

Taxonomic Significance of Major Alkaloids in the Genus *Priestleya*

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Key Word Index—*Priestleya*; Fabaceae; Lipariae; quinolizidine alkaloids; bipiperidyl alkaloids; chemotaxonomy; generic relationships.

Abstract—The genus *Priestleya* is known to contain the bipiperidyl alkaloid anabasine and an alkaloid tentatively identified as lupanine. In a study of 16 different species, several quinolizidine alkaloids and some bipiperidyl alkaloids were identified. A remarkable difference was found between the two sections of *Priestleya*: sect. *Priestleya* contains sparteine, lupanine and other tetracyclic quinolizidine alkaloids, while the sect. *Anisothea* contains the bipiperidyl alkaloid anabasine and the bicyclic quinolizidine lupanine. The chemical distinction agrees with morphological evidence that the generic concept of *Priestleya* is artificial and that two distinct genera should be recognized.

Introduction

As part of an evaluation of alkaloids as taxonomic characters in the tribe Lipariae, we investigated *Priestleya* DC., a genus of ca. 20 species endemic to the fynbos region of South Africa. The alkaloids of *Priestleya* have remained virtually unknown, despite interesting results obtained in a preliminary study [1]. Some species were found to contain the bipiperidyl alkaloid anabasine as major compound, while other species had an alkaloid tentatively identified as lupanine [1]. To evaluate the taxonomic significance of alkaloids, we extracted 23 samples from 16 different species. Our aim with this study was to provide data for a broad comparison at the generic level. The genus *Priestleya* is currently under taxonomic revision and more detailed studies to determine the significance of alkaloids at the species level are in progress.

Materials and Methods

Plant materials. Voucher specimens of the material used for extraction of alkaloids are listed below.

Priestleya sect. *Priestleya*: *Priestleya calycina* L. Bol.: 1, *Stokoe s.n. sub PRE 53058* (PRE); 2, *Boucher 1283* (STE); 3, *Le Maire 362* (STE). *P. capitata* (Thunb.) DC.: *Stokoe 2514* (PRE). *P. hirsuta* DC.: *Vlok 1242* (JRAU). *P. laevigata* (L.) DC.: 1, *Van Wyk 2639* (JRAU); 2, *C. M. van Wyk 1505* (PRE); 3, *Taylor 10111* (PRE); 4, *Van Wyk 2780* (JRAU). *P. latifolia* Benth.: *Kruger 81* (PRE). *P. umbellifera* (Thunb.) DC.: 1, *Schlechter 10201* (PRE); 2, *Esterhuysen 14906* (PRE). *Priestleya* sp. nov. A: *Van Wyk 2778* (JRAU). *Priestleya* sp. nov. B: 1, *C. M. van Wyk 1498* (PRE); 2, *Taylor 10499* (PRE). *Priestleya* sp. nov. C: *Boucher & Oliver s.n. 20.05. 1989* (STE).

Priestleya sect. *Anisothea* DC. (= *Xiphotheca* Eckl. & Zeyh.): *P. elliptica* DC.: *Haynes 1117* (PRE). *P. glauca* Salter: *Boucher 3885* (PRE). *P. guthriei* L. Bol.: *Oliver 4288* (PRE). *P. schlechteri* L. Bol.: *C. M. van Wyk 2581* (JRAU). *P. tecta* DC.: *Grobbelaar 2702* (PRE). *P. tomentosa* (L.) Druce: *Van Wyk 2749* (JRAU). *Priestleya* sp. nov. D: *Vlok 1381* (PRE).

Procedures. Dry plant material was extracted as described elsewhere [2] and the alkaloids identified by comparative TLC and comparative GC using authentic reference samples obtained in several previous studies [3-6]. A pure sample of anabasine was extracted from leaves and seeds of *Nicotiana glauca* R. C. Grah. Two TLC systems were used: Merck 60 F₂₅₄ silica gel plates (0.25 mm layer thickness) with CHCl₃-Cyclohexane-Et₃NH (4:5:1) as eluent and Merck aluminium oxide F₂₅₄ (type E) plates (0.25 mm layer thickness) with 1.5% MeOH in CHCl₃ as eluent. The plates were dried at 100°C for 3 min, studied under UV₂₅₄ and UV₃₆₅ and then sprayed with iodoplatinate reagent. GC spectra were obtained with a DB-1 fused silica capillary column (30

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m×0.25 mm i.d.; N₂ as carrier gas at 4 ml min⁻¹; column temperature 150°–320° at 6° min⁻¹; 15 min isotherm; injector 230°C; PND detection 300°C; split ratio 30:1; injection volume 1 µl. The identity of the major alkaloids was confirmed by GC-MS analyses of the following extracts: *P. laevigata* (sample 4), *P. capitata*, *P. latifolia*, *P. guthriei*, *P. schlechteri* and *Priestleya* sp. nov. B (sample 2). The MS spectra of all the major alkaloids were identical to results obtained in previous studies. Several unknown minor alkaloids occurred sporadically in some of the extracts but due to the small samples that were available, their identities could not yet be determined by spectroscopic methods.

