Alkaloid Composition of *Lupinus albescens* (Fabaceae) from South America

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Alkaloid extracts of leaves and seeds of the South American taxon *Lupinus albescens* Hooker & Arnott were analyzed by capillary gas-chromatography (GLC) and GLC-mass spectrometry. Multiflorine and albine figured as major, and ammodendrine, lupanine, 11,12-seco-12,13-didehydromultiflorine 13α-hydroxymultiflorine and 13-tigloyloxymultiflorine as minor alkaloids. Three new alkaloids were tentatively identified as a dehydromultiflorine, N-formylalbine and 13-methoxymultiflorine.

Introduction

Members of the genus *Lupinus*, of which more than 500 taxa are known, are distributed mainly in the New World (South and North America). Only 12 taxa occur in Europe and North Africa. It has been assumed that all lupin species produce quinolizidine alkaloids, but alkaloid contents and alkaloid profiles have been studied with modern techniques, such as GLC-MS, for a small limited number so far [1, 2]. The alkaloid profiles of South American lupins are especially interesting since it has been suggested that South America is the evolutionary origin of this genus. Except for *Lupinus mutabilis* [3] hardly anything is known about the majority of South American lupin taxa.

Lupinus albescens Hooker & Arnott, is native in Northeastern Argentina, Paraguay and Uruguay. It has been found frequently on sandy soils and dunes near the Paraná River and in its islands. *L. albescens* is a close relative of *L. aureonitens* Gilbert (occurring in South of the Buenos Aires Province in Argentina) and is moderately related to *L. multiflorus* Desr. from Uruguay. Planchuelo and Dunn [4] consider that these three species derived from *L. paraguariensis* Chodet & Hassler and the simple-leaved lupins from Brazil. Other relatives belong to the *L. lanatus* complex [5].

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The distribution of *L. albescens* and related species is restricted to the "Atlantic Subregion" if the whole area of speciation of the genus *Lupinus* is taken into account [6]. Neither the other lupin taxa growing in the same area and nor the ones of the "Andean Subregion" have morphological characteristics that suggest a close relationship with the *L. albescens* and its relatives.

The combination of high resolution gas liquid chromatography (GLC) and GLC-mass spectrometry (GLC-MS) is a powerful tool in the analysis of complex mixtures of quinolizidine alkaloids [2, 7–12].

In this study the alkaloid pattern of seeds and leaves of the South American *L. albescens* which had not been assessed before phytochemically was analyzed by capillary GLC and GLC-MS.

Materials and Methods

Samples of *L. albescens* were collected in January 1992, from an island of the Paraná River near the city of Santa Fe. Voucher specimens (No. Planchuelo 552) have been deposited in the Agricultural Science Herbarium of Córdoba National University of Argentina (AGROCOR).

Leaves and seeds were analyzed separately in order to compare the respective alkaloid profiles. Two samples, each of 2 g of leaves and 4.5 g of seeds, soaked in 0.5 M HCl overnight, were homogenized in 15 ml 0.5 M HCl. The homogenate was adjusted to pH 12 with ammonia (25%). Alkaloids

were extracted by solid liquid extraction using an Extrelut column and methylenechloride as an eluent. Alkaloid extracts were separated by high resolution gas liquid chromatography employing capillary columns (DB1 30 m × 0.3 mm; J&W Scientific) and a Carlo Erba instrument. GC-MS was performed as described in [10, 11].

Results and Discussion

The separation of alkaloid extracts from seeds and leaves of *L. albescens* by GLC is illustrated in Fig. 1. Both alkaloid profiles are very similar. GLC-MS analysis revealed the presence of at least 10 quinolizidine alkaloids which belong mostly to the multiflorine series. According to retention index and MS data from our laboratory [3, 7–12] and literature information [1, 2, 13–15], we were able to identify 7 alkaloids unambiguously: multiflorine as the main alkaloid, albine, 11,12-seco-12,13-didehydromultiflorine, and 13-tigloyloxymultiflorine as minor components, and only traces of 13α-hydroxymultiflorine.

