

Phylogeny and floral evolution of the Lysimachieae (Ericales, Myrsinaceae): evidence from ndhF sequence data

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Phylogeny and floral evolution of the *Lysimachieae* (*Ericales*, *Myrsinaceae*): evidence from *ndhF* sequence data

Abstract

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Earlier hypotheses of floral evolution in *Lysimachia* and related genera were tested by phylogenetic analysis of DNA sequences from the plastid gene *ndhF*. It is concluded that taxa with flowers provided with oil-producing trichomes are not advanced and buzz-pollinated species with anther cone are not ancestral as earlier proposed. Instead, species of *Lysimachia* with buzz-pollinated flowers belong to one derived clade, species with flowers provided with nectar producing trichomes belong to another, both groups having evolved from yellow flowered oil-producing ancestors. We also show that *Lysimachia* and most of its subgenera as well as the genera *Anagallis*, *Pelletiera* and *Asterolinon* are paraphyletic or polyphyletic as presently circumscribed, and that *Trientalis* and a few American species of *Lysimachia* are the closest relatives of the other species of *Lysimachia* + *Anagallis*, *Asterolinon*, *Pelletiera* and *Glaux*. The subgeneric classification and the biogeographical patterns in the group are briefly discussed.

Key words: *Anagallis*, *Asterolinon*, *Glaux*, *Lysimachia*, *Pelletiera*, *Trientalis*, cp DNA, pollination adaptation, parsimony, phylogeny.

Introduction

Lysimachia L. is traditionally placed in tribe *Lysimachieae* of the family *Primulaceae* (Pax & Knuth 1905), together with a few other genera, viz. *Trientalis* L., *Glaux* L., *Asterolinon* Hoffmanns. & Link, *Pelletiera* A. St.-Hil. and *Anagallis* L. (incl. *Centunculus* L.). The genera are small with only one or a few species. Exceptions are *Anagallis* with c. 30 species occurring mostly in Africa (Mabberley 1997), and the large and morphologically diverse genus *Lysimachia* with c. 180 species, most of which are found in China (Hu & Kelso 1996). *Lysimachia* displays a wide array of morphological variation, in growth habit, inflorescence structure and, most notably, also in floral morphology and pollination adaptation. Surprisingly in view of the large variation, there has been virtually no controversy regarding the circumscription of this genus, although Decrock (1901) considered it likely that *Lysimachia* had been the starting point leading

to the genera *Coris* L., *Glaux*, *Trientalis*, *Asterolinon* and *Anagallis*, the latter thought to be connected through *L. nemorum*. *Lysimachia* has attracted the attention of several workers focusing on anatomy, morphology, classification or floral biology. Handel-Mazzetti (1928) recognized five subgenera and his work is still acknowledged as the most important contribution to the classification of *Lysimachia*, although it has been modified considerably by later authors to comprise six subgenera: *Idiophyton*, *Heterostylandra*, *Lysimachia* (*Eulysimachia*), *Lysimachiopsis*, *Naumburgia* and *Palladia* (Chen & Hu 1979, Bennell & Hu 1983). Following Handel-Mazzetti, most authors have perceived *L.* subg. *Idiophyton* with its buzz-pollinated flowers with an anther cone as the ancestral group of the genus. Vogel (1986) discussed the floral evolution in *Lysimachia* based on variation in pollination strategies in different species and considered flowers with nectaries as primitive in relation to flowers with oil producing trichomes.

In recent years it has been demonstrated that *Lysimachia* and the other members of the *Lysimachieae* belong to the *Myrsinaceae* and not to *Primulaceae*, and the monophyly of *Lysimachia* has also been questioned in a series of papers (Anderberg & Ståhl 1995, Källersjö & al. 2000, Mast & al. 2001, Martins & al. 2003). The idea of a close relationship between *Myrsinaceae* and *Lysimachia* has been put forward earlier, but then with the frutescent *Myrsinaceae* as the primitive ancestral group (Handel-Mazzetti 1928, Wettstein 1935, Ray 1956, Cronquist 1981, Judd & al. 1994, Takhtajan 1997). In contrast, the previous studies have shown that the tropical and subtropical *Myrsinaceae*, which are shrubs or small trees, have evolved from temperate herbaceous primuloid ancestors. Compared to the commonly recognized view of family delimitation (e.g., Pax 1889, Pax & Knuth 1905, Cronquist 1981, Takhtajan 1997), the reclassification proposed by Källersjö & al. (2000) changed the text-book circumscription of all the three families *Primulaceae*, *Myrsinaceae* and *Theophrastaceae* and also pointed out the need for a fourth family, *Maesaceae*, which was later formally described (Anderberg & al. 2000).

Based on the findings of Källersjö & al. (2000), Hao & Hu (2001) analysed relationships in *Lysimachia* by cladistic analysis of morphological data. In their study, they found *L. crispidens* Hemsl. (subg. *Heterostylandra*) to be sister to the other taxa, but bootstrap support for most nodes, particularly the lower ones in their tree, were very low. Hao & al. (2004) analysed sequence data from *trnL-F* and nrITS data and again found *Lysimachia* to be paraphyletic. They concurred that *Glaux* evolved from an ancestor in *Lysimachia*, but rejected the idea of Anderberg & Ståhl (1995) and Källersjö & al. (2000) that *Anagallis*, *Asterolinon* and *Trientalis* were derived from within that genus. Their study was focussed on relationships within *Lysimachia*, included only a few species of other genera of *Lysimachieae* and did not discuss the interesting issue of floral evolution in the group.

In our present paper we have addressed the following questions based on the phylogenetic analysis of a protein coding gene (*ndhF*): (1) If *Lysimachia* is paraphyletic, what is the phylogenetic status of the other genera of the *Lysimachieae*? (2) Which are the major clades in the *Lysimachieae*? (3) Based on the phylogeny, what can be concluded regarding trends of floral evolution as outlined by Vogel (1986)?

Material and methods

Taxa. – For our analysis we tried to include representatives from as many species as possible from the recognized genera and subgenera of the *Lysimachieae* (Anderberg & Ståhl 1995, Källersjö & al. 2000), in some cases more than one sequence of each. The *ndhF* data set included 100 sequences, 86 of them belonging to our primary study group, 16 to other groups of the *Myrsinaceae*, *Primulaceae*, *Theophrastaceae* and *Maesaceae*. The included taxa are listed in Table 1, see Appendix. Observations of morphological characters discussed in the text are based on investigation of herbarium material and of living plants in the field.

Molecular methods. – DNA was extracted from leaves taken from herbarium specimens, material dried in silica gel or from living plants. Leaves were ground in liquid nitrogen with mortar and pestle and subsequently treated with the DNEasy plant DNA extraction kit from Qiagen (Qiagen,

Valencia, California, USA), following the manufacturer's protocol. The *ndhF* gene from the chloroplast genome was amplified using PCR with 10 µmol/L primers in 25-µL reactions using "Ready-to-go" PCR beads from Pharmacia Biotech (Amersham Pharmacia Biotech, Uppsala, Sweden) following the manufacturer's standard protocol. The thermal cycling profile was 95 °C for 5 min, followed by 35 cycles of 95 °C for 30 sec, 50°-60 °C for 30 sec, 72 °C for 2 min and finally 72 °C for 8 min. For sequencing reactions the "Big Dye Terminator Sequencing" kit (Applied Biosystems, Warrington, Cheshire, UK) was used and fragments were separated on an ABI377 from Applied Biosystems. Primers used for PCR and for sequencing were those used by Källersjö & al. (2000). Sequences were assembled and carefully checked with the Staden software (Staden & al. 1998) and aligned manually with the BioEdit software (Hall 1999). The 63 new sequences have been submitted to GenBank (Accession numbers AY839951-AY840013) and voucher information for material used is presented in Table 1.

Phylogenetic analysis. – The *ndhF* data set was analysed with parsimony jackknifing (Farris & al. 1996) using the computer software "Xac" (Farris 1997) with the following settings: 1000 pseudo-replicates, each with branch swapping and ten random addition sequences. The analysis of the *ndhF* gene was performed with all codon positions included. Källersjö & al. (2000) has shown that in the *Primulaceae-Myrsinaceae* the third position of *ndhF* provided most of the informative data and gave more structure to the resulting tree. In this group they demonstrated that including the third position gave a higher number of well supported groups than analyses of first and second codon positions only, or of transversions only.

