

## Paraphyly of Paederieae, recognition of Putorieae and expansion of *Plocama* (Rubiaceae-Rubioideae)

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Phylogenetic analyses of taxa that have been included in the tribe Paederieae are performed on the basis of a molecular dataset of plastid DNA sequences (the *rbcL* gene, *rps16* intron, and the regions *trnT-F*). The paraphyly of Paederieae as currently circumscribed is confirmed. A clade comprising *Paederia*, *Spermadictyon*, *Leptodermis*, and *Serissa* is retrieved and the circumscription of Paederieae is proposed to be restricted to this clade. *Paederia* is sister to a clade with *Spermadictyon*, *Leptodermis*, and *Serissa*. *Paederia* is subdivided into an Asian and an African clade, and within the African clade seven species endemic to Madagascar are sister to *P. pospischilii* in north-eastern Africa. The position of *Kelloggia* as sister to Rubieae is confirmed and a position of *Saprosma* in the Spermaceae alliance is supported. The genera *Putoria*, *Gaillonia* (with segregates), *Plocama*, and *Aitchisonia* form a clade that is sister to Theligoneae/*Kelloggia*/Rubieae. The name Putorieae is resurrected for the members of this clade. A synopsis of Putorieae is provided, where all species are placed in an expanded *Plocama* with 34 species. Thirty-one new combinations in *Plocama* are proposed. *Plocama pendula* on the Canary Islands is strongly supported as sister to a clade with *P. crocyllis* (former *Crocyllis*) in southern Africa and *P. yemenensis* and *P. tinctoria* (former *Gaillonia*) in southern Arabia/Horn of Africa.

**KEYWORDS:** *Gaillonia*, molecular phylogenetics, Paederieae, *Plocama*, Putorieae, *rbcL*, *rps16*, Rubiaceae, *trnT-F*

### INTRODUCTION

The tribe Paederieae has been recognized since 1830 (de Candolle), and according to recent circumscriptions (Robbrecht, 1988; Bremer & Manen, 2000) it comprises about 15 genera (see Table 1) and close to 100 species. It is distributed in the northern hemisphere, with the exception of the South African *Crocyllis anthospermoides* (≡ *Gaillonia crocyllis*), and some species of *Paederia* in Africa/Madagascar and South America. The Paederieae are shrubs, dwarf shrubs, climbers or perennial herbs with raphides, and divided or entire, interpetiolar stipules. The flowers have valvate corolla lobes, the stamens are inserted at the throat of the corolla tube, and the style is as long as, or longer than, the two to five stigma lobes. The ovary is two- to five-locular, each locule containing a single, erect ovule. The fruits are very variable and belong to one of following three types: fleshy and indehiscent, more or less dry and separating into two indehiscent mericarps, or dry and splitting into valves. The exotestal cells are parenchyma-like (e.g., Puff, 1982; Robbrecht, 1988). An unpleasant smell is evident in many members of Paeder-

ieae, and the chemistry of the group has been the subject of several studies (Hegnauer, 1973; Takeda & al., 1991). The pollen morphology in Paederieae is fairly uniform with suboblate to subprolate, three- (rarely four-)colpate pollen, featuring colpi with irregular margins, supratrigillar elements, and verrucose colpus membranes (Robbrecht, 1982; Igersheim, 1991; Dessein & al., 2005).

Jussieu (1789) described the family Rubiaceae and divided it into ten different groups, mainly based on fruit characters. Most of the taxa included in Paederieae by Robbrecht (1988) were included in his group number VII, along with taxa today placed in various other tribes, such as Psychotrieae, Anthospermeae, Ixoreae, Chiococceae, and Vanguerieae. When Paederieae was proposed by de Candolle (1830), he included three genera, *Lygodysodea*, *Lecotea*, and *Paederia*. He placed *Plocama*, *Putoria*, *Serissa*, as well as some genera today not associated with Paederieae, such as *Cuncea*, *Ernodea*, *Hydrophylax*, and *Scyphiphora* in Putoriinae (as “Putorieae”), as a subtribe of his tribe Spermaceae. Sweet (1839) recognized Paederieae with *Paederia* and *Lecotea*, and also gave tribal rank to Putorieae, to which he referred *Plocama*, *Putoria*,

Table 1. Overview of classifications of genera referred to Paederieae s.l.

Genera	De Candolle 1830	Hooker 1873	Schumann 1891	Puff 1982	Robbrecht 1988	Robbrecht 1993	Bremer & Manen 2000	Present study
<i>Aitchisonia</i>	—	—	Pae	Pae	Pae	Pae	—	Put
<i>Choulettia</i>	—	—	—	Pae	Pae	Pae	Pae	Put
<i>Crocyllis</i>	—	Ant	Ant	Pae	Pae	Pae	Pae	Put
<i>Gaillonia</i>	SSpe	Spe	Spe	Pae	Pae	Pae	Pae	Put
<i>Jaubertia</i>	—	—	—	Pae	Pae	Pae	Pae	Put
<i>Kelloggia</i>	—	Ant	Ant	Pae	Pae	Pae	Pae	—
<i>Leptodermis</i>	Gue	Pae	Pae	Pae	Pae	Pae	Pae	Pae
<i>Mitchella</i>	Gue	Ant	Ant	Pae	<i>Inc. sed.</i>	Mit	Mor-MI	Mor
<i>Paederia</i>	Pae	Pae	Pae	Pae	Pae	Pae	Pae	Pae
<i>Plocama</i>	SPut	Ant	Ant	Pae	Pae	Pae	Pae	Put
<i>Pseudogaillonia</i>	—	—	—	Pae	Pae	Pae	Pae	Put
<i>Pseudopyxis</i>	—	Pae	Pae	Pae	Pae	Pae	Pae	Pae
<i>Pterogaillonia</i>	—	—	—	Pae	Pae	Pae	Pae	Put
<i>Putoria</i>	SPut	Ant	Ant	Pae	Pae	Pae	Pae	Put
<i>Saprosma</i>	Cof	Psy	Psy	—	Psy	Pae	—	—
<i>Serissa</i>	SPut	Ant	Ant	Pae	Pae	Pae	Pae	Pae
<i>Spermadictyon</i>	Gue	Pae	Pae	Pae	Pae	Pae	Pae	Pae

Abbreviations: Ant, Anthospermeae; Cof, Coffeeae; Gue, Guettardeae; Mit or Mor-MI, *Mitchella*-group; Mor, Morindeae; Pae, Paederieae; Psy, Psychotriaceae; Put, Putorieae; Spe, Spermaceae; SPut, subtribe Putorineae of Spermaceae; SSpe, subtribe Spermacocinae of Spermaceae.

*Serissa*, and *Ernodea*. However, Putorieae has not been recognized by any author for well over a hundred years.

Schumann's (1891) circumscription of Paederieae long prevailed in the 20th century (e.g., Bremekamp, 1954, 1966; Verdcourt, 1958), but Puff (1982), followed by Robbrecht (1988, 1993) proposed a much broader circumscription of the tribe (Table 1) on the basis of various morphological characters. Lately some taxa of Paederieae have been included in molecular, phylogenetic studies of Rubiaceae (Andersson & Rova, 1999; Bremer & Manen, 2000). These studies indicated that the tribe is paraphyletic, but the sampling has been too restricted to allow any firm conclusions.

Genera that have been included in Paederieae are *Aitchisonia*, *Choulettia*, *Crocyllis*, *Gaillonia*, *Hondbessen*, *Jaubertia*, *Kelloggia*, *Lecontea*, *Leptodermis*, *Lygodisodea*, *Mitchella*, *Paederia*, *Plocama*, *Pseudogaillonia*, *Pseudopyxis*, *Pterogaillonia*, *Putoria*, *Saprosma*, *Serissa*, and *Spermadictyon*. Of these *Hondbessen*, *Lecontea*, and *Lygodisodea* are synonyms of *Paederia* (Puff, 1991), *Paederia* being conserved against *Hondbessen*.

*Choulettia*, *Crocyllis*, *Gaillonia*, *Jaubertia*, *Pseudogaillonia*, and *Pterogaillonia* belong to the "Gaillonia complex" (Linczevski, 1973; Qarar, 1973; Léonard, 1984), and have all at some point been included in *Gaillonia*. A historical outline of the group is given by Thulin (1998), who also pointed to the close relationship between *Gaillonia* in a wide sense, *Plocama* and *Putoria*.

*Kelloggia* and *Mitchella* were included in Paederieae by Puff (1982), but recent studies indicate that none of these genera is closely related to this tribe, *Kelloggia* being sister to Rubieae (Nie & al., 2005), and *Mitchella*

to *Damnacanthus* (Bremer & Manen, 2000) in the *Psychotria* alliance sensu Bremer & Manen (2000).

*Saprosma* has been placed in Psychotriaceae by several authors (e.g., Schumann, 1891; Robbrecht, 1988), but was moved to Paederieae by Robbrecht (1993), who pointed to various morphological similarities with this tribe. The position of the genera placed in Paederieae by Robbrecht (1988, 1993), compared to their position in previous classifications is given in Table 1.

The aims of the present study are to use molecular analyses of a wide and carefully selected sample of taxa to clarify further the phylogenetic relationships within Paederieae in a wide sense, and to propose a new classification for this part of Rubiaceae based on monophyletic entities.

## MATERIALS AND METHODS

**Taxon sampling.** — The intention has been to include taxa from all genera that at some point have been placed in Paederieae. This was achieved, with the sole exception of the monotypic Japanese genus *Pseudopyxis*, from which we had access to herbarium material but failed to amplify any of the tested regions. A total of 29 species of Paederieae s.l. were included in the analyses. The sampling of the larger genera *Gaillonia* s.l. and *Paederia* comprises taxa from all parts of their areas of distribution, except that South American members of *Paederia* are missing. Indications that Rubieae and Theligoneae are close to Paederieae (Bremer & Manen, 2000) led us to include members of these, as well as some representatives from

an additional set of tribes within Rubioideae. *Luculia* was selected as the most distant Rubiaceae outgroup. Available sequence data were retrieved from NCBI data base, GenBank. Voucher information and accession numbers are provided in the Appendix.

