

The Chemotaxonomic Significance of Integerrimine in *Buchenroedera* and *Lotononis* Section *Krebsia*

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Key Word Index—*Buchenroedera*; *Lebeckia*; *Lotononis*; Leguminosae; Crotalariaeae; pyrrolizidine alkaloid; integerrimine; chemotaxonomy; generic relationships.

Abstract—Integerrimine, a pyrrolizidine alkaloid previously known only from species of *Cacalia*, *Senecio* and *Crotalaria*, has been identified as the major alkaloid of several species of *Buchenroedera* and *Lotononis* section *Krebsia*. Chemotaxonomic implications are discussed. Our conclusions are that the genera *Buchenroedera* and *Lotononis* are more closely related to *Crotalaria* than to *Lebeckia*, despite morphological evidence to the contrary; that *Buchenroedera* and *Lotononis* may not be distinct at the generic level and that *Lebeckia microphylla* should be transferred to *Lotononis* section *Krebsia*.

Introduction

In a general survey of alkaloids in the tribe Crotalariaeae (Van Wyk, B-E., Verdoorn, G. H. and Schutte, A. L., *S. Afr. J. Botany*, in press) we have shown that useful chemotaxonomic information may be obtained. The pyrrolizidine alkaloids of the genus *Crotalaria* have been the subject of numerous studies, but the only previous reported occurrence of alkaloids from the remaining 16 genera was that of tetracyclic quinolizidine alkaloids in *Lebeckia plukenetiana* E. Mey. [1]. In view of suggestions that the genus *Buchenroedera* Eckl. & Zeyh. may be no more than a section of *Lotononis* (DC.) Eckl. & Zeyh. [2, 3], we investigated the major alkaloid of this genus and that of the closely related section *Krebsia* (Eckl. & Zeyh.) Benth. of *Lotononis*.

Results

The major alkaloid of *Buchenroedera lotonoides* Scott Elliot, previously referred to as alkaloid no. 26 (Van Wyk *et al.*, *op. cit.*), was isolated and identified as integerrimine, a pyrrolizidine alkaloid previously reported only from species of *Cacalia*, *Senecio* and *Crotalaria* [4, 5]. All the spectroscopic data we obtained was in correlation with that given for integerrimine in the literature [6-8]. A total yield of ca 140 µg/g

dry wt (0.014%) was obtained. Integerrimine was subsequently also identified by TLC and MS as the major alkaloid of all the species of *Buchenroedera* and *Lotononis* section *Krebsia* examined (Table 1). At least four other minor alkaloids were present in all of these species, but not in sufficient quantities to allow their identification. This is the first report of the presence of pyrrolizidine alkaloids in *Buchenroedera* and *Lotononis*.

Discussion

In the Leguminosae, pyrrolizidine alkaloids were at first thought to be limited to *Crotalaria*, but have subsequently also been found in the tribe Genisteae (*Adenocarpus*, *Cytisus* and *Laburnum*) [9, 10], where they may co-occur with quinolizidine alkaloids. The presence of a pyrrolizidine alkaloid in *Buchenroedera* and *Lotononis* would seem to erode the apparent isolation of *Crotalaria* within the tribe Crotalariaeae and indicates new possibilities for testing current tribal and generic concepts. A more detailed survey of *Lotononis* (a diverse genus of some 130 species) and related genera would therefore be most worthwhile. This may well lead to a better understanding of evolutionary relationships within a group that shows a high incidence of convergence and conflicting character information.

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TABLE 1. TAXA AND PLANT MATERIAL EXAMINED

Species	Locality	Date collected	Collector and No.
<i>Buchenroedera lotononoides</i> Scott Elliot	Sani Pass, Natal	April 1987	B-E. van Wyk 2630
<i>B. meyeri</i> Presl	Mhlahlane, Transkei	January 1986	B-E. van Wyk 1765
<i>B. multiflora</i> Eckl. & Zeyh.	Zuurberg, Eastern Cape	April 1986	B. & M. van Wyk 1523
<i>B. tenuifolia</i> Eckl. & Zeyh. var. <i>tenuifolia</i>	Penhoek Pass, Queenstown	December 1985	B-E. van Wyk 1593
var. <i>pulchella</i> (E. Mey.) Harv.	Craddock, Eastern Cape	July 1985	B-E. van Wyk 1334
<i>B. trichodes</i> Presl	Katberg Pass, Eastern Cape	January 1986	B-E. van Wyk 1693
<i>Lotononis caerulea</i> (E. Mey.) B-E. van Wyk <i>ined.</i>			
(= <i>Lebeckia microphylla</i> E. Mey.)	Nonesi's Nek, Queenstown	April 1987	B-E. van Wyk 1632
<i>L. divaricata</i> (Eckl. & Zeyh.) Benth.	Burgersdorp, Eastern Cape	January 1987	B-E. van Wyk 2597
	Rouxville, Orange Free State	April 1987	B-E. van Wyk 2629
<i>L. trisegmentata</i> Phill. var. <i>robusta</i> Phill. forma <i>robusta</i>	Clarens, Orange Free State	May 1987	B-E. van Wyk 1561
forma <i>sericea</i> Phill.	Sani Pass, Natal	April 1987	B-E. van Wyk 1968

The presence of integerrimine as a major alkaloid in extracts of all these species was confirmed by mass spectrometry. Voucher specimens are deposited in the Rand Afrikaans University Herbarium.

Some species of *Lotononis* are morphologically very similar to species of *Lebeckia* Thunb. This similarity has led to considerable taxonomic and nomenclatural confusion over the last two centuries. The alkaloidal metabolites however, now appear to be quite different. We suggest that the two genera are not closely related and that similarities may be ascribed to convergent evolution in response to the same environmental pressures, such as increasing summer drought during recent geological times. The phenomenon that leaflets are individually shed leaving the petioles, is a clear example of such an adaptation that occurs in both genera. The presence of integerrimine as a major alkaloid of *Lebeckia microphylla* supports the morphological evidence that was found for the transfer of this species to *Lotononis* (Van Wyk, B-E., *S. Afr. J. Botany*, in press).

The *Lotononis* species listed in Table 1 all belong to the section *Krebsia*, which is so closely related to *Buchenroedera* that the status of the latter has been questioned [2, 3]. Only a single diagnostic character (fruit shape) was found to separate the two genera and sectional rank for the latter within *Lotononis* was therefore suggested. This view is strongly supported by the evidence at hand.

Experimental

Plant materials. Collection details of all the species examined are listed in Table 1.

Procedures. Ground air-dried leaves and twigs were extracted with cold CH_2Cl_2 . Extracts were acidified with 2 N

HCl and re-extracted with CH_2Cl_2 (50 ml \times 3) after basifying the aqueous phase with conc. NH_3 . *Buchenroedera lotononoides* (930 g) yielded 600 mg of crude alkaloidal material after 2 days of extraction and an additional 300 mg after 4 more days. The first 600 mg portion was chromatographed on silica gel 60 using cyclohexane- CHCl_3 - Et_3N (20:25:6) as eluent. Fractions of 2 ml of each were collected. Fractions 28 to 64 with R_f 0.18 were combined and upon evaporation of the solvent gave 88 mg of a white crystalline solid. Colourless prisms obtained from CHCl_3 were used to determine the mp and specific rotation, as well as IR, ^1H NMR, ^{13}C NMR and MS spectra. Mixtures of all the other species were studied by TLC (silica gel: eluent cyclohexane- CHCl_3 - Et_3NH , 5:4:1, detecting reagent iodoplatinate) and the identity of integerrimine as the major alkaloid was confirmed by MS.

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