For all analyses *Maesa* Forssk. was used as outgroup (Farris 1972) because it has been shown in previous studies (Källersjö & al. 2000, Anderberg & al. 2002) to be the sister group of *Theophrastaceae*, *Primulaceae* and *Myrsinaceae*. The alignment was unproblematical and without informative indels. Gaps were treated as missing information.

Results

The 100 aligned *ndhF* sequences comprised 1965 characters, 464 (23.6 %) of which were informative. The consensus tree from the "Xac" analysis is shown in Fig. 1. It should be noted that only jackknife support frequencies >50 % are shown.

The two species of *Trientalis* form one small clade. The three species *Lysimachia ciliata*, *L. hybrida* and *L. quadriflora* form a second clade (100 %), which is sister to *L. andina* but only with low support (51 %). This group is part of an unresolved trichotomy together with *Trientalis* and the large group comprising the rest of the *Lysimachieae* taxa.

The remaining taxa at the next higher node form one large clade with weak support (53 %). Resolution within this large clade is high, except for the first level where there are five principal clades.

In the first of these, *Lysimachia nummularia* and *L. punctata* are sisters (100 %).

The second clade is formed by *Anagallis arvensis*, *A. monelli* (i.e. *Anagallis* s.str.), the two *Asterolinon* species, the two *Pelletiera* species, as well as *Lysimachia nemorum* and *L. serpyllifolia* from *L.* subg. *Lysimachia* sect. *Lerouxia*. *Asterolinon linum-stellatum* is closest to *Pelletiera wildpretii*, whereas *Asterolinon adoëense* is closest to *Anagallis arvensis*, *A. monelli* and the two *Lysimachia* species.

The third clade (99 %) consists of *Lysimachia alpestris* (*L.* subg. *Lysimachia* sect. *Rosulatae*) together with all the species from *L.* subg. *Idiophyton*.

The fourth clade is composed of the remaining *Anagallis* species, with *A. tenella* as sister to the rest (100 %).

The fifth and last of the clades in the pentatomy (89 %) contains all the remaining *Lysimachia* species and the single species of the monotypic genus *Glaux*. The clade with the three species *L. vulgaris*, *L. thyrsoflora* and *L. terrestris* (100 %) is sister to that comprising the remaining taxa (79 %), where the species from *L.* subg. *Palladia* (*L. africana*, *L. atropurpurea*, *L. candida*, *L.*

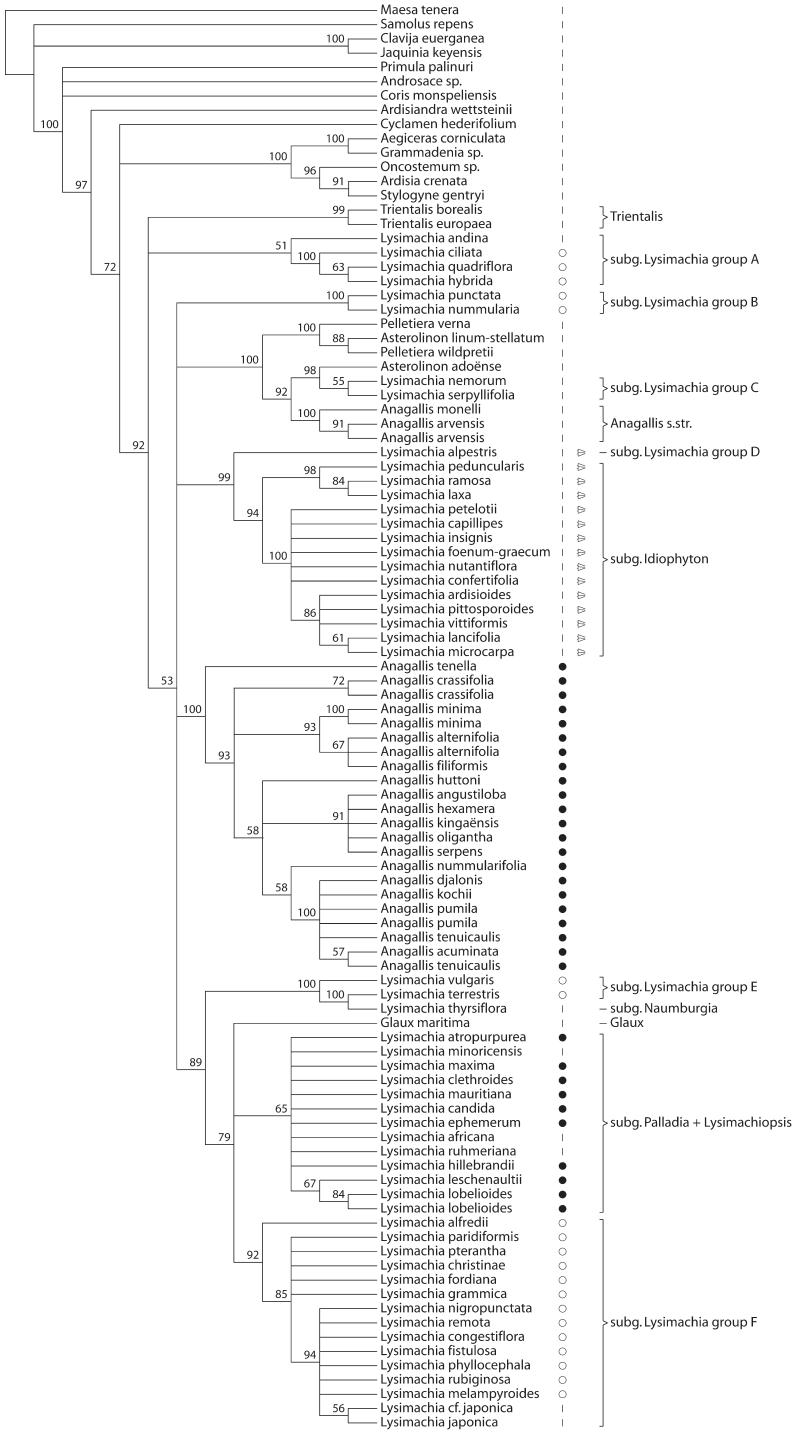


Fig. 1. Jackknife tree obtained from the Xac analysis of *ndhF*. Jackknife support values (> 50 %) are indicated above the nodes. ○ = Flowers with oil-producing triebones; ● = flowers with nectar-producing triebones.

clethroides, *L. ephemerum*, *L. leschenaultii*, *L. lobelioides*, *L. minoricensis*, *L. mauritiana*, *L. ruhmeriana*) and the species from *L. subg. Lysimachiopsis* (*L. hillebrandii*, *L. maxima*) form one group (65 %), while the majority of species from *L. subg. Lysimachia* sect. *Nummularia* form another group (92 %), in which *L. alfredii* is sister to the rest of the species, i.e. *L. christinae*, *L. congestiflora*, *L. fistulosa*, *L. fordiana*, *L. grammica*, *L. japonica*, *L. cf. japonica*, *L. melampyroides*, *L. nigropunctata*, *L. paridiformis*, *L. phyllocephala*, *L. pterantha*, *L. remota* and *L. rubiginosa*. *Glaux* is also part of the fifth clade and the jackknife support for its position together with the two *Lysimachia* groups is 79 %, but its exact relationships cannot be ascertained at this point.

Discussion

Our present results corroborate the conclusion that *Lysimachia* is paraphyletic, since there is strong support in the jackknife analysis for *Glaux* being an apetalous, derived member of that genus. Furthermore, there is also strong support for a closer relationship between the species *L. nemorum* and *L. serpyllifolia* to *Anagallis* s.str. than to any of the other *Lysimachia* species, and this will be discussed further in a later paragraph. The genus *Asterolinon* is shown to be polyphyletic and *Pelletiera* to be paraphyletic. The paraphyly of *Lysimachia* is thus well established and also that the traditional subgeneric classification needs to be re-evaluated.