**Morphology.** — Investigation of morphological characters, particularly in the “*Gaillonia* complex”, was made on herbarium material from BM, FUMH, K, MPU, P, S and UPS.

**DNA extraction.** — Total DNA was extracted using the CTAB method as described by Oxelman & al. (1999), starting from fresh, silica-gel dried (Doyle & Doyle, 1987; Chase & Hills, 1991) or herbarium material. The leaf material was not ground manually, but with a Mini-Beadbeater (Techtum Lab AB). DNA was purified with the QiaQuick PCR purification kit (Qiagen™).

**cpDNA regions and primers.** — For this study, three different chloroplast regions were selected, the *rbcL* gene, the intron of *rps16*, and the *trnT-F* region. The *rbcL* gene encodes the large subunit of ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCO), and is frequently used for molecular plant studies (e.g., Ritland & Clegg, 1987; Kim & al., 1992; Bremer & al., 1995). Amplification of the ca. 1,400 base pairs of *rbcL* was done in two steps. The first step using the primer pair Z1 and 1020R, in few instances Z1 and 895R, and the second step using the primer pairs 427BS and 3' (Zurawski DNAX Research Institute; Olmstead & al., 1992; Olmstead & al., 1993; Bremer & al., 2002). The intron of *rps16* is a group II intron of 800–900 base pairs (e.g., Kelchner, 2002), and is easily amplified in one reaction, as demonstrated in several studies (e.g., Oxelman & al., 1997; Andersson & Rova, 1999; Downie & Katz-Downie, 1999; Wallander & Albert, 2000). Primers used for amplification as well as sequencing of the *rps16* intron were F and R2 (Oxelman & al., 1997). The *trnT-trnF* region consists of approximately 1,200–1,600 base pairs, and is composed of three main sections. Starting with the *trnT* gene, the *trnT-trnL* spacer is followed by the *trnL* gene, including the *trnL* intron, and is completed by the *trnL-trnF* spacer (Taberlet & al., 1991). The *trnT-trnF* region was amplified in two steps, the first step using the primer pair A1 and D (or A and D), and the second step using the primer pair C and F (Taberlet & al., 1991; Bremer & al., 2002).

**Amplification.** — PCR reactions were performed in 50 µl volumes and mixed as follows: 29.25 µl ddH<sub>2</sub>O, 5 µl 10× buffer, 5 µl MgCl<sub>2</sub>, 5 µl TMACL (Chevet & al., 1995), 4 µl 10 mM dNTP, 0.5 µl 20 µM forward primer, 0.5 µl 20 µM reverse primer, 0.5 µl BSA, 1% 0.25 µl Taq DNA Polymerase. To 49 µl of the mix 1 µl DNA template was added. The amplifying reactions were run on a Mastercycler gradient (Eppendorf) with 1 min. at 95°C followed by 33–35 cycles of 95°C 1 min.; 55°C–58°C 1 min. 30 sec.; 72°C 1 min. 30 sec., finishing with 72°C for 7 min.

PCR products were purified with MultiScreen PCR plates (Millipore Corporation) prior to sequencing.

**Sequencing.** — Primers used for sequencing of *rbcL* were z1 (Olmstead & al., 1993), 895R, 234, 895, 1020, and 1204R (Zurawski). Sequencing of *trnT-F* were made with the primers a1, i (Bremer & al., 2002), a, b, c, d, e, and f (Taberlet & al., 1991). The sequence reactions were performed with the DYEnamic ET terminator Cycle Sequencing Kit (Amersham Biosciences), on a GeneAmp® PCR System 9700 (Applied Biosystems), following the protocol of the manufacturer, and were run on a MegaBACE™ 1000 DNA Analysis System (Amersham Biosciences). A few sequences were retrieved by the commercial services of MacroGen Inc. ([www.macrogen.com](http://www.macrogen.com)). GenBank accession numbers are listed in the Appendix.

**Alignment and matrices.** — Sequences were aligned manually, as the large number of insertions and deletions of different sizes produced ambiguous results from automated multiple sequence alignments by ClustalW (Thompson & al., 1997). After alignment, potentially informative insertion and deletion events were identified and recoded, following principles by Simmons & Ochoterena (2000). Recoded indels were given uniform weights of one. Sections still difficult to align were excluded from the analyses. The analyses were run both with and without these deletions and codings, and the major topographic structures were retained.

**Analyses.** — Parsimony and bootstrap analyses were performed using PAUP 4.0b10 (Swofford, 2002) under emulation of MacOS 9.2.2 by MacOS 10.3.8 on a 1 GHz Macintosh PowerBook G4 computer with 512 Mb RAM allocated, using only informative characters. The three matrices were run separately, and all combined, making a total of four different analyses. The search method was heuristic, with 1,000 replicates of random stepwise additions of sequences. Branch swapping algorithm employed was tree-bisection-reconnection (TBR), with swapping on best trees only. Support values were calculated with bootstrap analyses, in 1,000 replicates, each with 10 random stepwise additions sequences, MULPARS option off, and TBR branch swapping.

## RESULTS

Tree statistics from the four different analyses are presented in Table 2. The analyses show only minor contradictions between the different matrices, all trees were largely congruent, and differences were mainly on the level of resolution in the trees from the single gene matrices. The combined analysis resulted in one single tree, which is shown with results from the bootstrap analysis in Fig. 1. Corresponding information for each of the separate analyses are given in Figs. 2–4 (electronic supplement).

Table 2. Statistics from the four analyses.

	Combined	<i>rbcL</i>	<i>rps16</i>	<i>trnT-F</i>
Nr. of MPT:	1	2,318	3	240
Tree lengths	2,976	970	849	1,535
Inf. aligned characters	862	221	246	464
Consistency Index (CI)	0.673	0.511	0.680	0.711
Retention index (RI)	0.783	0.661	0.797	0.803
Rescaled CI	0.527	0.338	0.542	0.571

Abbreviations: Inf., informative; MPT, most parsimonious tree.

Of the 40 internal nodes (Fig. 1) receiving bootstrap support above 50%, no less than 19 have a support of 100%, and 27 receive a support of 90% or more. The results show univocal paraphyly of Paederieae, the bulk of which is separated into two major clades corresponding to nodes C and K in Fig. 1. Paederieae s.str. (node C), comprising *Paederia*, *Spermadietyon*, *Leptodermis*, and *Serissa*, forms the sister group to a larger assembly (node F). This larger assembly includes the tribes Rubieae and Theligoneae (*Theligonum*), and the genus *Saprosma*, and a very well supported clade (node K) including *Putoria*, *Gaillonia* and segregates, *Plocama*, and *Aitchisonia*. This clade is referred to as Putorieae (see below).

## DISCUSSION

The present study demonstrates, with strong support, that Paederieae is paraphyletic as earlier indicated. Hence taxonomic changes are needed in order to obtain monophyletic entities. Results mainly from the combined analysis (Fig. 1) are discussed below, with the nodes A to Q in alphabetic order.

**Node A, Spermacoceae alliance.** — This strongly supported (100%) node corresponds to the Spermacoceae alliance as defined by Bremer & Manen (2000). Representatives for all seven tribes of this alliance were included in our analysis. In the study by Bremer & Manen (2000), all the seven tribes were monophyletic and successively nested albeit with low support, with exception of Theligoneae and Rubieae, and the paraphyletic Paederieae. The present study gives a similar topology, except that Anthospermeae, Argostemmataeae, and Spermacoceae together form the sister group of the rest of the alliance.

The placement of *Mitchella* outside the Spermacoceae alliance, previously shown in the analyses by Bremer & Manen (2000) and Nie & al. (2005), is confirmed in our study.

**Node B, Paederieae s.str. and sister group.** — This strongly (92%) supported node comprises practically all the taxa included in Paederieae by Robbrecht (1988), and the paraphyly of the tribe is clearly shown. In the study by Bremer & Manen (2000) this node is

also retrieved and represented by six genera, *Didymaea* and *Rubia* in Rubieae, *Theligonum* in Theligoneae, and *Serissa*, *Plocama*, and *Putoria* in the paraphyletic Paederieae. The relationship between these taxa in the present analysis is identical, but much more strongly supported, and the addition of 19 genera and 29 terminal taxa has, as intended, increased the resolution. Members of node B generally have ovaries with a single basal (or almost so) ovule in each locule. With the exception of Rubieae, they also all seem to have foetid substances, generally referred to as paederosidic acid (Kapadia & al., 1979; Suzuki & al., 1993) or paederoside. Hegnauer (1973) mentioned this as a compound of potential systematic interest, and it is also mentioned in later studies (e.g., Takeda & al., 1991). However, foetid substances have been shown to be present in other taxa of Rubioideae as well, e.g., in *Carpacoce*, *Coprosma*, and *Pentanisia* (Verdcourt, 1958; Puff, 1982).

**Node C, Paederieae s.str.** — The support for this node comprising *Paederia*, *Spermadietyon*, *Leptodermis* and *Serissa* is 100%, and we propose to accommodate these genera in a recircumscribed Paederieae. De Candolle (1830) grouped *Serissa* and *Plocama* together on the basis of their fleshy fruits, but this relationship has no support in our study (for further information on the fruits of *Plocama pendula*, see under node Q). Ecological aspects of fruit evolution have been discussed by, e.g., Bremer & Eriksson (1992) and Novotny & al. (2002), and it seems clear that development of fleshy fruits is often the result of ecological pressure. *Paederia*, *Spermadietyon* and *Leptodermis* all have dry fruits, in which the exocarp splits into valves to release the two to five one-seeded pyrenes with seeds enclosed in endocarp (Puff, 1982). This fruit type seems to be a synapomorphy for Paederieae in this new sense. As *Serissa* is nested within these genera, its fleshy indehiscent fruit has probably evolved from a dry fruit of this type. *Pseudopyxis*, the monotypic Japanese genus that could not be included in the analysis (see under Material and Methods), probably belongs to Paederieae s.str. as well. It differs from the other four genera by being herbaceous, but the fruits agree well with those of *Paederia*, *Spermadietyon* and *Leptodermis* (Puff, 1982).