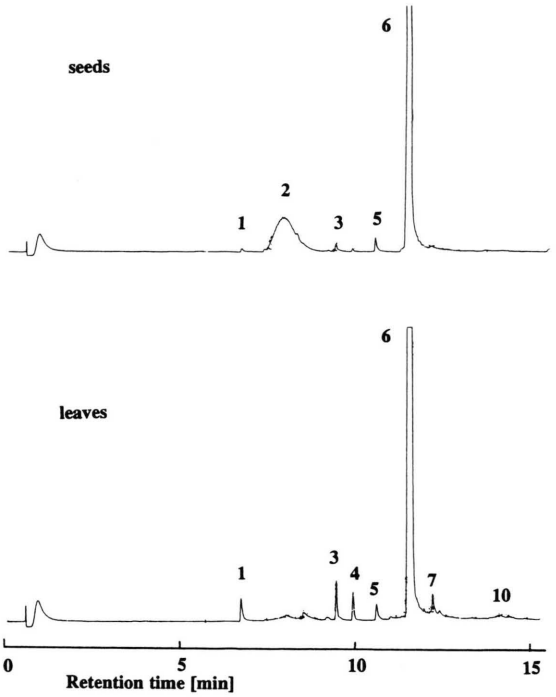

Fig. 1. Separation of quinolizidine alkaloids from *L. albescens* by capillary GLC. Conditions: injector: 250 °C, nitrogen-specific detector: 300 °C, oven: 150 °C, 1 min isothermal, then 15 °C/min to 300 °C. Numbering as in Table I.

Three new alkaloids have not been reported before which are identified tentatively according to their MS fragmentation patterns: RI 2110 shows a molecular ion at *m/z* 244 which could be expected for a "dehydromultiflorine". The position of the double bond is not clear but the MS spectrum differs significantly from that of 5,6-dehydromultiflorine [13]. RI 2543 has a base peak at *m/z* 219, thus 14 mass units higher than that of albine. In analogy to the fragmentation pattern of N-formylcytisine in the cytisine series [2], "N-formylalbine" could be a plausible candidate which would fit into the fragmentation pattern observed. RI 2840 must be a derivative of 13-hydroxymultiflorine according to the intensive fragment at *m/z* 244. In analogy to 13-methoxylupanine we conclude that this new alkaloids represents 13-methoxymultiflorine, since a methoxy group would explain the mass difference of 30 (between *m/z* 274 and *m/z* 244) (Table I). Because of shortage in material a further structure elucidation was not possible at this stage.

Table I. Identification of quinolizidine alkaloids from *L. albescens* by GLC and GLC-MS. RI, Kovats retention index; M^+, molecular ion.

Compound	RI	M^+	Characteristic ions (abundance %)				
1 Ammodendrine	1865	208	208 (65)	191 (45)	165 (100)	136 (70)	110 (60)
2 Albine	1900	232	232 (25)	191 (100)	149 (40)	122 (40)	110 (55)
3 "Dehydromultiflorine"*	2110	244	244 (100)	160 (20)	146 (20)	134 (75)	110 (20)
4 Lupanine	2165	248	248 (40)	149 (55)	136 (100)	110 (25)	98 (20)
5 11,12-Seco-12,13-didehydromultiflorine	2210	246	246 (10)	205 (90)	134 (10)	110 (20)	58 (100)
6 Multiflorine	2310	246	246 (55)	189 (10)	149 (20)	134 (100)	110 (25)
7 "N-Formylalbine"*	2543	260	260 (10)	219 (100)	207 (5)	148 (5)	96 (25)
8 13-Hydroxymultiflorine	2570	262	262 (35)	164 (10)	150 (100)	134 (10)	110 (15)
9 "13-Methoxymultiflorine"*	2840	274	274 (10)	244 (100)	162 (100)	134 (40)	96 (100)
10 13-Tigloyloxymultiflorine	2935	344	344 (10)	244 (70)	149 (50)	132 (100)	110 (40)

* New alkaloids; tentative identification.

Table II. Alkaloid composition and alkaloid content of *L. albescens*.

Alkaloid	Alkaloid composition (total = 100%) Seeds	Leaves
Ammodendrine	0.2	0.8
Albine	16.5	0.7
"Dehydromultiflorine"	0.4	2.4
Lupanine	0.02	0.2
11,12-Seco-12,13-didehydro-multiflorine	0.7	0.9
Multiflorine	81.0	92.3
"N-Formylalbine"	tr	tr
13-Hydroxymultiflorine	tr	tr
"13-Methoxymultiflorine"	tr	tr
13-Tigloyloxymultiflorine	0.6	2.3
Alkaloid content (mg/g dry weight)	2.6	6.1

Multiflorine is the major alkaloid, both in the seeds and leaves (Table II), representing 81 or 92% of total alkaloids, respectively. In seeds, albine is the second main alkaloid with 16.5% followed by 11,12-seco-12,13-didehydromultiflorine with only 0.7%. On the other hand, in leaves, albine is in the 6th place with 0.7%. Two other alkaloids are more pronounced in leaves than in seeds, such as "dehydromultiflorine" and 13-tigloyloxymultiflorine with 2.4 or 2.3% respectively. All other alkaloids figure as minor components in both seeds and leaves. The dominance of multiflorine and derivatives is quite rare among lupins. We had found similar profiles only in Old World lupins, such as *L. micranthus, L. palaestinus, L. atlanticus* and *L. pilosus* [8, 16] but not in New World species [1, 8, 10, 16].

Since we do not know the alkaloid profiles of other South American lupin taxa, it is too early to speculate on the evolutionary and systematic implications of our findings.

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