Status of subgenera in *Lysimachia*

***Lysimachia* subg. *Lysimachia*.** – Species placed in this group have yellow flowers that are solitary in leaf axils or arranged in racemes, panicles or heads. The anthers open with slits and have longer filaments that form a thin ring adnate to the corolla base. Characteristic of most taxa are the flowers that have oil-producing trichomes in various places on the corolla and the anther filaments. The taxa are rather similar in floral morphology, at least when compared to the species of other subgenera, but the similarity seems to be symplesiomorphic. Our study included species from several of the subgeneric sections recognized by Chen & Hu (1979), i.e. *L. sect. Seleucia*, *Theopyxis*, *Nummularia*, *Lerouxia*, *Lysimachia* and *Rosulatae*, and found them dispersed in several places in the tree, belonging to different monophyletic groups and thus not being closely related (Fig. 1, groups A-F). It is evident that the subgenus *Lysimachia* is strongly paraphyletic in its present circumscription, which becomes interesting in the light of Vogel's (1986) hypotheses of floral evolution in *Lysimachia*, as most species have flowers with oil-producing trichomes.

The two sections *Seleucia* (*Lysimachia ciliata*, *L. quadriflora* and *L. hybrida*) and *Theopyxis* (*L. andina*) are both American groups and were found to have a basal but yet unresolved position in the *Lysimachieae* complex together with *Trientalis*. Ray (1956) considered *Trientalis* with its hexa- or heptamerous white flowers as a distinct genus, but noted its similarity to the species of the North American *L. sect. Seleucia* in other respects. The whorled leaves and axillary flowers in *Trientalis* were interpreted as an extreme of the subverticillate condition in that group. Species belonging to *L. sect. Seleucia* are distinguished by pollen grains with indistinct endoapertures, referred to as tricolporoidate by Bennell & Hu (1983), and often have staminodial structures in the corolla. Hao & al. (2004) did not include any taxon from *L. sect. Theopyxis* in their study, but like us they found *L. ciliata* in a basal position in relation to other *Lysimachia*.

The species placed in *Lysimachia* sect. *Nummularia* in the sense of Chen & Hu (1979) do not form a monophyletic group (Fig. 1, subg. *Lysimachia* groups B, F) and the majority of species included in that section (i.e., *L. alfredii*, *L. congestiflora*, *L. fistulosa*, *L. melampyroides*, *L. nigropunctata*, *L. phyllocephala*, *L. pterantha*, *L. remota* and *L. rubiginosa*) do not group with the type of the section (*L. nummularia*). Instead, they form a trichotomy together with *Glaux* and the

white, pink or red flowered species of subgenera *Palladia* and *Lysimachiopsis*. *L. nummularia* and *L. punctata* form another group that has unresolved relationships to many of the other *Lysimachia* clades. The analyses by Hao & al. (2004) also indicated that *L. sect. Nummularia* is paraphyletic.

Lysimachia sect. *Lerouxia* comprises only three species, two of which were included in our study (*L. nemorum* and *L. serpyllifolia*) and were found to be very closely related to *Anagallis* s.str. (*A. arvensis*, *A. monelli*). The same relationships were found by Martins & al. (2003) in their analysis of ITS sequence data, whereas Hao & al. (2004) found *L. nemorum* to be sister to *L. nummularia*. This seems to be a conflict, but a digital image of the voucher (at IBSC), kindly provided by Hao, revealed that the herbarium voucher is a mix of both *L. nemorum* and *L. nummularia*, and their sequence of *L. nemorum* probably originates from the *L. nummularia* material. Several morphological features also support the systematic position of *L. nemorum* and *L. serpyllifolia* near *Anagallis* (Fig. 2 A-C). We will return to discuss this matter below in the context of that genus and its relatives. Both Hao & al. (2004) and our present investigation place *L. alpestris* of the monotypic section *Rosulatae* as sister to the species of subgenus *Idiophyton*, a group with which it shares the presence of an anther cone and buzz-pollinated flowers. The former was for unclear reasons placed close to the Hawaiian species in *L. subg. Lysimachiopsis* by Handel-Mazzetti (1928), but moved to *L. subg. Lysimachia* by Chen & Hu (1979).

Lysimachia sect. *Lysimachia* includes the type of the genus, *L. vulgaris*, and a few more species, e.g., *L. terrestris*. In our analysis, as well as in that of Hao & al. (2004), the two species of *L. sect. Lysimachia* form a strongly supported group together with *L. thyrsoflora*, the only species of subgenus *Naumburgia*.

***Lysimachia* subg. *Naumburgia*.** – It is sometimes (e.g., Fedorov 1981) treated as a separate monotypic genus. It is diagnosed by hexa- or heptamerous flowers aggregated in dense head-like racemes in the axils of the middle cauline leaves. Like in most other groups the anthers open with slits and have long filaments attached to the base of the corolla. As mentioned earlier, we have shown that *L. thyrsoflora* is closely related to *L. vulgaris* and *L. terrestris* of *L. subg. Lysimachia* and it also hybridizes with the latter (Ray 1956). Its unique features are apparently autapomorphies, and since it is closely related to the species that provides the type of the name *Lysimachia* (*L. vulgaris*), the derived *L. thyrsoflora* cannot be recognized in a separate subgenus. Seeds of all the three species of this small clade have a characteristic tawny, almost spongy seed coat layer, which is possibly a floating device evolved as an adaptation to living in wet habitats, and a synapomorphy that provides morphological support for their close evolutionary relationship.

***Lysimachia* subg. *Idiophyton*.** – It is characterized by having yellow buzz-pollinated flowers without nectaries, reflexed corolla lobes, basifixed poricidal anthers forming a protruding cone and short filaments adnate to the corolla base. Handel-Mazzetti (1928) viewed *L. insignis* as a probable link between *Lysimachia* and the *Myrsinaceae* proper, where pollen flowers like those in *L. subg. Idiophyton* are found in many genera (e.g., *Ardisia*), and the similarity in flower morphology is reflected by the species epithet of *L. ardisioides*. The species of this subgenus are easy to recognize. *L. alpestris* of *L. subg. Lysimachia*, which we found with strong support to be the sister group of *L. subg. Idiophyton*, shares the presence of an anther cone, buzz-pollination and the same kind of pollen (Bennell & Hu 1983) with the other species of this clade. The pollen in subgenus *Idiophyton* is comparatively small compared to other groups, something that Bennell & Hu (1983) interpreted as primitive, whereas Vogel (1986) viewed the small pollen in *L. subg. Idiophyton* as a mere adaptation to buzz-pollination. This group has also been conceived as the ancestral complex from which the group of Hawaiian endemics (*L. subg. Lysimachiopsis*) have evolved (see below), but our results show that these groups are not closely related.

***Lysimachia* subg. *Palladia* and *Lysimachiopsis*.** – There is a close relationship between the species of subgenus *Palladia* and the Hawaiian endemic group of species constituting *L. subg. Lysimachiopsis* (Marr & Bohm 1997). The former have white or sometimes pink or red flowers arranged in terminal racemes, which give them a characteristic appearance. Both groups have anthers opening with slits and anther filaments adnate some distance up the corolla. Subgenus

Lysimachiopsis is diagnosed by red, red-tinged or whitish flowers, but also by a fruticose growth habit and a corolla with a variable number of corolla lobes. They are unique in having tetracolporate pollen (Huynh 1970, 1971, Bennell & Hu 1983) in contrast to the tricolporate pollen found in all other groups. The monophyly of this Hawaiian group has never been questioned, but several suggestions have been put forward as to its origin. As mentioned above, Handel-Mazzetti (1928) hypothesized the yellow flowered Chinese species *L. alpestris* to be their closest relative, whereas Hu (pers. comm.) proposed a close relationship with the species belonging to subgenus *Idiophyton* occurring in SE Asia, particularly *L. laxa* (Marr & Bohm 1997). Evidence in support of such an assumption was the shared presence of basifixed anthers and sometimes lignified stems. In their morphological analysis, Hao & Hu (2001) found the Hawaiian taxa, represented by *L. hillebrandii* and *L. forbesii* Rock, to be part of the same clade as the species of *L. subg. Idiophyton*, but Hao & al. (2004) later concluded that the Hawaiian species were in fact closely related to the taxa of *L. subg. Palladia*. We also found subgenera *Palladia* and *Lysimachiopsis* to be very close and to form a clade, corroborating their results, but where they obtained strong support for *L. subg. Lysimachiopsis* as sister to *L. subg. Palladia*, our data are inconclusive whether they are sister groups or if one is part of the other. Although white flowers may be found also in other groups, a white or red coloured flower seems to be a synapomorphy for this group, as its closest relatives are groups with predominantly yellow flowers. Nectar producing trichomes, which are found in flowers of species from these two groups, is evidently another synapomorphy and an indication of a common ancestry, and not due to parallel evolution as assumed by Vogel (1986). Apart from species of these two groups, flowers with nectar producing trichomes are only found in *L. crispidens* of the monotypic *L. subg. Heterostylandra*. Like most species of subgenus *Palladia*, this species also has white flowers and anther filaments basally forming a thin ring adnate to the corolla, but is unique in having almost heterostylous flowers. This species was found to be sister to the subgenera *Palladia* and *Lysimachiopsis* by Hao & al. (2004), but was unfortunately not available to us.

