*, e. g. another *Lamiastrum* spp. or variety, we should like to investigate this, too:-
Please contact us at our E-mail address: aitzetm@uni-muenster.de ❑

BIBLIOGRAPHY OF RECENT TAXONOMIC PUBLICATIONS ON THE LAMIALES

The following list of publications has been abstracted from the Kew Record of Taxonomic Literature and from the Kew Economic Botany Bibliographic Database (November 1996 - February 1998), and we are, again, extremely grateful to the editors and compilers for their assistance in preparing this bibliography. Regrettably our selection from the Economic Botany Database is not complete due to space constraints (the original list ran to c. 80 pages!). As was the case for the last newsletter, some authors have sent us notification of publications not listed on the "Kew Record" and these have also been included. Where possible articles are listed under the applicable genus, or occasionally, tribe or family - and are arranged alphabetically. Where a number of references to different taxa in the Lamiales are made in a single paper they have tended to be listed under the "General" heading. Any title completely enclosed within brackets is the English translation of a title written in a "symbol" font such as Arabic, Chinese, Japanese or Russian. All diacritical marks have been removed to facilitate editing.

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