Piperidyl alkaloids. Anabasine: Rt 8.52, M⁺ 162. Ammodendrine: Rt 15.10, M⁺ 208.

Bicyclic quinolizidine alkaloids. Lupinine: Rt 8.05, M⁺ 169.

Tetracyclic quinolizidine alkaloids. α-Isosparteine: Rt 12.62, M⁺ 234. Sparteine: Rt 13.77, M⁺ 234. Δ11,12-Sparteine: Rt 14.76, M⁺ 232. Isolupanine (11-*epi*-lupanine): Rt 19.60, M⁺ 248. Lupanine: Rt 20.45, M⁺ 248. 13α-Hydroxylupanine: Rt 24.27, M⁺ 264.

Esters of quinolizidine alkaloids. Virgiline-pyrrolyl-carboxylic acid ester: Rt 32.08, M⁺ 357.

Results

The distribution of major alkaloids is presented graphically in Fig. 1. Major alkaloids were defined as all those which constitute more than 10% of any of the extracts. Ammodendrine did not meet this requirement but was included to facilitate the comparison with *Liparia splendens*. Results for the two sections of *Priestleya* were remarkably different. The sect. *Priestleya* contained several tetracyclic quinolizidine alkaloids such as α-isosparteine, sparteine, 11,12-dehydrosparteine, isolupanine (11-*epi*-lupanine), lupanine and 13α-hydroxylupanine. One species, *P. calycina* L. Bol. also had large quantities of virgiline-pyrrolyl-carboxylic acid ester. In contrast, all the species of sect. *Anisothoeae* DC. contained only two major alkaloids, both of which appear to be totally absent in sect. *Priestleya*: lupinine (a bicyclic quinolizidine) and anabasine (a bipiperidyl alkaloid).

Discussion

The results in Fig. 1 are highly significant from a taxonomic point of view. They show that the generic concept of *Priestleya* should be re-evaluated and agree with morphological evidence that the two sections recognized by de Candolle [7] should be raised to the generic rank. *Priestleya sensu lato* was indeed treated in this way by Ecklon and Zeyher in 1836 [8] who proposed the name *Xiphotheca* Eckl. & Zeyh. for a new genus to accommodate all the species of sect. *Anisothoeae*. These species differ from *Priestleya sensu stricto* in their condensed, one- or two-flowered inflorescences and non-intrusive calyces. The racemose inflorescence structure and intrusive calyx of the section *Priestleya* suggest an affinity with genera of the tribe Podalyrieae, where tetracyclic quinolizidine alkaloids are present in *Virgilia* [9] and in *Podalyria* [10, 11]. Furthermore, the presence of virgiline-pyrrolyl-carboxylic acid ester in three samples of *Priestleya calycina* L. Bol. may be viewed as additional evidence of a relationship with the genus *Virgilia*, one of only three known sources of the carboxylic acid esters of virgiline and lupanine (the other two being *Cadia* [12, 13] and *Calpurnia* [13–15], both of the tribe Sophoreae).

We have previously examined the alkaloids of the genus *Liparia* [2] and found large quantitative differences between the two species. *L. parva* contained sparteine and lupanine as major alkaloids, while *L. splendens* had large quantities of ammodendrine. *L. parva* scarcely differ from the species of *Priestleya sensu stricto* except ~~that~~ the many-flowered capitate inflorescences and the close similarity of their alkaloids is very obvious in Fig. 1. Here again, the alkaloidal pattern closely agree with morphological evidence, namely that *Priestleya* s.s and *Liparia* are monophyletic. The generic status of the latter seems a matter of tradition or convenience rather than a true reflection of intergeneric relationships. The distinctive appearance of the two species of *Liparia* may have resulted from changes in pollination strategy [2] which is known to exaggerate taxonomic distance between closely related species [16, 17]. Generic delimitations in the tribe Liparieae is somewhat doubtful [18] and a final decision should only be based