Glaux

Our study gives strong support for *Glaux* being nested within *Lysimachia* in a group together with the species from the subgenera *Palladia* + *Lysimachiopsis* and parts of *L. subg. Lysimachia* (viz., sect. *Lysimachia*, sect. *Nummularia* p.p.). The flowers in *Glaux* have stamens alternating with the perianth lobes, whereas species with distinct corolla and calyx have stamens opposite the corolla lobes. Therefore, the single-rowed perianth in *Glaux* is best interpreted as being a coloured calyx, and the corolla lost by reduction. Reduction in corolla size is also evident in *Asterolinon* and *Pelletiera*, but they belong to a different clade and in both cases reduction has evolved independently. Superficially, *Glaux* somewhat resembles a small *L. mauritiana* from *L. subg. Palladia*, with which it shares fleshy leaves, but these two species are not particularly closely related. Hao & al. (2004) received weak support for *Glaux* as sister to *L. subg. Lysimachia* sect. *Nummularia*. At this point it seems clear that *Glaux* cannot be maintained as a separate genus, but even so, this circumpolar apetalous species still holds an isolated position and its closest relative among the *Lysimachia* is yet to be found.

Anagallis, Asterolinon, Pelletiera and Lysimachia sect. *Lerouxia*

The conclusion that *Anagallis* is derived within *Lysimachia* (Anderberg & Ståhl 1995, Källersjö & al. 2000) is corroborated by our present study. Hao & al. (2004) included only one *Anagallis* species, reaching the conclusion that *Anagallis* was not nested within *Lysimachia*, but our analysis indicates that it is. Our analysis includes 19 species of *Anagallis* that are shown to belong in two different clades, each with maximum support (100 %). *A. arvensis* and *A. monelli* share a common ancestor with the two small genera *Pelletiera* and *Asterolinon* and with the species of *L. subg. Lysimachia* sect. *Lerouxia* (*L. nemorum*, *L. serpyllifolia*). Thus *Anagallis arvensis* and *A. monelli* are more closely related to species of other genera than to the other species of *Anagallis*.

Similarly *L. nemorum* and *L. serpyllifolia* are more closely related to *Anagallis* s.str., *Asterolinon*

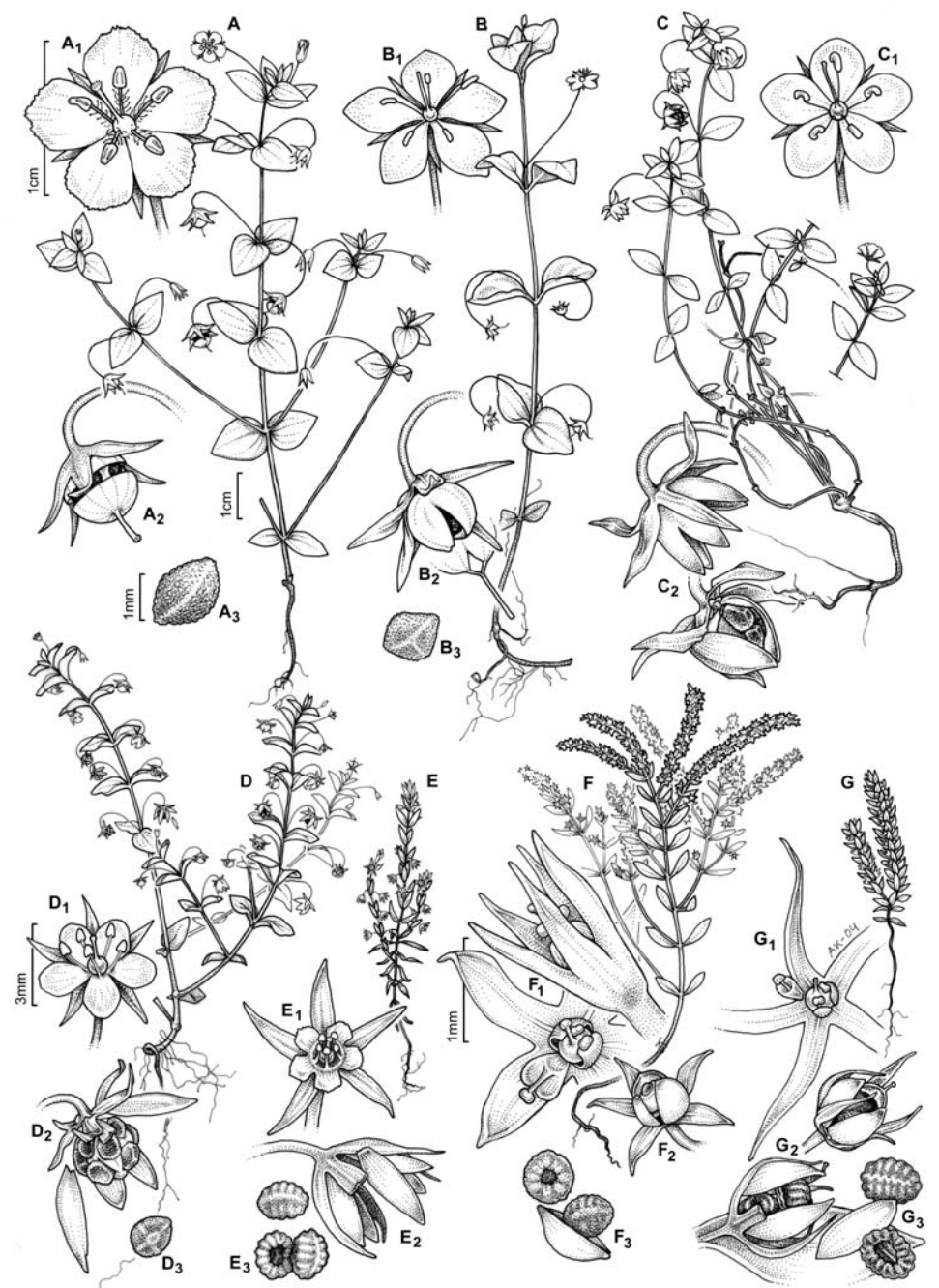


Fig. 2. A-A3: *Anagallis arvensis* – A: habit; A1: flower with 5-merous sympetalous corolla longer than calyx; A2: operculate capsule with style on lid; A3: papillose seed. – B-B3: *Lysimachia nemorum* – B: habit; B1: flower with 5-merous sympetalous corolla longer than calyx; B2: capsule opening with valves (and distal portion of younger stage with style still in place); B3: papillose seed. – C-C2: *L. serpyllifolia* – C: habit; C1: flower with 5-merous sympetalous corolla longer than calyx; C2: capsule opening with valves (and transection of cap-

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and *Pelletiera* than to any other species of *Lysimachia*. *Anagallis arvensis* and *A. monelli* are sisters and together form the sister group of *Asterolinon adoënsense* and the *Lysimachia* species pair. The clade with these five taxa has *Asterolinon linum-stellatum* and the two *Pelletiera* species as their sister group. *Anagallis arvensis* and *A. monelli* belong to *Anagallis* s.str., a species complex with relatively unclear species delimitation. The *Anagallis* species belonging to the other clade are from *A.* subg. *Jirasekia* or *A.* subg. *Centunculus* (Taylor 1955), and it was recently demonstrated that circumscissile capsules have evolved in parallel in the two groups and that they in other characters differ from the species of *A.* subg. *Anagallis* (Manns & Anderberg 2005).