**Node D.** — This node, supported by 100%, comprises the Asian shrubby genera, *Spermadietyon* (1 species), *Leptodermis* (30 species), and *Serissa* (2 species). The internal node grouping *Leptodermis* and *Serissa* is also supported by 100%.

**Node E, *Paederia*.** — This tropical genus of some 30 species of climbing shrubs is represented in the analysis by ten species that group together with a support of 100%. The *Paederia* clade is divided into two monophyletic groups, one Asian with *P. foetida* (type of genus) and *P. pilifera* (100%) and one African with eight species (95%). In the latter, seven species endemic to Madagascar form a mono-

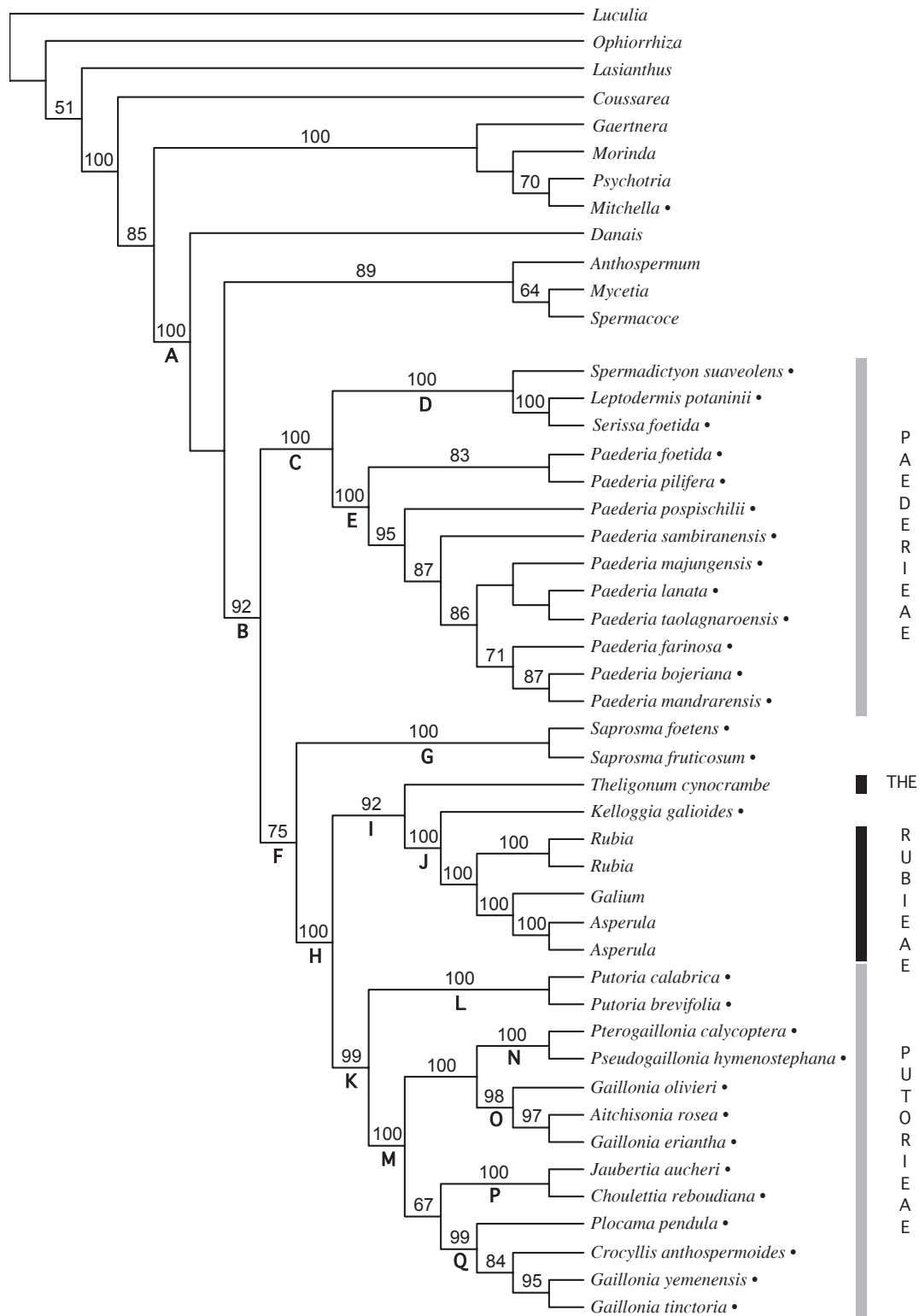


Fig. 1. The single most parsimonious tree from the combined analysis. Dots indicate taxa that at any point have been included in Paederieae. Numbers above the bars show the bootstrap values. Capital letters below the bars designate nodes that are discussed in the text. Grey bars indicate new circumscription according to this paper. THE = Theligoneae. The tribal positions of *Kelloggia* and *Saprosma* are left undecided. Taxa in the tribe Putorieae are labelled according to pre-revision classification. Taxa of the tribe Rubieae are composed of sequence data from several species, therefore only generic names are given.

phyletic group, sister to *P. pospischilii* in eastern Ethiopia, Somalia, and northern and eastern Kenya. For the African clade the name *Lecontea* is available, but we do not propose any taxonomic changes as *Paederia* s.l. apparently is a very coherent group (Puff, 1991), and also as no South American members are included in the study. For the South American species the name *Lygodisodea* is available.

**Node F, the sister group of Paederieae s.str.**

— This group comprises the representatives of the tribes Rubieae and Theligoneae, *Kelloggia*, the remaining part of Paederieae (node K, see below), and the genus *Saprosma*. This entire group yields a support of 75% in the combined analysis, the reason for this moderate support apparently being related to the placement of the genus *Saprosma*. However, *Saprosma* is in all analyses unambiguously and with strong support included in the Spermaceae alliance (node A) and obviously belongs to this complex. There is no molecular support for a close relationship of *Saprosma* with Psychotrieae as previously sometimes has been suggested (Schumann, 1891; Robbrecht, 1988).

**Node G, *Saprosma*.** — The placement of this Indo-Malaysian genus is unexpected. In the classification of Rubioideae by Bremer & Manen (2000) *Saprosma* is not mentioned, although it has been placed in Psychotrieae by several authors (e.g., Robbrecht, 1988). However, Robbrecht (1993) included it in Paederieae based on its valvate-induplicate aestivation (valvate in Paederieae), placentation, obturator, ovule structure, and presence of paederoside. To verify the primary results in our study a second species of *Saprosma* was sequenced and included in the analyses. Both sequences are similar, showing several insertion/deletion events, albeit not identical. The placement of *Saprosma* as sister group to node H receives moderate support, (< 50% from *rbcl*, *rps16*, and *trnT-F* [Figs. 2–4 in the electronic supplement], in congruence with previous unsuccessful attempts to place *Saprosma* using molecular markers, but 75% in the combined analysis). Probably *Saprosma* is best accommodated in a new monogeneric tribe in Rubioideae, but we refrain from making a formal proposal about this, pending further phylogenetic and morphological studies of the genus.

**Node H.** — This node, supported by 100%, comprises Rubieae, Theligoneae, *Kelloggia*, and the rest of Paederieae (node K). The relationship between these entities (except *Kelloggia*) was proposed already by Bremer & Manen (2000). However, these authors refrained from making any formal taxonomic changes with reference to the present study.

**Node I, Rubieae, Theligoneae and *Kelloggia*.**

— Since the inclusion of the herbaceous *Theligonum*, previously in Theligonaceae, in Rubiaceae (Wunderlich, 1971), it has generally been placed in a monogeneric tribe, Theligoneae. The genus is highly unusual in the family by having, for example, upper leaves with strong anisophylly

and male flowers with up to 30 stamens, but the close relationship between Theligoneae and Rubieae has been widely accepted since 1995 (Bremer & al., 1995; Natali & al., 1995). The bootstrap support for node I, where *Theligonum* is sister to Rubieae plus *Kelloggia*, is 92%. The same topology, but with lower support, was revealed in the study by Nie & al. (2005).

**Node J, *Kelloggia* and Rubieae.** — This is a strongly supported node (100%) placing the herbaceous *Kelloggia*, previously in Paederieae and with one species in North America and one in eastern Asia, as sister to Rubieae. Rubieae has been much discussed in recent years (e.g., Natali & al., 1996), and has been suggested to be monophyletic in several studies (Manen & al., 1994; Manen & Natali, 1995; Natali & al., 1995; Huysmans & al., 2003), as is also the case in the present study. The monophyly of *Kelloggia* was clearly demonstrated by Nie & al. (2005), who also, like us, found it to be sister to Rubieae with strong support. Nie & al. (2005) considered the inclusion of *Kelloggia* in Rubieae, but pointed to differences in pollen morphology and leaf arrangement (leaves opposite in *Kelloggia*, verticillate in Rubieae), and the absence of a specific 50-bp deletion in the *atpB-rbcL* sequences of *Kelloggia* as arguments against such an action. We agree with their conclusion that *Kelloggia* may be best placed in a tribe of its own, but, like them, refrain from any formal proposal of this at present. To avoid monogeneric tribes an alternative could be, for example, to expand the herbaceous Rubieae to comprise also *Kelloggia* and *Theligonum*. A proposal along this line was also recently made by Robbrecht & Manen (2006), who erected the monogeneric subtribes Theligoninae and Kelloggiinae to accommodate *Theligonum* and *Kelloggia* within Rubieae.

