Molecular phylogeny confirms the placement of enigmatic *Stachys persepolitana* in *Lamium* (Lamiaceae; subfam. Lamioideae)

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Abstract

The main purpose of this study was to test the hypothesis that *Stachys persepolitana* is extraneous in *Stachys* (tribe Stachydeae, subfam. Lamioideae, Lamiaceae) and instead belongs to the genus *Lamium* (Lamiaceae, Lamioideae). We investigated the phylogenetic position of *S. persepolitana* using plastid (*rps16* intron, *trnL*-F and *matK* regions) and nuclear (nrITS) DNA sequence data with both parsimony and Bayesian phylogenetic approaches. Plastid and nuclear data strongly support that *S. persepolitana* is extraneous in *Stachys* and belongs in fact to the genus *Lamium*. Morphological characters also corroborate its placement in *Lamium*. Most of the morphological features used to distinguish *S. persepolitana* from the rest of *Stachys* fit with *Lamium*. Anterior pair of stamens bending outward after pollination, anthers hairy and mericarps triquetrous are the most important characters correlating *S. persepolitana* to *Lamium*.

Introduction

The rare species *Stachys persepolitana* Boissier (1846: 56) occurs in subalpine habitats with humid conditions and is endemic to south and south-west of Iran. Several years after description of this species, Boissier (1879) expressed himself serious doubts about the placement of this species in the genus *Stachys* Linnaeus (1753: 580). He stated that the samples lacked any corolla, but in general vegetative morphology they resembled *Stachys corsica* Persoon (1806: 124) (Boissier 1879); hence, it seems that the main reason why this species had been treated as a member of the genus *Stachys* was this vegetative similarity rather than floral morphology. Later, Bornmüller (1915) found that the cleistogamous flowers in *S. persepolitana* are a remarkable character; a feature that is shared with some *Lamium* Linnaeus (1753: 579) species (Sato et al. 2013) but not observed elsewhere in *Stachys*. Bhattacharjee (1982) placed the species in *Stachys* section *Olisia* Dumortier (1827: 44) subsection *Annuae* Bhatacharjee (1982: 86) because of its fruiting calyx that is usually gibbous at the base with lower teeth curved towards the mouth. *Stachys* section *Olisia* has recently been shown to be heterogeneous and polyphyletic (Salmaki et al. 2013). Consequently, in the revision of *Stachys* in Iran, Salmaki et al. (2012a) mentioned that the existence of such unusual characters might indicate an isolated position of *S. persepolitana*. In the most recent edition of Flora of Iran (Jamzad 2012), this species was treated as a member of the genus *Lamium*. However, a detailed discussion on morphological evidence leading to this treatment is missing. Moreover, the description provided is in Persian and of limited access to plant taxonomists.

The aim of the present study is to determine the position of this taxon using a phylogenetic approach based on DNA sequences and to provide an emended description of easier availability.
Infrageneric classification of *Lamium* was presented first by Bentham (1832–1836, 1848), but revised by Briquet (1895–1897). These classifications have been updated by Mennema (1989) who recognized three subgenera in the genus: (1) subg. *Lamium* comprising species with hairy anthers; (2) subg. *Orvala* (Linnaeus 1753: 578) Briquet (1897: 254), with the single species *L. orvala* Linnaeus (1759: 1099) that has glabrous anthers; and (3) subg. *Galeobdolon* (Adanson 1763: 190) Ascherson (1864: 525), also with glabrous anthers. Within subg. *Lamium*, Mennema (1989) discerned the following three sections: (1) sect. *Lamium* comprising species with bracteoles and a straight corolla tube; (2) sect. *Lamiotypus* Dumortier (1827: 45) consisting of species with bracteoles and a sigmoid corolla tube that is abruptly dilated and ventrally saccate and (3) sect. *Amplexicaule* Mennema (1989: 48) including species that lack bracteoles.

The molecular phylogenetic analyses performed here do not suggest a definite placement of *Lamium persepolitanum* in the genus *Lamium*, as there are some conflicts in the branching pattern between the nrITS and cpDNA trees (Figs. 1, 2). However, the cpDNA topology, which is generally more strongly supported, suggests also a close relationship between *L. persepolitanum* and *L. amplexicaule* (BS=95, PP=1.00). The genus *Lamium* is evidently the subject of complex evolutionary history caused mainly by intensive hybridization events (Bendiksby et al. 2011b). Therefore, the genus is still in need of a comprehensive molecular phylogenetic study including adequate numbers of taxa and markers.

**FIGURE 3.** Photographs of *Lamium persepolitanum*. A. Lectotype, G-BOIS, B. Habit, C. Calyx, D. Corolla, E. Inflorescence of *L. persepolitanum* (from Yasaman Salmaki).

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