Asterolinon is polyphyletic, the only character diagnosing the genus seems to be a small size and a pentamerous corolla being shorter than the calyx (Fig. 2D-E), something that Leblebici (1978) considered insufficient to merit generic separation from *Lysimachia*. The fact that the two species of *Asterolinon* do not constitute a monophyletic group is not surprising (cf. Anderberg & Ståhl 1995: 1715). *A. adoënsense* shares a very characteristic aspect with *Anagallis* s.str. (Fig. 2A) and with *Lysimachia nemorum* (Fig. 2B), to some extent also with *L. serpyllifolia* (Fig. 2C). They all have opposite, ovate to cordate sessile leaves and solitary flowers on thin pedicels from the leaf axils, which after flowering curve like arcs, and have globose capsule with spreading sepals at the base. The main difference is found in the size and colour of the corolla and in the way the capsules open. The two *Anagallis* species of this clade have red or blue flowers and capsules opening with a circular lid. *L. nemorum* and *L. serpyllifolia* (*L.* sect. *Lerouxia*) and *Asterolinon adoënsense* have yellow flowers and capsules opening with valves. Operculate capsules cannot be used to diagnose *Anagallis* s.str. as they occur also in the second, more distantly related clade of *Anagallis* species, and a capsule opening with a lid is most parsimoniously interpreted as a parallelism in these groups. The fruit opening mechanism has changed several times and seems to be a homoplasious character. Manns & Anderberg (2005) showed that even if the two *Anagallis* clades were sisters, then it would be more parsimonious with two gains of lid capsule than with the corresponding gain and loss events that must have occurred in the ancestors of the *Asterolinon-Pelletiera* clade and in the clade with *Asterolinon adoënsense* as well as in the two species from *Lysimachia* sect. *Lerouxia*.

The widely distributed Mediterranean *Asterolinon linum-stellatum*, which provides the type of the name *Asterolinon*, forms a strongly supported clade with the two species of *Pelletiera* (100 %). The three species are all small, delicate annuals with lanceolate or narrowly elliptical leaves and a corolla much shorter than the calyx (Fig. 2E-G). In *A. linum-stellatum* the corolla is pentamerous and sympetalous, whereas both *Pelletiera* species have five sepals but only three free corolla segments. The seeds in these three species have a unique appearance (Fig. 2E3, F3, G3) with wrinkled, minutely papillose surface and a deeply sunken hilum. This constitutes a synapomorphy for the three species, whereas the seeds of *A. adoënsense* have the plesiomorphic character state, being more or less smooth with a finely papillose surface just like in *Anagallis arvensis*, *Lysimachia nemorum* and many other species groups in *Lysimachia*. Our analyses indicate that *Pelletiera* is

sule showing seeds). – D-D3: *Asterolinon adoënsense* – D: habit; D1: flowers with 5-merous sympetalous corolla somewhat shorter than calyx; D2: capsule with valves falling apart; D3: papillose seed. – E-E3: *A. linum-stellatum* – E: habit; E1: flower with 5-merous sympetalous corolla shorter than calyx; E2: capsule with valves falling apart; E3: wrinkled seeds with sunken hilum. – F-F3: *Pelletiera wildpretii* – F: habit; F1: two flowers, natural half-closed shape, the lower opened showing 3-merous choripetalous corolla much shorter than calyx, and one petal folded backwards; F2: capsule with valves falling apart; F3: wrinkled seeds with sunken hilum, and one free capsule valve. – G-G3: *P. verna* – G: habit; G1: flower with 3-merous choripetalous corolla much shorter than calyx, one petal lifted out; G2: two capsules with valves falling apart; G3: wrinkled seeds with sunken hilum, and one free capsule valve. – Material: A: *Nicholson s.n.* (S), A1: *Segelberg 11458/14* (S), A2-A3: *Cacciato s.n.* (S), B-B3: *Geesteranus 4187* (S); C: *Segelberg s.n.* (S); C1: *Kretaexpeditionen 1989 s.n.* (S), C2: *Bornmüller 1092* (S); D-D3: *Ambjörn 383* (S); E-E1: *Wall 21* (S); E2-E3: *Bremer 1861* (S); F-F3: *Paulo s.n.* (S); G: *Malme 46* (S), G1-G3: *Urtubey & Tremetsberger 119* (S). – Five different scales are used: scale bar in A is also for B, C, D, E, F, G, scale in A1 is also for A2, B1, B2, C1, C2, scale bar in A3 is also for B3, E1-E3, F2-F3, G2-G3, scale bar in D1 is also for D2 and scale bar in F1 also for F2 and G1.

not monophyletic and that the Macaronesian *P. wildpretii* is closer to the likewise Old World species *A. linum-stellatum* (88 %) than to the other species, *P. verna* from South America. Our results also indicate that the character combination of a trimerous, choripetalous corolla, triandrous flowers and a trivalvate capsule have evolved independently in the two *Pelletiera* species, or else that *A. linum-stellatum* has regained a pentamerous flower with a sympetalous corolla. This is surprising considering that the flowers in the group seem to form a reduction series. Valdés (1980) found that of the three free corolla segments in *P. wildpretii* one is emarginate and another bilobed, which reflects the condition in its closest relative. This seems to be based on the notion of a successive reduction of the flowers from a sympetalous, pentamerous and medium short corolla to a choripetalous, trimerous corolla with extremely short petals. Taking into consideration also the triandrous flowers and the trivalvate capsule it would thus be easier to assume that *A. linum-stellatum* was sister to the two *Pelletiera* species. Also from a biogeographical standpoint the obtained results are difficult to explain. The question of how the disjunction between the two *Pelletiera* species originated is puzzling. Species of *Anagallis* s.str. are chiefly Mediterranean but occur as weeds elsewhere. *Asterolinon adoënsis* is East African, *L. nemorum* and *L. serpyllifolia* are European and the third species of *L. sect. Lerouxia* (*L. cousiniana* Coss.) is North African. Thus, all species from this clade, except *P. verna* and the cosmopolitan weeds are Old World taxa. At this point we do not know if their transatlantic disjunction is a relict of an ancient Laurasian distribution, or if the common ancestor of *P. wildpretii*-*A. linum-stellatum* was introduced in the Old World by dispersal. The relationship between the two *Pelletiera* species and *A. linum-stellatum* that we found here is certainly an interesting issue that would need further investigation.

Apart from *Pelletiera verna*, only a few species of *Lysimachieae* grow in the New World, i.e. the white-flowered *Trientalis* with two similar but polymorphic and partly sympatric species, *T. borealis* and the almost circumpolar *T. europaea* in North America, the four likewise white flowered and morphologically distinct species of *Lysimachia* sect. *Theopyxis* in South America (*L. andina*, *L. mexicana* R. Knuth, *L. sertulata* Baudo and *L. steyermarkii* Standl.), the yellow flowered *L. sect. Seleucia* in North America (e.g., *L. ciliata*, *L. hybrida*, *L. quadriflora*) as well as a few species of *Anagallis* (*A. alternifolia* and *A. filiformis*). Species of other clades are almost entirely distributed in Asia, where the vast majority of *Lysimachia* species occur, and only relatively few species in Europe and Africa. Exceptions are *L. subg. Lysimachiopsis* that is endemic to Hawaii and the species of *Anagallis*, *Asterolinon*, *Pelletiera* and *L. sect. Lerouxia* that have almost all species concentrated to Africa and Europe. The Hawaiian endemics are closely related to the species of *L. subg. Palladia* and to subg. *Heterostylandra*, and it seems safe to assume that the ancestors of the Hawaiian group came from E Asia and later evolved in the islands. Interestingly, the only *Lysimachia* in Hawaii that is not part of the endemic *L. subg. Lysimachiopsis* is the E Asian species *L. (subg. Palladia) mauritiana*.