**Node K, Putorieae.** — This node, with a support of 99%, comprises the members of the former Paederieae with fruits splitting into two indehiscent one-seeded mericarps (*Aitchisonia*, *Choulettia*, *Crocyllis*, *Gaillonia*, *Jaubertia*, *Pseudogaillonia*, *Pterogaillonia* and *Putoria*), plus *Plocama* (with fleshy indehiscent fruit). For this group, which generally consists of shrubs or shrublets, the name Putorieae is available and we propose to resurrect it as a tribe with this new circumscription.

The generic subdivision of Putorieae needs reconsideration. Puff (1982) considered *Choulettia*, *Jaubertia*, *Pseudogaillonia* and *Pterogaillonia* to be not sufficiently different from *Gaillonia* (as *Neogaillonia*) to warrant their separation. He also pointed to similarities between *Gaillonia* and *Crocyllis*, *Plocama* and *Putoria*. Thulin (1998) treated *Crocyllis* as a synonym of *Gaillonia* and, again, pointed to the doubtful distinction between *Gaillonia*, *Plocama* and *Putoria*. Similarities between *Aitchisonia* and *Gaillonia* were pointed out already by Hemsley (1882). We have carefully but in vain searched for morphological support, in herbarium material and in literature, for the

internal nodes L–Q (see also below), and our conclusion is that all members of node K are best accommodated in a single genus, for which the oldest name is *Plocama*. Due to the lack of morphological support we also refrain from proposing any subgenera or sections. The formal nomenclatural changes are found below under “Synopsis of Putorieae”. However, in the analyses presented in this paper the old names have been used, to demonstrate the width of the sampling. There are several well supported nodes within *Plocama* s.l. and some of these (nodes L–Q in Fig. 1) are further discussed below.

**Node L, Mediterranean *Plocama*.** — This node comprises, with 100% support, the Mediterranean taxa previously placed in *Putoria*. The fruit in *Putoria* is fleshy and reddish when young, and has often been referred to as a drupe with two pyrenes (e.g., Thonner, 1908) or as a “bacca” (de Candolle, 1830; Lange, 1868; Halácsy, 1901), but as pointed out by Puff (1982) it is a schizocarp that finally splits into two indehiscent one-seeded mericarps, just as in *Gaillonia*, but with a fleshy outer wall that dries up and remains on the endocarp as a skin. The fleshy and reddish young fruits may be seen as a synapomorphy for this node, but also in *Gaillonia* the finally dry fruits may be, to various degrees, fleshy when young and *Plocama pendula* has true drupes. We have found no other characters that separate the members of this node from the rest of the Putorieae and therefore refrain from recognizing the genus *Putoria*.

**Node M, non-Mediterranean *Plocama*.** — This node comprises, with strong support (100%), the remaining part of Putorieae, with the taxa previously placed in *Aitchisonia*, *Choulettia*, *Crocylis*, *Gaillonia*, *Jaubertia*, *Pseudogaillonia*, *Pterogaillonia*, as well as *Plocama pendula*. The monotypic *Aitchisonia*, once suggested to be close to Dipsacales (Bremekamp, 1966), is now firmly placed in Putorieae. The suggestion of a dipsacalean affinity was due to the stamens being inserted at different levels, but this has also been observed in species of *Gaillonia* (Puff, 1982). The previously monotypic *Plocama* from the Canary Islands was regarded as closely related to *Putoria* already by de Candolle (1830). The genus *Gaillonia*, often segregated into several smaller genera, is paraphyletic in every sense that has been suggested. Although this node is strongly supported by the molecular data, we have not found any morphological support for it.

**Node N, former *Pterogaillonia* and *Pseudogaillonia*.** — The former genera *Pterogaillonia*, with two species, and *Pseudogaillonia* (monotypic), both mainly in south-western Asia, group together with high support (100%). They both have enlarged fruiting calyces, probably as an adaptation for wind dispersal (Ehrendorfer & Schönbeck-Temesy, 2005; Schönbeck-Temesy & Ehrendorfer, 2005).

**Node O, former *Gaillonia* in part and *Aitchisonia*.** — This node comprises, with 98% support, *Gaillonia*

*olivieri* (type of *Gaillonia*), *Aitchisonia rosea*, and *G. eriantha*, with *A. rosea* and *G. eriantha* as strongly supported sisters at an internal node. According to the classification by Ehrendorfer & Schönbeck-Temesy (2005), *G. olivieri* belongs to *G.* sect. *Gaillonia*, along with *G. bruguieri*, *G. iljinii*, *G. kandaharensis*, *G. szowitzii*, and *G. vassilczenkoi*, whereas *G. eriantha* belongs to *G.* sect. *Eriantha*, along with *G. afghanica*, *G. dubia* and *G. macrantha*. These species are all distributed in south-central and south-western Asia, a region to which are confined also *G.* sect. *Pseudoasperula* with the single species *G. asperuliformis*, *G.* sect. *Bucharica* with *G. bucharica*, *G. inopinata*, *G. mestscherjakovii*, and *G. trichophylla*, and *G.* sect. *Surchania* with *G. botschantzevii*. Although only three out of these 17 species have been sequenced they all have fruits splitting into two indehiscent, one-seeded mericarps with basal placentation and without hooked hairs, non-foliaceous stipules, and foetid substances (as far as known), a unique set of characters in Rubiaceae that is present in every member of node K (*Plocama*), sampled or not (the only exception is the fruit of *P. pendula*, see under node Q below). Furthermore, it is highly likely that the non-sampled species enumerated above all belong to node O along with the sampled species, although we cannot point to any particular character supporting this. However, all these species together form a geographically and morphologically coherent group and our sample includes the most aberrant species of them all, *Aitchisonia rosea*. In *Aitchisonia*, as in the members of node N, probable adaptations for wind dispersal are found (Schönbeck-Temesy & Ehrendorfer, 2005), but in this case the bracts enlarge, become membranous, and surround the fruits.

**Node P, former *Jaubertia* and *Choulettia*.** — This node comprises, with 100% support, the previous monotypic genera *Jaubertia* (in Oman, United Arab Emirates, Iran, Afghanistan and Pakistan) and *Choulettia* (in Morocco and Algeria). They both have bristle-like hispid bracts forming conspicuous involucre that remain attached to the fruits and evidently support their dispersal by wind. Due to this and other similarities *Choulettia* was recently made a synonym of *Jaubertia* by Ehrendorfer & Schönbeck-Temesy (2005).

**Node Q, *Plocama pendula*, former *Crocylis*, remaining former *Gaillonia*.** — This strongly (99%) supported node shows *Plocama pendula* on the Canary Islands as sister to a clade with the southern African *Crocylis anthospermoides* and, on a strongly supported internal node, *Gaillonia yemenensis* (Yemen and Oman) and *G. tinctoria* (Socotra and Somalia). *G. tinctoria* was placed in *G.* sect. *Anisostephus* by Ehrendorfer & Schönbeck-Temesy (2005), along with *G. puberula*, *G. putorioides* and *G. thymoides* from Socotra. Other close relatives are *G. calcicola* and *G. somaliensis* in Somalia and *G. jolana* in Yemen. These six non-sampled species form a geo-

graphically and morphologically coherent group along with *G. yemenensis* and *G. tinctoria*, and it is highly likely that they all belong to node Q, although we cannot point to any single character that they have in common. In any case, with their fruits splitting into two indehiscent mericarps, with basal placentation and without hooked hairs, non-foliaceous stipules, and foetid substances, they are all clearly members of node K (*Plocama*).

*Plocama pendula* is unique in Putorieae in having fleshy fruits that do not split into mericarps, but are drupes with two or rarely three one-seeded pyrenes. They are first whitish, but become black when ripe (Bramwell & Bramwell, 1974). The smelling fruits are staple food for the lizard *Gallotia galloti*, also endemic on the Canary Islands (Barquin Diez & Wildpret, 1975; Mendoza-Heuer, 1987). According to Barquin Diez & Wildpret (1975) ten studied excrements from this lizard contained 495 pyrenes of *Plocama pendula*. Observations of *Gallotia stehlini*, a lizard endemic on Gran Canaria, eating fruits from the hanging branches of *Plocama*, have been reported by Molina Borja (1986). It thus seems likely that the fleshy fruit of *Plocama* is an adaptation to saurochory.

Mendoza-Heuer (1987) also reported functional dioecy to be the normal condition in *Plocama pendula*, although occasional plants were found to be functionally monoecious. Flowers in Putorieae are otherwise, as far as known, hermaphrodite, with the exception of *Crocylis anthospermoides* that is gynodioecious (Puff, 1982; Puff & Mantell, 1982).

**Biogeography of *Plocama* s.l.** — In a review of molecular phylogenetic studies of taxa considered to represent biogeographical links between Macaronesia and southern Arabia and/or distant regions of Africa, Andrus & al. (2004) concluded that these putative relationships are, with few exceptions, not supported. One of their examples is *Plocama pendula*, where the postulation by Thulin (1998) of a close relationship between this species and species of *Gaillonia* in southern Arabia and north-eastern and southern Africa is cited. The study by Andersson & Rova (1999), indicating a sister relationship between *Plocama pendula* and *Theligonum*, is cited as counter-evidence. However, this result of Andersson & Rova is clearly an artifact as their sampling did not include any member of the “*Gaillonia* complex”. Instead, node Q, showing *Plocama pendula* to be sister to a clade with *Crocylis anthospermoides* in southern Africa and two species of *Gaillonia* in southern Arabia/Horn of Africa clearly indicates that this biogeographical link is valid. A similar result was obtained by Kornhall & al. (2001) in *Camptoloma*, but in this case *C. rotundifolium* in southern Africa (Angola and Namibia) was moderately (75%) supported as sister to a clade with *C. canariense* on the Canary Islands (Gran Canaria) and *C. lyperiiflorum* in southern Arabia/Socotra/northern Somalia.

## SYNOPSIS OF PUTORIEAE

**Putorieae** Sweet, Hort. Brit., ed. 3: 325. 1839 ≡ Putorinae DC., Prodr. 4: 470. 1830 as “Putorieae” – Type: *Putoria* Pers.