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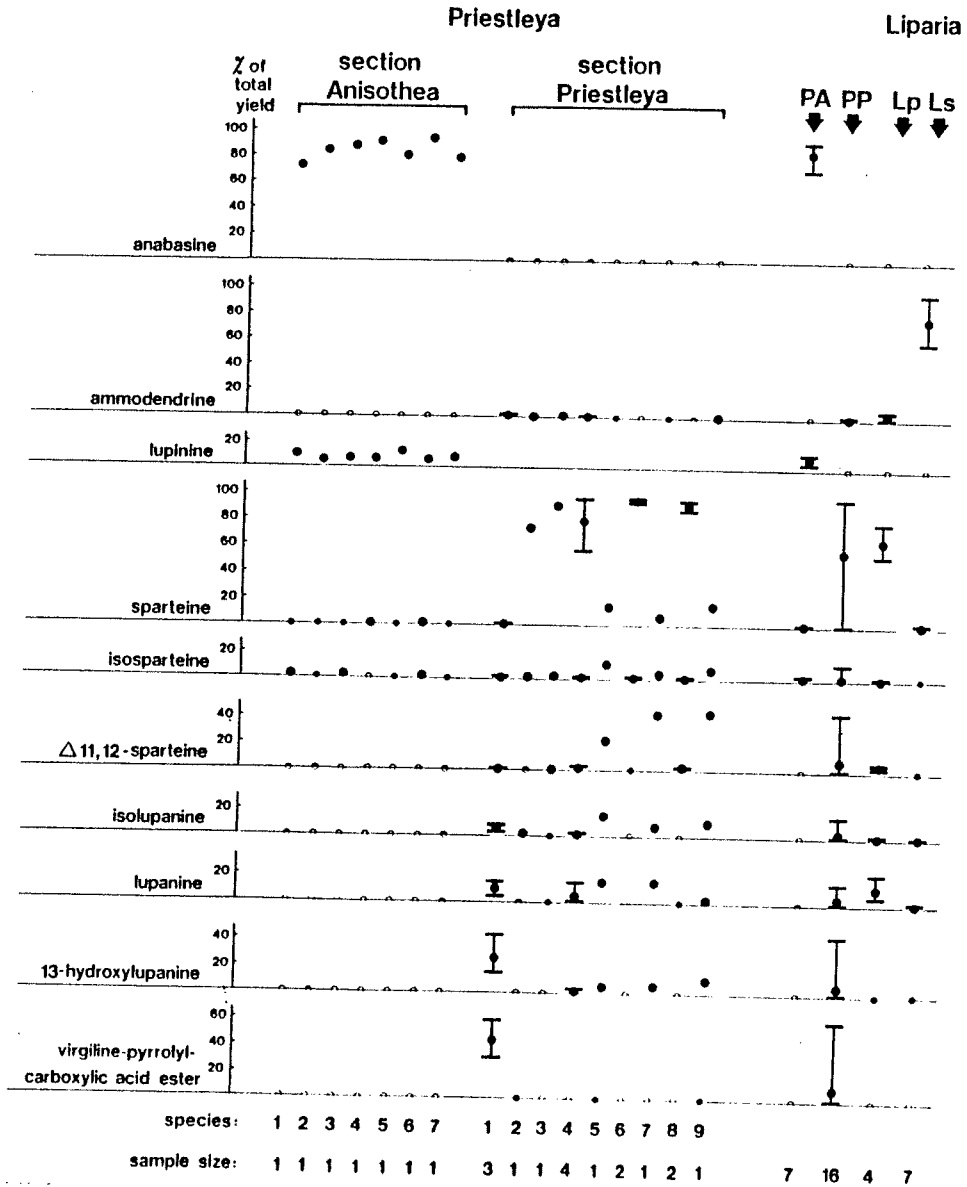


FIG. 1. THE DISTRIBUTION OF MAJOR ALKALOIDS IN THE TWO SECTIONS OF THE GENUS *PRIESTLEYA* AND THE TWO SPECIES OF THE GENUS *LIPARIA*. Note the diagnostically different alkaloid patterns in *Priestleya* sect. *Anisothea* (PA), *Priestleya* sect. *Priestleya* (PP), *Liparia parva* (Lp) and *Liparia splendens* (Ls). (Mean value and range of each alkaloid are shown; small dots indicate trace amounts; circles indicate absence.)

on a rigorous analysis of all available evidence. It is clear from our results that alkaloids provide useful clues about evolutionary relationships.

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