Floral evolution

Like *Trientalis*, the species of the South American *Lysimachia* subg. *Theopyxis* have white flowers without any apparent attractive agents, whereas the species of *L. sect. Seleucia* have yellow flowers with glandular trichomes. Many species of *L. subg. Lysimachia* sect. *Lysimachia* and sect. *Nummularia* are similar to the species of *L. sect. Seleucia* in being provided with minute glandular hairs on the inside of the corolla and on the base of the stamens. Vogel's (1986) thorough investigation of flowers in *Lysimachia* showed that the floral glands are of two different kinds although they were superficially similar in appearance, one producing oil and the other producing nectar. Vogel investigated the anatomy and ultrastructure of the floral glands and made microchemical analysis of gland exudates in no less than 102 species of *Lysimachia*. Flowers with oil-producing glandular hairs were found in a number of *Lysimachia* species in *L. subg. Lysimachia* (e.g., *L. sect. Seleucia*, sect. *Lysimachia* and sect. *Nummularia*). In these species, bees collect the oil for their nesting and breeding needs while carrying out pollination. Without exception, oil-producing flowers were found to be yellow, with more or less open corolla and with basally connate anther filaments. Vogel (1986) hypothesized that the oil-producing flowers

formed one group whereas the nectar-producing taxa were dispersed within several different groups. Vogel's supposition is not supported by our results, since species with oil-producing flowers do not form a single clade (Fig. 1, open circles).

Our analysis reveals a pattern of distribution of oil-producing glandular hairs and nectar glands in *Lysimachia* that is new and conflicting with earlier hypotheses. Although future investigations may find exceptions, the present monophyletic groups are diagnosed by the same floral type. Vogel's study revealed that flowers with nectar-producing trichomes occur only in the three subgenera *Palladia*, *Heterostylandra* and *Lysimachiopsis* and the flowers in these groups are either white or red, but not yellow. Nectar-producing taxa are found in an array of taxa in the *Primulaceae*, as circumscribed at the time of Vogel's investigation, and he considered the presence of trichome nectaries also in some perennial *Anagallis* as an indication that they were plesiomorphic in *Lysimachia*. As a consequence, Vogel considered the possibility that the nectar-producing trichomes found in the Hawaiian endemic taxa and those found in *L.* subg. *Palladia* were due to parallel evolution and not evidence of monophyly. Contrary to Vogel, we conclude that nectar-producing trichomes is a derived trait within the *Lysimachieae* and their occurrence in the clade formed by species belonging to *L.* subg. *Palladia* and *Lysimachiopsis* is evidence that at least this group evolved from ancestors with yellow, oil-producing flowers. In the perennial *Anagallis* species (i.e., the clade with *A. tenella*, etc.) the nectar trichomes have apparently evolved independently from those occurring in *Lysimachia*. Vogel (1986) described several cellular and ultrastructural differences between the oil glands and the nectar glands and, considering their parallel evolution, it would be interesting to investigate if also nectar glands in *Lysimachia* and in *Anagallis* could be shown to differ.

Besides the oil and nectar trichome flowers, there is one *Lysimachia* species group to which the third kind of flowers is confined. Species of *L.* subg. *Idiophyton* generally have a characteristic floral morphology with poricidal basifixed anthers on very short filaments, forming a protruding cone, and they often have somewhat recurved corolla lobes. In our tree all species with these characteristics form a single clade. This flower type is buzz-pollinated and typical of species belonging to *L.* subg. *Idiophyton*. Handel-Mazzetti (1928) thought *L. insignis* of this group was the most primitive species of the genus and a link to *Myrsinaceae*, but our results show that *L. insignis* is an integrated part of *L.* subg. *Idiophyton* and not an ancestral taxon from which the remaining species of that clade have evolved. Vogel (1986) was convinced that the lack of glandular floral trichomes in *L.* subg. *Idiophyton* contradicted the assumption put forward by Handel-Mazzetti, as such a group could not be the predecessor of the oil- or nectar-producing taxa. Our results agree with Vogel's view, as the species of *L.* subg. *Idiophyton* form a separate clade that represents a separate evolutionary lineage, likely evolved from oil-producing ancestors. It is neither ancestral to oil-producing nor to nectar-producing taxa in *Lysimachia* as hypothesized by many earlier workers. The relationships of *L.* subg. *Idiophyton* are, however, not fully understood as the support for the basal nodes are weak. Our analyses have the American group with *L. ciliata*, *L. quadriflora* and *L. hybrida* on the next lower node. The analysis of Hao & al. (2004) found weak support for *L.* subg. *Idiophyton* as the sister group of all other *Lysimachia* groups, i.e., all except the American *L. ciliata* and *L. quadrifolia*, and thus it is corroborated that *L.* subg. *Idiophyton* is not basal.

Trientalis, *Asterolinon*, *Pelletiera*, *Anagallis* s.str., as well as a number of *Lysimachia* species, which are often autogamous, have flowers without particular attractive agents. Vogel (1986) hypothesized these to represent reductions from the other flower types, which is corroborated by our molecular phylogeny where species with such flowers occur in several clades (Fig. 1). The yellow flowered *L. japonica* and *L. thyrsoflora* have evolved independently from ancestors with oil-producing yellow flowers, whereas other species without attractants such as *L. africana*, *L. minoricensis* and *L. ruhmeriana* have evolved from ancestors with nectar-producing white flowers within subg. *Palladia*. The two species, *L. nemorum* and *L. serpyllifolia*, also lacking oil- and nectar-producing trichomes, belong to the same clade as *Asterolinon*, *Pelletiera* and *Anagallis arvensis* and *A. monelli* of *Anagallis* s.str., in which this kind of flowers seem to have evolved earlier, already in the ancestor of the group.

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References

- Anderberg, A. A. & Ståhl, B. 1995: Phylogenetic interrelationships in the order *Primulales*, with special emphasis on the family circumscriptions. – *Canad. J. Bot.* **73**: 1699-1730.
- , Rydin, C. & Källersjö, M. 2002: Phylogenetic relationships in the order *Ericales* s.l.: analyses of molecular data from five genes from the plastid and mitochondrial genomes. – *Amer. J. Bot.* **89**: 677-687.
- , Ståhl, B. & Källersjö, M. 2000: *Maesaceae*, a new primuloid family in the order *Ericales* s.l. – *Taxon* **49**: 183-187. [[CrossRef](#)]
- Bennell, A. P. & Hu, C.-M. 1983: The pollen morphology and taxonomy of *Lysimachia*. – *Notes Roy. Bot. Gard. Edinburgh* **40**: 425-458.
- Chen, F.-H. & Hu, C.-M. 1979: Taxonomic and phytogeographic studies on Chinese species of *Lysimachia*. – *Acta Phytotax. Sin.* **17**: 21-53.
- Cronquist, A. 1981: An integrated system of classification of flowering plants. – New York.
- Decrock, E. 1901: Anatomie des Primulacées. – *Ann. Sci. Nat., Bot.*, ser. 8, **13**: 1-199.
- Farris, J. S. 1972: Inferring phylogenetic trees from distance matrices. – *Amer. Naturalist* **106**: 645-668. [[CrossRef](#)]
- 1997: “Xac.” Computer program and manual. – Stockholm.
- , Albert, V. A., Källersjö, M., Lipscomb, D. & Kluge, A. G. 1996: Parsimony jackknifing outperforms neighbor-joining. – *Cladistics* **12**: 99-124. [[CrossRef](#)]
- Fedorov, A. A. 1981: *Primulaceae*. – Pp. 63-87 in: Fedorov, A. A. (ed.), *Flora partis Europaeae URSS* **5**. – Leningrad.
- Hall, T. A. 1999: BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. – *Nucleic Acids Symposium Ser.* **1999**: 41, 95.
- Handel-Mazzetti, H. 1928: A revision of the Chinese species of *Lysimachia*, with a new system of the whole genus. – *Notes Roy. Bot. Gard. Edinburgh* **16**: 51-122.
- Hao, G. & Hu, C.-M. 2001: Phylogenetic relationships in *Lysimachia* (*Primulaceae*): a cladistic analysis. – *J. Trop. Subtrop. Bot.* **9**: 93-100.
- , Yuan, Y.-M., Hu, C.-M., Ge, X.-J. & Zhao, N.-X. 2004: Molecular phylogeny of *Lysimachia* (*Myrsinaceae*) based on chloroplast *trnL-F* and nuclear ribosomal ITS sequences. – *Molec. Phylogenet. Evol.* **31**: 323-339. [[CrossRef](#)]
- Hu, C.-M. & Kelso, S. 1996: *Primulaceae*. – Pp. 39-189 in: Wu, Z.-Y. & Raven, P. H. (ed.), *Flora of China* **15**. – Beijing & St Louis.
- Huynh, K.-L. 1970: Le pollen et la systématique chez le genre *Lysimachia* (*Primulaceae*) I. Morphologie générale du pollen et palynotaxonomie. – *Candollea* **25**: 267-296.
- 1971: Le pollen et la systématique chez le genre *Lysimachia* (*Primulaceae*) II. Considérations générales. – *Candollea* **26**: 279-295.
- Judd, W. S., Sanders, R. W. & Donoghue, M. J. 1994: Angiosperm family pairs: preliminary phylogenetic analyses. – *Harvard Pap. Bot.* **5**: 1-51.
- Källersjö, M., Bergqvist, G. & Anderberg, A. A. 2000: Generic realignment in primuloid families of the *Ericales* s.l.: a phylogenetic analysis based on DNA sequences from three chloroplast genes and morphology. – *Amer. J. Bot.* **87**: 1325-1341. [[CrossRef](#)]