- Plocama*** Aiton, Hort. Kew. 1: 292, 3: 508. 1789 – Type: *Plocama pendula* Aiton  
 = *Putoria* Pers., Syn. Pl. 1: 524. 1805 – Type: *Putoria calabrica* (L. f.) DC.  
 = *Gaillonia* A. Rich. ex DC., Prodr. 4: 574. 1830 ≡ *Neogaillonia* Lincz. in Novosti Sist. Vyssh. Rast. 10: 226. 1973, nom. superfl. – Type: *G. oliveri* A. Rich. ex DC., lectotype selected by Linczevski, loc. cit.  
 = *Jaubertia* Guill. in Ann. Sci. Nat., Bot., sér. 2, 16: 60. 1841 – Type: *J. aucheri* Guill.  
 = *Crocylis* E. Mey. ex Hook. f. in Benth. & Hook. f., Gen. Pl. 2: 26, 136. 1873 – Type: *C. anthospermoides* E. Mey. ex K. Schum.  
 = *Choulettia* Pomel, Nouv. Mat. Fl. Atl.: 81. 1874 – Type: *C. reboudiana* (Coss. & Durieu) Pomel  
 = *Aitchisonia* Hemsl. in J. Linn. Soc., Bot. 19: 166. 1882 – Type: *A. rosea* Hemsl.  
 = *Pterogaillonia* Lincz. in Novosti Sist. Vyssh. Rast. 10: 233. 1973 – Type: *P. calycoptera* (Decne.) Lincz.  
 = *Pseudogaillonia* Lincz. in Novosti Sist. Vyssh. Rast. 10: 235. 1973 – Type: *P. hymenostephana* (Jaub. & Spach) Lincz.

Shrubs or herbs with a foetid smell when crushed. Leaves opposite but sometimes crowded and appearing whorled; stipules mostly sheathing at the base. Flowers hermaphrodite or sometimes functionally unisexual, generally 4–5-merous, in cymose inflorescences or solitary; bracts mostly inconspicuous or obsolete or sometimes enlarged and forming an involucre. Calyx-lobes often unequal, sometimes obsolete, or sometimes variously enlarged after anthesis. Corolla funnel-shaped with a short to long tube; lobes valvate in bud. Stamens inserted at throat of corolla, sometimes unequal; anthers included to exerted. Ovary 2(–3)-celled, each cell with a single ovule attached near the base; style filiform, with 2(–3) linear lobes. Fruit a drupe or mostly splitting into two 1-seeded indehiscent mericarps when ripe, glabrous or hairy but without hooked hairs, crowned by persistent calyx-lobes.

Thirty-four species, the majority in south-western Asia, a few in Africa, one on the Canary Islands, and one widespread Mediterranean species also in southern Europe. The circumscription of the species follows the recent treatments by Thulin (1998), Ehrendorfer & Schönbeck-Temesy (2005), and Schönbeck-Temesy & Ehrendorfer (2005).

Chromosome numbers seem to have been counted in two members of the genus only, *P. calabrica* ( $2n = 22$ ,



see Backlund & Thulin, 2007) and *P. pendula* ( $2n = 44$ , see Larsen, 1958).

*Plocama afghanica* (Ehrend.) M. Backlund & Thulin, **comb. nov.** = *Gaillonia afghanica* Ehrend. in Biol. Skr. 10: 122. 1959 = *Neogaillonia afghanica* (Ehrend.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973. Afghanistan, Pakistan.

*Plocama asperuliformis* (Lincz.) M. Backlund & Thulin, **comb. nov.** = *Gaillonia asperuliformis* Lincz., Fl. URSS 23: 689. 1958 = *Neogaillonia asperuliformis* (Lincz.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973.

= *Gaillonia kerstani* Ehrend. in Biol. Skr. 10: 117. 1959 = *Neogaillonia kerstani* (Ehrend.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973.

= *Gaillonia chitralensis* Nazim. in Nasir & Ali, Fl. Pakistan 190: 106. 1989. Afghanistan, Pakistan, Tadjikistan.

*Plocama aucheri* (Guill.) M. Backlund & Thulin, **comb. nov.** = *Jaubertia aucheri* Guill. in Ann. Sci. Nat., Bot., sér. 2, 16: 60. 1841 = *Gaillonia aucheri* (Guill.) Jaub. & Spach in Ann. Sci. Nat., Bot., sér. 2, 20: 87. 1843 = *Neogaillonia aucheri* (Guill.) Puff in J. Linn. Soc., Bot. 84: 375. 1982.

Oman, United Arab Emirates, Iran, Afghanistan, Pakistan.

*Plocama botschantzevii* (Lincz.) M. Backlund & Thulin, **comb. nov.** = *Neogaillonia botschantzevii* Lincz. in Novosti Sist. Vyssh. Rast. 10: 232. 1973 = *Gaillonia botschantzevii* (Lincz.) Ehrend. in Rechinger, Fl. Iranica 176: 25. 2005. Uzbekistan.

*Plocama brevifolia*, **comb.** based on *Putoria brevifolia* Coss. & Durieu ex Pomel. The complex taxonomy and nomenclature of this species is dealt with by Backlund & Thulin (2007). Morocco, Algeria.

*Plocama bruguieri* (A. Rich. ex DC.) M. Backlund & Thulin, **comb. nov.** = *Gaillonia bruguieri* A. Rich. ex DC., Prodr. 4: 574. 1830 = *Neogaillonia bruguieri* (A. Rich. ex DC.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 226. 1973.

= *Gaillonia richardiana* Jaub. & Spach in Ann. Sci. Nat., Bot., sér. 2, 20: 83. 1843.

= *Gaillonia incana* Jaub. & Spach in Ann. Sci. Nat., Bot., sér. 2, 20: 84. 1843 = *Neogaillonia incana* (Jaub. & Spach) Lincz. in Novosti Sist. Vyssh. Rast. 10: 227. 1973. Iran, Turcomania, Afghanistan.

*Plocama bucharica* (B. Fedtsch. & Desjat.) M. Backlund & Thulin, **comb. nov.** = *Gaillonia bucharica* B. Fedtsch. & Desjat. in B. Fedtsch., Rastit. Turkest.: 708. 1915 = *Neogaillonia bucharica* (B. Fedtsch. & Desjat.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973. Tadjikistan.

*Plocama calabrica*, **comb.** based on *Asperula calabrica* L. f. The complex taxonomy and nomenclature of this species is dealt with by Backlund & Thulin (2007). Mediterranean region, widespread.

*Plocama calcicola* (Puff) M. Backlund & Thulin, **comb. nov.** = *Neogaillonia calcicola* Puff in Nordic J. Bot. 8: 333. 1988 = *Gaillonia calcicola* (Puff) Thulin in Nordic J. Bot. 18: 36. 1998. Somalia.

*Plocama calycoptera* (Decne.) M. Backlund & Thulin, **comb. nov.** = *Spermacoce calycoptera* Decne. in Ann. Sci. Nat., Bot., sér. 2, 2: 267. 1834, as “*calyptera*” = *Gaillonia calycoptera* (Decne.) Jaub. & Spach in Ann. Sci. Nat. Bot., sér. 2, 20: 86. 1843 = *Pterogaillonia calycoptera* (Decne.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 233. 1973 = *Jaubertia calycoptera* (Decne.) Täckh. & Boulos in Publ. Cairo Univ. Herb. 5: 95. 1974 = *Neogaillonia calycoptera* (Decne.) Puff in J. Linn. Soc., Bot. 84: 374. 1982.

= *Pterogaillonia stscherbinovskii* Lincz. in Novosti Sist. Vyssh. Rast. 10: 234. 1973 = *Neogaillonia stscherbinovskii* (Lincz.) Puff in J. Linn. Soc., Bot. 84: 374. 1982.

Egypt, Sudan, Israel, Jordania, Saudi Arabia, Oman, United Arab Emirates, Bahrain, Iran, Pakistan. The epithet “*calycoptera*”, meaning “winged calyx”, was originally spelled “*calyptera*” by Decaisne, an orthographic error that is corrected under Art. 60.1.

*Plocama crocyllis* (Sond.) M. Backlund & Thulin, **comb. nov.** = *Anthospermum crocyllis* Sond., Fl. Cap. 3: 32. 1865 = *Crocyllis anthospermoides* E. Mey. ex K. Schum. in Engler & Prantl, Nat. Pflanzenfam. 4(4): 132. 1891 = *Gaillonia crocyllis* (Sond.) Thulin in Nordic J. Bot. 18: 36. 1998. Namibia, South Africa.

*Plocama crucianelloides* (Jaub. & Spach) M. Backlund & Thulin, **comb. nov.** = *Gaillonia crucianelloides* Jaub. & Spach in Ann. Sci. Nat., Bot., sér. 2, 20: 86. 1843 = *Pterogaillonia crucianelloides* (Jaub. & Spach) Lincz. in Novosti Sist. Vyssh. Rast. 10: 235. 1973 = *Neogaillonia crucianelloides* (Jaub. & Spach) Puff in J. Linn. Soc., Bot. 84: 374. 1982. Bahrain, Iran.