- Leblebici, E. 1978: *Lysimachia*. – Pp. 135-138 in: Davis, P. H. (ed.), Flora of Turkey and the East Aegean Islands **6**. – Edinburgh.
- Mabberley, D. J. 1997: The Plant-Book, ed. 2. – Cambridge.
- Manns, U. & Anderberg, A. A. 2005: Molecular phylogeny of *Anagallis* (*Myrsinaceae*) based on ITS, *trnL-F*, and *ndhF* sequence data. – *Int. J. Pl. Sci.* **166**: 1019-1028. [[CrossRef](#)]
- Marr, K. L. & Bohm, B. A. 1997: A taxonomic revision of the endemic Hawaiian *Lysimachia* (*Primulaceae*) including three new species. – *Pacific Sci.* **51**: 254-287.
- Martins, L., Oberprieler, C. & Hellwig, F. 2003: A phylogenetic analysis of *Primulaceae* s.l. based on internal transcribed spacer (ITS) DNA sequence data. – *Pl. Syst. Evol.* **237**: 75-85. doi:10.1007/s00606-002-0258-1
- Mast, A. R., Kelso, S., Richards, J., Lang, D. J., Feller, D. M. S. & Conti, E. 2001: Phylogenetic relationships in *Primula* L. and related genera (*Primulaceae*) based on noncoding chloroplast DNA. – *Int. J. Pl. Sci.* **162**: 1381-1400. [[CrossRef](#)]
- Pax, F. 1889: *Myrsinaceae*. – Pp. 84-97 in: Engler, A. & Prantl, K. (ed.), Die natürlichen Pflanzenfamilien **4(1)**. – Leipzig.
- & Knuth, R. 1905: *Primulaceae*. – In: Engler, A. (ed.), Das Pflanzenreich **237**. – Leipzig.
- Ray, J. D. 1956: The genus *Lysimachia* in the new world. – *Illinois Biol. Monogr.* **24(3-4)**.
- Staden, R., Beal, K. & Bonfield, J. K. 1998: The Staden package. – Pp. 115-130 in: Misener, S. & Krawets, S. A. (ed.), Bioinformatics methods and protocols. – *Computer Meth. Molec. Biol.* **132**.
- Takhtajan, A. L. 1997: Diversity and classification of flowering plants. – New York.
- Taylor, P. 1955: The genus *Anagallis* in Tropical and South Africa. – *Kew Bull.* **3**: 321-350.
- Valdés, B. 1980: A new species of *Pelletiera* (*Primulaceae*) from Macaronesia. – *Candollea* **35**: 641-648.
- Vogel, S. 1986: Ölblumen und ölsammelnde Bienen 2. *Lysimachia* und *Macropis*. – *Trop. Subtrop. Pflanzenwelt* **54**.
- Wettstein, R. 1935: Handbuch der Systematischen Botanik, ed. 4. – Leipzig & Wien.

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Appendix

Table 1. Voucher material of species used for the analysis of *ndhF* sequences (AY839951-AY840013). Some earlier used voucher material (marked with *) is cited in Källersjö & al. (2000). (2) = two sequences used from the same voucher.

Taxon	Voucher /literature reference	GenBank No.
<i>Aegiceras corniculatum</i> (L.) Blanco	Källersjö & al. (2000)	AF213734*
<i>Anagallis acuminata</i> Schinz	Tanzania: <i>Bidgood & al. 4122</i> (K)	AY856417
<i>Anagallis alternifolia</i> Cav.	Chile, Atacama: <i>Frödin 434</i> (UPS)	AY839972
<i>Anagallis alternifolia</i> Cav.	Chile: <i>Gardner & Knees 4720</i> (K)	AY856418
<i>Anagallis angustiloba</i> (Engl.) Engl.	Rwanda: <i>Plumtree s.n.</i> (K)	AY856419
<i>Anagallis arvensis</i> L.	Källersjö & al. (2000)	AF213735*
<i>Anagallis arvensis</i> L.	Greece: <i>Snogerup & Snogerup 16875a</i> (UPS)	AY856420
<i>Anagallis crassifolia</i> Thore (2)	Spain: <i>Sandwith 6250</i> (K)	AY856421
<i>Anagallis djaloni</i> A. Chev.	Ethiopia: <i>Gilbert & al. 571</i> (UPS)	AY856422
<i>Anagallis filiformis</i> Cham. & Schltldl.	Brazil: <i>Rambo s.n.</i> (S)	AY856423
<i>Anagallis hexamera</i> P. Taylor	Ethiopia: <i>Johansson s.n.</i> (S)	AY856424
<i>Anagallis huttoni</i> Harv.	Lesotho: <i>Hedberg & Hedberg 82005</i> (UPS)	AY856425
<i>Anagallis kingaënsis</i> Engl.	Tanzania: <i>Thulin & Mhoro 1238</i> (UPS)	AY856426
<i>Anagallis kochii</i> H. E. Hess	Zambia: <i>Fitz Gerald 4068</i> (K)	AY856427
<i>Anagallis minima</i> (L.) E. H. L. Krause (2)	Sweden, Halland: <i>Anderberg s.n.</i> (S)	AY839968
<i>Anagallis monelli</i> L.	Portugal: <i>Anderberg 7278</i> (S)	AY839967
<i>Anagallis nummularifolia</i> Baker	Madagascar: <i>Viguier & Humbert 1536</i> (B)	AY856429
<i>Anagallis oligantha</i> P. Taylor	Malawi: <i>Richards 16641</i> (K)	AY856430
<i>Anagallis pumila</i> Sw.	Kenya: <i>Faden & Ng'weno 87/29</i> (UPS)	AY839969
<i>Anagallis pumila</i> Sw.	Ecuador: <i>Øllgaard & Navarrete 105421</i> (AAU)	AY856431
<i>Anagallis serpens</i> A. DC.	Tanzania: <i>Hedenäs & Bisang s.n.</i> (S)	AY839971
<i>Anagallis tenella</i> L.	France: <i>Lundqvist 20869</i> (S)	AY856434
<i>Anagallis tenuicaulis</i> Baker	Tanzania: <i>Bidgood & al. 2492</i> (UPS)	AY839970
<i>Anagallis tenuicaulis</i> Baker	Madagascar: <i>Nilsson & al. D57</i> (UPS)	AY856435
<i>Androsace</i> sp.	Källersjö & al. (2000)	AF213736*
<i>Ardisia crenata</i> Sims	Källersjö & al. (2000)	AY839951
<i>Ardisiandra wettsteinii</i> J. Wagner	Källersjö & al. (2000)	AF213733*
<i>Asterolinon adoëense</i> Kunze	Eritrea: <i>Ryding & al. 1091</i> (UPS)	AY839965
<i>Asterolinon linum-stellatum</i> (L.) Duby	Sweden: cult., <i>Anderberg s.n.</i> (S)	AY839964
<i>Clavija euerganea</i> J. F. Macbr.	Källersjö & al. (2000)	AF213737*
<i>Coris monspeliensis</i> L.	Källersjö & al. (2000)	AF213738*
<i>Cyclamen hederifolium</i> Aiton	Källersjö & al. (2000)	AF213740*
<i>Glaux maritima</i> L.	Källersjö & al. (2000)	AF213743*
<i>Grammadenia</i> sp.	Källersjö & al. (2000)	AF213744*
<i>Jaquinia keyensis</i> Mez	Källersjö & al. (2000)	AF213746*
<i>Lysimachia africana</i> Engl.	Ethiopia: <i>Gilbert & al. 488</i> (UPS)	AY839979
<i>Lysimachia alfredii</i> Hance	China: <i>Hao 394</i> (IBSC)	AY839984
<i>Lysimachia alpestris</i> Benth.	Hong Kong: <i>Law & Kendrick 00/13</i> (S)	AY839997
<i>Lysimachia andina</i> Sandwith	Ecuador: <i>Ståhl 4442</i> (GB)	AY839991
<i>Lysimachia ardisioides</i> Masam.	Taiwan: <i>Chung & Anderberg 1353</i> (S)	AY840000
<i>Lysimachia atropurpurea</i> L.	England: cult., <i>Kew Gardens 1993</i>	AY839954
<i>Lysimachia candida</i> Lindl.	China, Hubei: <i>Hao 204</i> (IBSC)	AY839973
<i>Lysimachia capillipes</i> Hemsl.	China, Chongqing: <i>Hao 409</i> (IBSC)	AY839999
<i>Lysimachia christinae</i> Hance	China, Hubei: <i>Hao 203</i> (IBSC)	AY840008
<i>Lysimachia ciliata</i> L.	USA, Vermont: <i>Dragon 02-68</i> (S)	AY839977
<i>Lysimachia clethroides</i> Duby	Sweden: cult., <i>Anderberg s.n.</i> (S)	AY839955
<i>Lysimachia confertifolia</i> C. Y. Wu	China, Yunnan: <i>Hao 254</i> (IBSC)	AY840012
<i>Lysimachia congestiflora</i> Hemsl.	Taiwan: <i>Peng & al. 17435</i> (S)	AY839963