- Plocama dubia* (Aitch. & Hemsl.) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia dubia* Aitch. & Hemsl. in Trans. Linn. Soc. London, ser. 2, 3: 73. 1888 ≡ *Neogaillonia dubia* (Aitch. & Hemsl.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973. Afghanistan.
- Plocama eriantha* (Jaub. & Spach) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia eriantha* Jaub. & Spach in Ann. Sci. Nat. Bot., sér. 2, 20: 85. 1843 ≡ *Neogaillonia eriantha* (Jaub. & Spach) Lincz. in Novosti Sist. Vyssh. Rast. 10: 229. 1973. Iran.
- Plocama hymenostephana* (Jaub. & Spach) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia hymenostephana* Jaub. & Spach in Ann. Sci. Nat., Bot., sér. 2, 20: 85. 1843 ≡ *Pseudogaillonia hymenostephana* (Jaub. & Spach) Lincz. in Novosti Sist. Vyssh. Rast. 10: 236. 1973 ≡ *Neogaillonia hymenostephana* (Jaub. & Spach) Puff in J. Linn. Soc., Bot. 84: 375. 1982.  
= *Gaillonia humifusa* Jaub. & Spach in Ann. Sci. Nat., Bot., sér. 2, 20: 85. 1843. Oman, United Arab Emirates, Iran, Afghanistan, Pakistan, India (Punjab).
- Plocama iljinii* (Lincz.) M. Backlund & Thulin, **comb. nov.** ≡ *Neogaillonia iljinii* Lincz. in Novosti Sist. Vyssh. Rast. 10: 227. 1973 ≡ *Gaillonia iljinii* (Lincz.) Ehrend. & Schönb.-Tem. in Rechinger, Fl. Iranica 176: 37. 2005. Uzbekistan.
- Plocama inopinata* (Lincz.) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia inopinata* Lincz. in Bot. Mater. Gerb. Bot. Inst. Komarova Akad. Nauk S.S.S.R. 22: 216. 1963 ≡ *Neogaillonia inopinata* (Lincz.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 232. 1973. Uzbekistan.
- Plocama jolana* (Thulin) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia jolana* Thulin in Nordic J. Bot. 18: 34. 1998. Yemen.
- Plocama kandaharensis* (Ehrend. & Qarar ex Ehrend. & Schönb.-Tem.) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia kandaharensis* Ehrend. & Qarar ex Ehrend. & Schönb.-Tem. in Rechinger, Fl. Iranica 176: 38. 2005. Afghanistan.
- Plocama macrantha* (Blatt. & Hallb.) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia macrantha* Blatt. & Hallb. in J. Indian Bot. 1: 170. 1920 ≡ *Neogaillonia macrantha* (Blatt. & Hallb.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973. Iran, Afghanistan, Pakistan.
- Plocama mestscherjakovii* (Lincz.) M. Backlund & Thulin, **comb. nov.** ≡ *Neogaillonia mestscherjakovii* Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973 ≡ *Gaillonia mestscherjakovii* (Lincz.) Ehrend. in Rechinger, Fl. Iranica 176: 25. 2005. Uzbekistan, Tadjikistan.
- Plocama olivieri* (A. Rich. ex DC.) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia olivieri* A. Rich. ex DC., Prodr. 4: 574. 1830 ≡ *Neogaillonia olivieri* (A. Rich. ex DC.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 226. 1973. Iraq.
- Plocama pendula* Aiton, Hort. Kew. 1: 292. 1789. Canary Islands.
- Plocama puberula* (Balf. f.) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia puberula* Balf. f. in Proc. Roy. Soc. Edinburgh 11: 836. 1882 ≡ *Neogaillonia puberula* (Balf. f.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 229. 1973. Yemen (Socotra).
- Plocama putorioides* (Radcl.-Smith) M. Backlund & Thulin, **comb. nov.** ≡ *Jaubertia putorioides* Radcl.-Sm. in Hooker's Icon. Pl. 7(4): t. 3682. 1971 ≡ *Neogaillonia putorioides* (Radcl.-Sm.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973 ≡ *Gaillonia putorioides* (Radcl.-Sm.) Thulin in Nordic J. Bot. 18: 37. 1998. Yemen (Socotra).
- Plocama reboudiana* (Coss. & Durieu) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia reboudiana* Coss. & Durieu in Bull. Bot. Soc. Fr. 2: 250. 1855 ≡ *Choulettia reboudiana* (Coss. & Durieu) Pomel, Nouv. Mat. Fl. Atl.: 81. 1874 ≡ *Neogaillonia reboudiana* (Coss. & Durieu) Puff in J. Linn. Soc., Bot. 84: 375. 1982 ≡ *Jaubertia reboudiana* (Coss. & Durieu) Ehrend. & Schönb.-Tem. in Rechinger, Fl. Iranica 176: 39. 2005. Morocco, Algeria.
- Plocama rosea* (Hemsl.) M. Backlund & Thulin, **comb. nov.** ≡ *Aitchisonia rosea* Hemsl. in J. Linn. Soc., Bot. 19: 166. 1882. Iran, Afghanistan, Pakistan.
- Plocama somaliensis* (Puff) M. Backlund & Thulin, **comb. nov.** ≡ *Neogaillonia somaliensis* Puff in Nordic

J. Bot. 8: 331. 1988 ≡ *Gaillonia somaliensis* (Puff) Thulin in Nordic J. Bot. 18: 36. 1998. Somalia.

***Plocama szowitsii*** (DC.) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia szowitsii* DC., Prodr. 4: 574. 1830, as “sowitzii” ≡ *Jaubertia szowitsii* (DC.) Takht., Fl. Erevana, ed. 2: 205. 1972 (as *szovitsii*) ≡ *Neogaillonia szowitsii* (DC.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 226. 1973 (as *szovitsii*).

Armenia, Azerbaijan, Iran. The epithet was spelled “sowitzii” in Candolle (1830) but as he acknowledged “Szowits” as the collector, this appears to be a typographical error to be corrected under Art. 60.1. On the other hand, “Szowits” was clearly a deliberate spelling of the collector’s name, being a widely accepted transliteration of Сович until relatively recent standardization. The supposed “correction” to “szovitsii” by Russian authors, although understandable, is not permitted under Art. 60.

***Plocama thymoides*** (Balf. f.) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia thymoides* Balf. f. in Proc. Roy. Soc. Edinburgh 11: 837. 1882 ≡ *Neogaillonia thymoides* (Balf. f.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973. Yemen (Socotra).

***Plocama tinctoria*** (Balf. f.) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia tinctoria* Balf. f. in Proc. Roy. Soc. Edinburgh 11: 836. 1882 ≡ *Neogaillonia tinctoria* (Balf. f.) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973.

= *Gaillonia tinctoria* var. *glabra* Radcl.-Sm. in Kew Bull. 25: 186. 1971. Yemen (Socotra), Somalia.

***Plocama trichophylla*** (Popov ex Tscherneva) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia trichophylla* Popov ex Tscherneva in Vvedensky, Fl. Uzbekistana 5: 642. 1961 ≡ *Neogaillonia trichophylla* (Popov ex Tscherneva) Lincz. in Novosti Sist. Vyssh. Rast. 10: 231. 1973. Uzbekistan.

***Plocama vassilczenkoi*** (Lincz.) M. Backlund & Thulin, **comb. nov.** ≡ *Neogaillonia vassilczenkoi* Lincz. in Novosti Sist. Vyssh. Rast. 10: 228. 1973 ≡ *Gaillonia vassilczenkoi* (Lincz.) Ehrend. & Schönb.-Tem. in Rechinger, Fl. Iranica 176: 37. 2005. Uzbekistan, Tadjikistan.

***Plocama yemenensis*** (Thulin) M. Backlund & Thulin, **comb. nov.** ≡ *Gaillonia yemenensis* Thulin in Nordic J. Bot. 18: 32. 1998. Yemen, Oman.

### Excluded species

*Gaillonia pulchella* Podlech in Mitt. Bot. Staatssamml. München 7: 107. 1968 ≡ *Neogaillonia pulchella* (Podlech) Lincz. in Novosti Sist. Vyssh. Rast. 10: 230. 1973 ≡ *Asperula pulchella* (Podlech) Ehrend. & Schönb.-Tem. in Rechinger, Fl. Iranica 176: 136. 2005.

*Putoria indica* DC., Prodr. 4: 577. 1830 ≡ *Neanotis indica* (DC.) W.H. Lewis in Ann. Missouri Bot. Gard. 53: 38. 1966.

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Appendix. List of taxa investigated and GenBank accession numbers for the sequences used. For new sequences voucher information is given and species names and accession numbers are in bold.

Species name, tribe<sup>a</sup>, voucher/reference<sup>b</sup>, *rbcL*, *rps16*, *trnT-F*.

*Aitchisonia rosea* Hemsl., Pae, *Rafei & Zangoeei 25651*, FUMH, **DQ662172, DQ662195, DQ662134**. *Anthospermum herbaceum* L. f., Ant, 1, X83623, –, –, *Anthospermum tricostatum* Sond., Ant, 11, –, AF257898, –, *Anthospermum welwitschii* Hiern, Ant, Luke 8928, UPS, –, –, **DQ662220**. *Argostemma hookeri* King, Arg, 3, Z68788, –, –, *Argostemma rupestre* Ridl., Arg, 6, –, AF002756, –, *Asperula arcadiensis* Sims, Rub, 6, –, AF004029, –, *Asperula cynanchica* L., Rub, *Andreasen 321*, UPS, –, –, **DQ662135**. *Asperula laevigata* L., Rub, 2, X81092, –, –, *Asperula tinctoria* L., Rub, *Andreasen 322*, UPS, –, –, **DQ662136**. *Choulettia reboudiana* Pomel, Pae, *Samuelsson 6846*, S, **DQ662173, DQ662196, DQ662137**. *Coccocypselum hirsutum* Bartling, Cou, 10, X87145, –, –, *Coccocypselum lanceolatum* Pers., Cou, 6, –, AF004036, –, *Coprosma antipoda* W.R.B. Oliv., Ant, 11, –, AF257902, –, *Coprosma pumila* Hook. f., Ant, 10, X87146, –, –, *Coussarea macrophylla* Müll. Arg., Cou, 4, 6, Y11847, AF004040, –, *Coussarea* sp., Cou, 8, –, –, AF152612. *Craterispermum brachynematum* Hiern, Cra, 7, AJ288629, –, –, *Craterispermum laurinum* Benth., Cra, 12, –, AF331645, –, *Dannacanthus indicus* Gaertn. f., Mor, 3, 12, Z68793, AF331647, –, *Danais* sp., Dan, 12, –, AF331648, –, *Danais xanthorrhoea* Bremek., Dan, 3, *Bremer 3079*, UPS, Z68794, –, **DQ662138**. *Gaertnera paniculata* Benth., Gae, 6, –, AF002736, –, *Gaertnera* sp., Gae, 3, Z68797, –, –, *Gaertnera* sp., Gae, *Bremer & al. 4008*, UPS, –, –, **DQ662139**. *Gaillonia crocylis* (Sond.) Thulin, Pae, *Acocks 18171*, UPS, **DQ662174, DQ662197, DQ662140**. *Gaillonia eriantha* Jaub. & Spach, Pae, *Ehrendorfer 4210*, S, **DQ662175, DQ662198, DQ662141**. *Gaillonia olivieri* A. Rich. ex DC., Pae, *Khorasan 691*, S, **DQ662176, DQ662199, DQ662142**. *Gaillonia tinctoria* Balf. f., Pae, *Thulin 10946*, UPS, **DQ662177, DQ662200, DQ662143**. *Gaillonia yemenensis* Thulin, Pae, 7, *Thulin 9365*, UPS, AJ288630, **DQ662201, DQ662221**. *Galium album* Mill., Rub, 2, 6, X81090, AF004050, –, *Galium thunbergianum* Eckl. & Zeyh., Rub, *Luke 8876*, UPS, –, –, **DQ662144**. *Hedyotis fruticosa* L., Spe, 3, Z68799, –, –, *Hedyotis serpens* H.B. & K., Spe, 13, –, AF333377, –, *Jaubertia aucheri* Guill., Pae, *Thulin 9963*, UPS, **DQ662178, DQ662202, DQ662145**. *Kelloggia galioides* Torr., Pae, *Holmgren & al. 2437*, UPS, **DQ662179, DQ662203, DQ662146**. *Lasianthus coffeoides* Fyson, Las, 6, –, AF004061, –, *Lasianthus kilimandscharicus* K. Schum., Las, *Lantz 119*, UPS, –, –, **DQ66214**. *Lasianthus pedunculatus* E.A. Bruce, Las, 3, Z68802, –, –, *Leptodermis potaninii* Batalin, Pae, *Andreasen 230*, UCBG, **DQ662180, DQ662204, DQ662148**. *Luculia grandifolia* Ghose, unplaced, 1, *Bremer 2713*, UPS, X83648, **DQ662205, DQ662149**. *Mitchella repens* L., Mor, 3, 6, Z68805, AF001441, –, *Morinda candollei* Beauvis., Mor, *Munzinger & McPherson 701*, UPS, –, –, **DQ662150**. *Morinda citrifolia* L., Mor, 1, 9, X83651, AJ320078, –, *Mycetia malayana* Craib, Arg, 3, 6, 8, Z68806, AF002771, AF152622. *Ophiorrhiza mungos* L., Oph, 1, 6, *Bremer 3301*, UPS, X83656, AF004064, **DQ662151**. *Paederia bojeriana* Drake, Pae, *Razafimandimbison & H. Bremer 483*, UPS, **DQ662181, DQ662206, DQ662152**. *Paederia farinosa* Puff, Pae, *Kårehed & al. 225*, UPS, **DQ662182, DQ662207, DQ662153**. *Paederia foetida* L., Pae, 12, 6, 8, AF332373, AF004065, AF152619. *Paederia lanata* Puff, Pae, PDB 1152, UPS, **DQ662183, DQ662208, DQ662154**. *Paederia majungensis* Homolle, Pae, *Nilsson & al. D152*, UPS, **DQ662184, DQ662209, DQ662155**. *Paederia mandranensis* Homolle, Pae, *Razafimandimbison & H. Bremer 504*, UPS, **DQ662185, DQ662210, DQ662156**. *Paederia pilifera* Hook. f., Pae, *Puff 971228-1/1*, WU, **DQ662186, DQ662211, DQ662157**. *Paederia pospischilii* K. Schum., Pae, *Luke & al. 9456*, UPS, **DQ662187, DQ662212, DQ662158**. *Paederia sambiranensis* Homolle, Pae, *Kårehed & al. 238*, UPS, **DQ662188, DQ662213, DQ662159**. *Paederia taolagnaroensis* S.G. Razafim. & C.M. Taylor, Pae, *Razafimandimbison 515*, UPS, **DQ662189, DQ662214, DQ662160**. *Paederia thouarsiana* Baill., Pae, *Kårehed & al. 236*, UPS, –, –, **DQ662161**. *Pentas carnea* Benth., Spe, 6, –, AF002750, –, *Pentas lanceolata* Defl., Spe, 1, X83659, –, –, *Phyllis nobla* L., Ant, 3, 6, Z68814, AF003613, –, *Plocama pendula* Aiton, Pae, 3, 6, *Andreasen 1*, UPS, Z68816, AF004071, **DQ662162**. *Pseudogaillonia hymenostephana* (Jaub. & Spach) Lincz., Pae, *Thulin 9993*, UPS, **DQ662190, DQ662215, DQ662163**. *Psychotria acuminata* Benth., Psy, 14, –, AF149359, –, *Psychotria kirkii* Hiern, Psy, 1, 15, X83663, –, AY538469. *Pterogaillonia calycoptera* (Decne.) Lincz., Pae, *Miller 6604*, UPS, **DQ662191, DQ662216, DQ662164**. *Putoria brevifolia* Coss. & Durieu. ex Pomel, Pae, *Thulin 2329*, UPS, **DQ662192, DQ662217, DQ662165**. *Putoria calabrica* (L. f.) DC., Pae, 7, 6, *Jonsell 4216*, UPS, AJ288620, AF004072, **DQ662166**. *Rubia fruticosa* Aiton, Rub, 6, 5, –, AF004078, AF102475. *Rubia horrida* (Thunb.) Puff, Rub, *Bremer & al. 4266*, UPS, –, –, **DQ662167**. *Rubia tinctorum* L., Rub, 2, X81104, –, –, *Saprosma foetens* K. Schum., Psy, *Klackenberg 325*, S, **DQ662193, DQ662218, DQ662168**. *Saprosma fruticosum* Blume, Psy, *Ridsdale 157*, L, **DQ662194**, –, **DQ662169**. *Schradera* sp., Sch, 6, –, AF003617, –, *Schradera subandina* Krause, Sch, 4, Y11859, –, –, *Serissa foetida* Lam., Pae, 3, 6, 8, Z68822, AF004081, AF152618. *Spermacoce confusa* Rendle, Spe, 6, –, AF003619, –, *Spermacoce filituba* Verdc., Spe, *Luke 9022*, UPS, –, –, **DQ662170**. *Spermacoce hispida* L., Spe, 7, AJ288623, –, –, *Spermadictyon suaveolens* Roxb., Pae, 3, *Bremer 3133*, UPS, Z68824, **DQ662219, DQ662171**. *Theligonum cynocrambe* L., The, 1, 6, 8, X83668, AF004087, AF152621. *Urophyllum ellipticum* Thwaites, Uro, 7, AJ288627, –, –, *Urophyllum glabrum* Jack, Uro, 6, –, AF004089, –.

<sup>a</sup>Tribes shortening: Pae, Paederieae; Ant, Anthospermeae; Arg, Argostemmatae; Rub, Rubieae; Cou, Coussareae; Cra, Craterispermeae; Mor, Morindeae; Dan, Danaideae; Gae, Gaertnerae; Spe, Spermacoceae; Las, Lasiantheae; Oph, Ophiorrhizeae; Psy, Psychotriaceae; Sch, Schradereae; The, Theligoneae; Uro, Urophyllae.

<sup>b</sup>Reference numbers correspond to: 1, (Bremer & al., 1995); 2, (Manen & Natali, 1995); 3, (Bremer, 1996); 4, (Bremer & Thulin, 1998); 5, (Struwe & al., 1998); 6, (Andersson & Rova, 1999); 7, (Bremer & Manen, 2000); 8, (Rova & al., 2002); 9, (Novotny & al., 2002); 10, (Bremer, 1997); 11, (Anderson & al., 2001); 12, (Andersson, unpubl.); 13, (Andersson & al., 2002); 14, (Andersson & Taylor, unpubl.); (Andersson & Antonelli, 2005).