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<i>Lysimachia ephemerum</i> L.	Sweden: cult., <i>Anderberg s.n.</i> (S)	AY839976
<i>Lysimachia fistulosa</i> Hand.-Mazz.	China, Chongqing: <i>Hao 399</i> (IBSC)	AY839978
<i>Lysimachia foenum-graecum</i> Hance	China, Yunnan: <i>Hao 250</i> (IBSC)	AY840007
<i>Lysimachia fordiana</i> Oliv.	China, Guangdong: <i>Ye & al. 3940</i> (IBSC)	AY840009
<i>Lysimachia grammica</i> Hance	China, Hubei: <i>Hao 209</i> (IBSC)	AY840010
<i>Lysimachia hillebrandii</i> A. Gray	Hawaii, Lyon Arboretum: <i>Acc. No. P1626</i>	AY839982
<i>Lysimachia hybrida</i> Michx.	USA, Vermont: <i>Dragon 0801-70</i> (S)	AY839981
<i>Lysimachia insignis</i> Hemsl.	China, Yunnan: <i>Bo Xiao s.n.</i> (S)	AY840001
<i>Lysimachia</i> cf. <i>japonica</i> Thunb.	Taiwan: <i>Chung & Anderberg 1354</i> (HAST)	AY839985
<i>Lysimachia japonica</i> Thunb.	Taiwan: <i>Peng 18731</i> (S)	AY839986
<i>Lysimachia lancifolia</i> Craib	Thailand, Nan: <i>Larsen & al. 43680</i> (AAU)	AY840006
<i>Lysimachia laxa</i> Baudo	Thailand: <i>Larsen & al. 44962</i> (AAU)	AY840005
<i>Lysimachia leschenaultii</i> Duby	India: <i>Klackenberg & Lundin 487</i> (S)	AY839957
<i>Lysimachia lobelioides</i> Wall. (2)	China, Yunnan: <i>Hao 303</i> (IBSC)	AY839958
<i>Lysimachia mauritiana</i> Lam.	Taiwan: <i>Gudmundsson s.n.</i> (S)	AY839956
<i>Lysimachia maxima</i> (R. Knuth) H. St.John	Källersjö & al. (2000)	AF213748*
<i>Lysimachia melampyroides</i> R. Knuth	China, Hunan: <i>Luo 0117</i> (HAST)	AY839992
<i>Lysimachia minoricensis</i> J. J. Rodr.	Källersjö & al. (2000)	AF213749*
<i>Lysimachia microcarpa</i> C. Y. Wu	China, Yunnan: <i>Han 5</i> (IBSC)	AY840013
<i>Lysimachia nemorum</i> L.	Källersjö & al. (2000)	AF213747*
<i>Lysimachia nigropunctata</i> Masam.	Taiwan: <i>Peng & al. 17283</i> (S)	AY839961
<i>Lysimachia nummularia</i> L.	Sweden: cult., <i>Anderberg s.n.</i> (S)	AY839988
<i>Lysimachis nutantiflora</i> F. H. Chen & C. M. Hu	China, Guangxi: <i>Hao 239</i> (IBSC)	AY840011
<i>Lysimachia paridiformis</i> Franch.	China, Hunan: <i>Luo 0143</i> (HAST)	AY839995
<i>Lysimachia peduncularis</i> Wall.	Thailand: <i>Larsen & al. 44904</i> (AAU)	AY840004
<i>Lysimachia petelotii</i> Merr.	China, Yunnan: <i>Bo Xiao s.n.</i> (S)	AY839998
<i>Lysimachia phyllocephala</i> Hand.-Mazz.	China, Chongqing: <i>Hao 405</i> (IBSC)	AY839983
<i>Lysimachia pittosporoides</i> C. Y. Wu	China, Yunnan: <i>Bo Xiao s.n.</i> (S)	AY840002
<i>Lysimachia pterantha</i> Hemsl.	China, Sichuan: <i>Chen & al. 960472</i> (HAST)	AY839996
<i>Lysimachia punctata</i> L.	Sweden: cult., <i>Anderberg s.n.</i> (S)	AY839987
<i>Lysimachia quadriflora</i> Sims	USA, Michigan: <i>Stevens 1625</i> (UPS)	AY839975
<i>Lysimachia ramosa</i> Wall.	Ceylon: <i>Klackenberg 156</i> (S)	AY839974
<i>Lysimachia remota</i> Petitm.	Taiwan: <i>Peng & al. 17246</i> (S)	AY839962
<i>Lysimachia rubiginosa</i> Hemsl.	China, Sichuan: <i>Hao 619</i> (IBSC)	AY839989
<i>Lysimachia ruhmeriana</i> Vatke	Ethiopia, Bale: <i>Friis & al. 3460</i> (UPS)	AY839980
<i>Lysimachia serpyllifolia</i> Schreb.	Crete: <i>Kretaexpeditionen 1989 s.n.</i> (S)	AY839993
<i>Lysimachia trestrestris</i> Britton & al.	Canada, Quebec: <i>Blondeau S2002-383</i> (S)	AY839994
<i>Lysimachia thyrsiflora</i> L.	Sweden: <i>Anderberg s.n.</i> (S)	AY839959
<i>Lysimachia vittiformis</i> F. H. Chen & C. M. Hu	China, Yunnan, <i>Hao 237</i> (IBSC)	AY840003
<i>Lysimachia vulgaris</i> L.	Sweden, Uppland: <i>Anderberg s.n.</i> (S)	AY839960
<i>Maesa tenera</i> Mez	Källersjö & al. (2000)	AF213750*
<i>Oncostemum</i> sp.	Madagascar: <i>Villiers & al. 4937</i> (S)	AY839953
<i>Pelletiera verna</i> A. St.-Hil.	Argentina: <i>Urtubey & Trementsberger 119</i> (S)	AY839990
<i>Pelletiera wildpretii</i> Valdés	Madeira, Salvagem Grande: <i>Paulo s.n.</i> (S)	AY839966
<i>Primula palinuri</i> Petagna	Källersjö & al. (2000)	AF213756*
<i>Samolus repens</i> Pers.	Källersjö & al. (2000)	AF213760*
<i>Stylogyne gentryi</i> Lundell	Ecuador: <i>Ståhl & Knudsen 1363</i> (S)	AY839952
<i>Trientalis borealis</i> Raf.	USA: <i>Breckling s.n.</i> (S)	AY856438