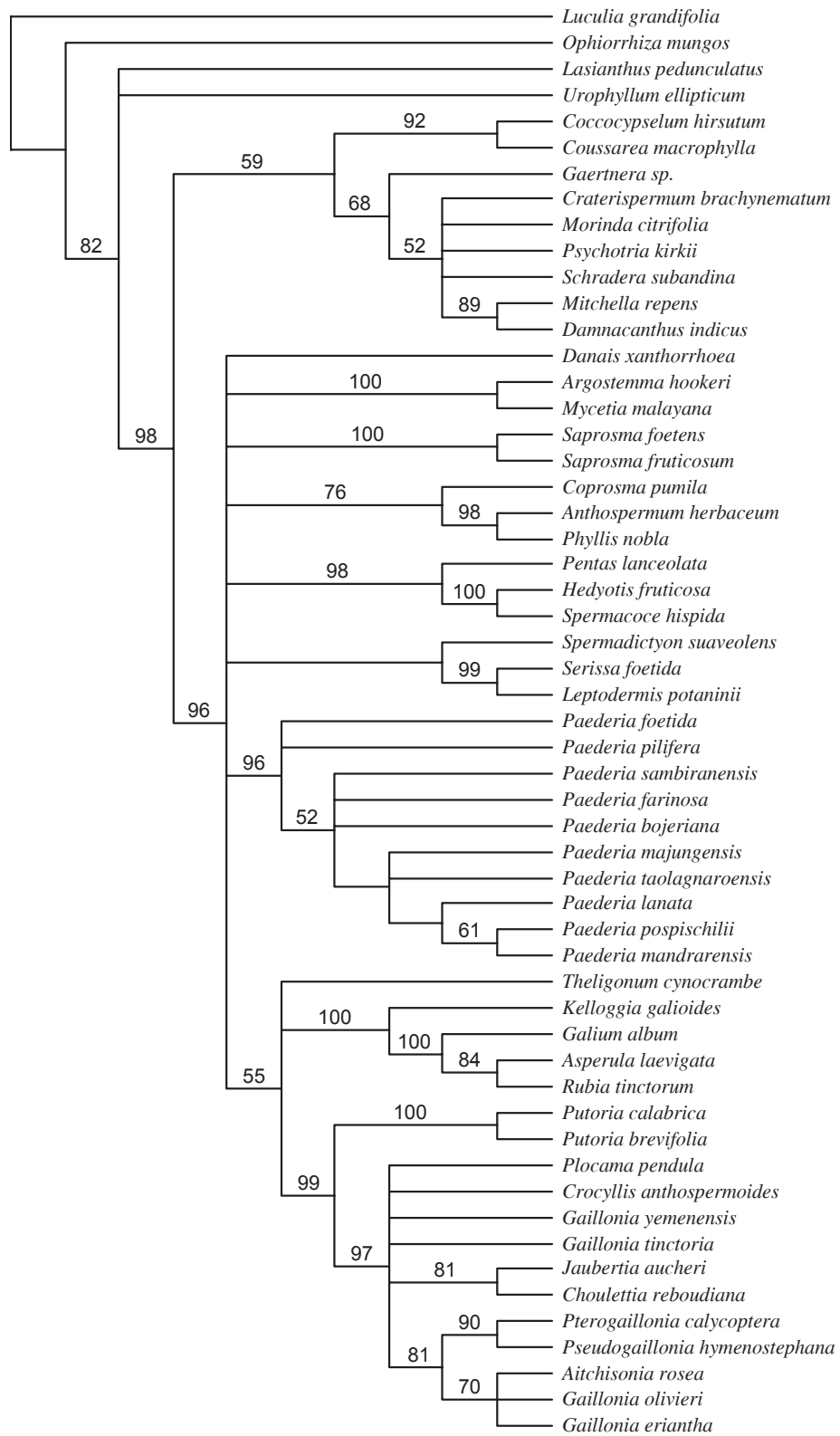


Fig. 2. The consensus tree of the *rbcL* analysis. Bootstrap values are shown above the bars.

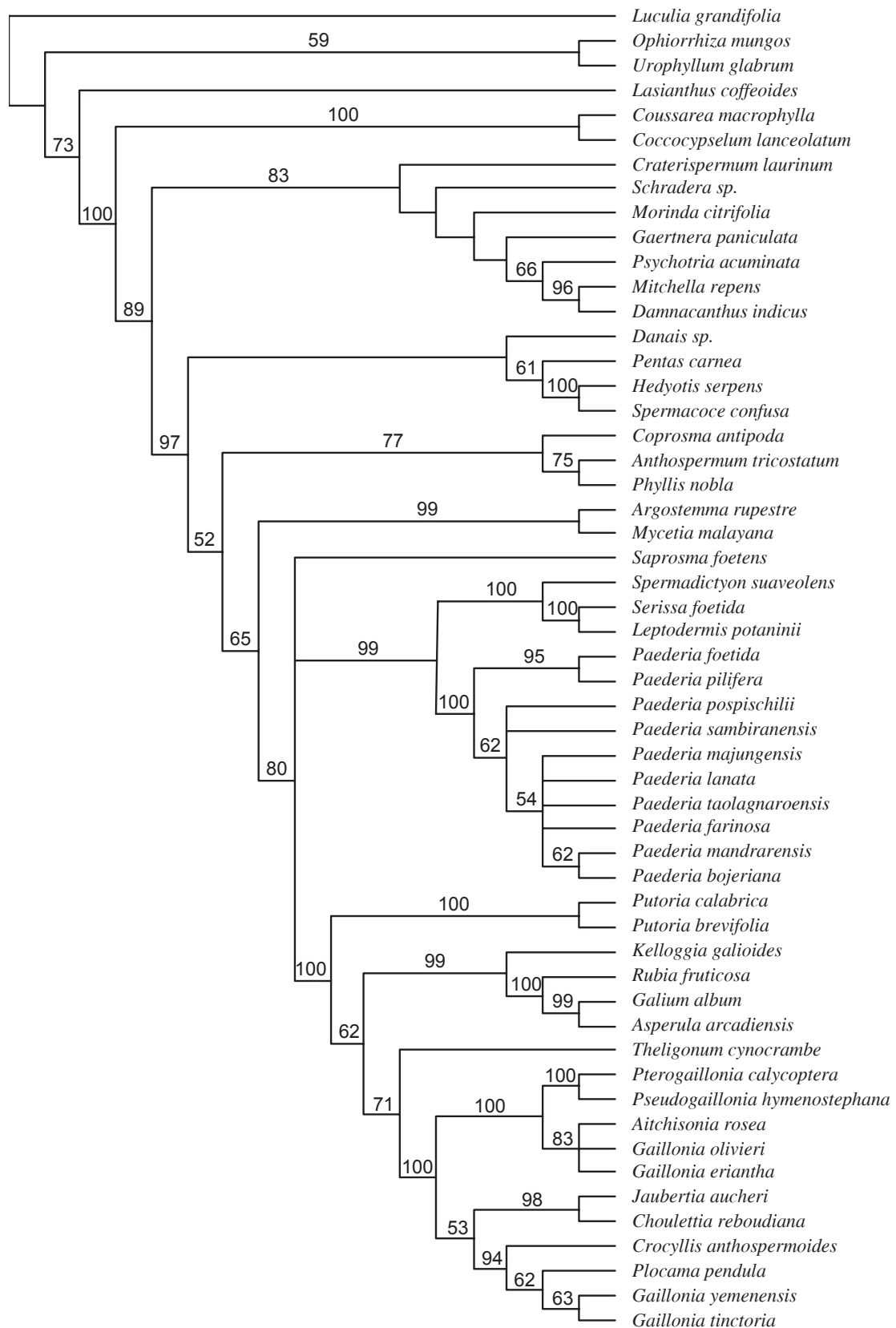


Fig. 3. The consensus tree of the *rps16* analysis. Bootstrap values are shown above the bars.



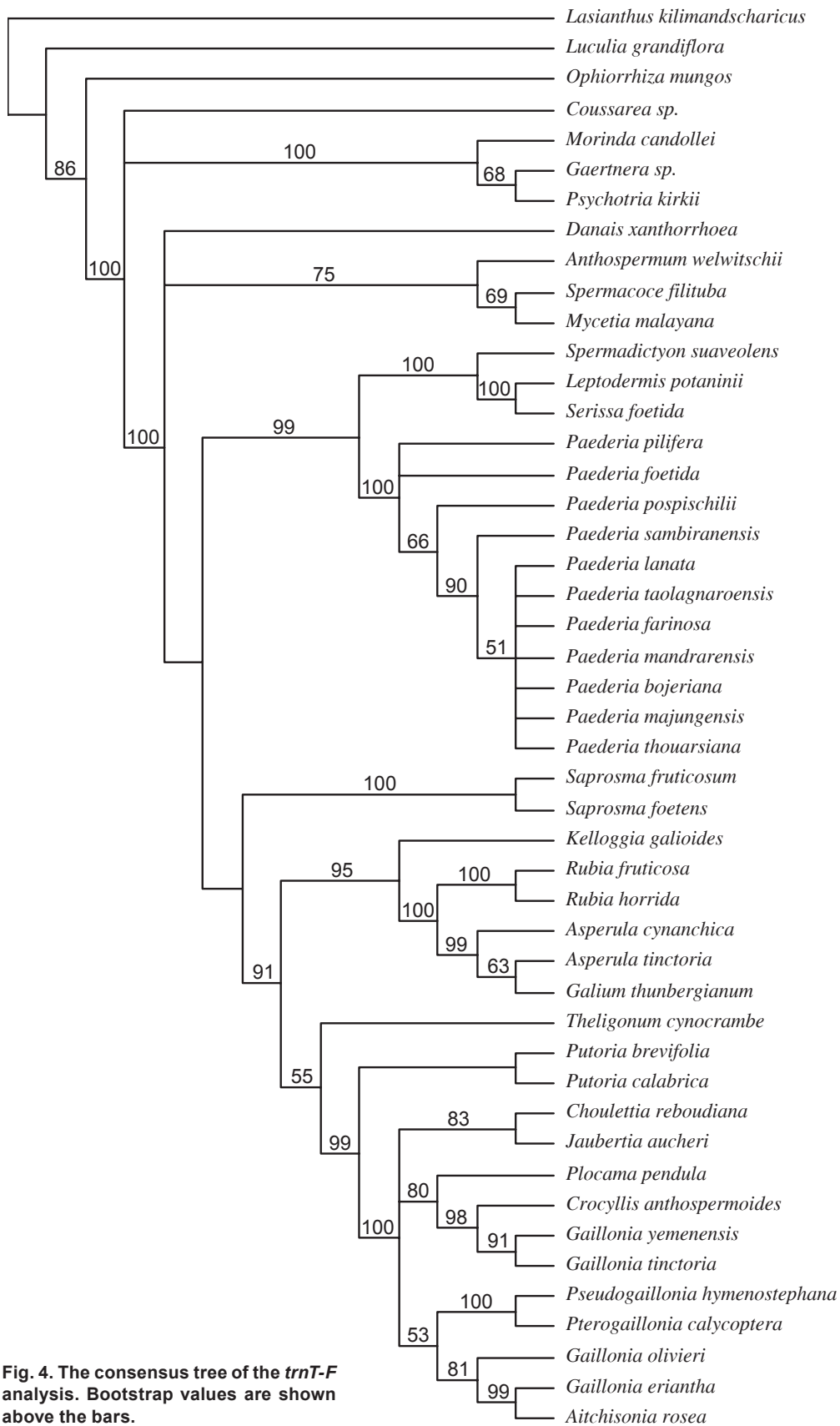


Fig. 4. The consensus tree of the *trnT-F* analysis. Bootstrap values are shown above the bars.