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FLAVONOIDS OF *LEPTARRHENA PYROLIFOLIA**

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Key Word Index—*Leptarrhena pyrolifolia*; Saxifragaceae; flavonols; (+)-dihydromyricetin; dihydroflavonol stereochemistry; chemotaxonomy.

Abstract—The flavonoids of *Leptarrhena pyrolifolia* comprise (+)-dihydromyricetin and mono-, di- and triglycosides of kaempferol, quercetin, isorhamnetin and myricetin. This is the first report of a dihydroflavonol in the Saxifragaceae.

INTRODUCTION

Leptarrhena R.Br. is a monotypic genus of the Saxifragaceae consisting of *L. pyrolifolia* (D.Don) R.Br. The plant is found along streams and in wet subalpine meadows from Alaska to Oregon and east to western Montana and Alberta [1]. Engler [2] placed *Leptarrhena* in the tribe Saxifrageae, subtribe Leptarrhenineae along with the monotypic Japanese genus *Tanakea*. In recent flavonoid investigations we have examined a number of genera which Engler [2] placed in subtribe Saxifraginae. Thus, the acquisition of several collections of *L. pyrolifolia* enabled us to determine its flavonoids and to compare these data with the known chemistry of other members of the Saxifraginae. Hegnauer [3] lists *Leptarrhena* as possessing procyanidin, prodelphinidin, myricetin, quercetin and ellagic acid.

RESULTS

The leaf flavonoid glycoside fraction of *Leptarrhena* was found to be a complex mixture of flavonol mono-, di- and triglycosides. The monoglycosides identified were the 3-*O*-glucosides, 3-*O*-galactosides, 3-*O*-arabinosides and 3-*O*-xylosides of kaempferol, quercetin, isorhamnetin and myricetin. Among the diglycosides were the 3-*O*-rhamnosylgalactosides and 3-*O*-rhamnosylglucosides (rutinosides) of kaempferol, quercetin and isorhamnetin and traces of quercetin and myricetin 3-*O*-diglucosides. The triglycoside fraction was not studied other than to note the presence of all four aglycones after hydrolysis.

Initial 2D-TLC showed the presence of a large, UV-absorbing spot with chromatographic behaviour similar to a flavonol monoglycoside in all of the leaf samples. The compound gave no colour with diphenylboric acid ethanolamine complex and exhibited a UV spectrum suggestive of a flavonoid with a reduced C-ring system [4]. It was identified as dihydromyricetin, by mp and UV spectral comparison with published data [5] and by dehydrogenation to give myricetin. The NMR spectrum was consistent with this structural assignment: δ 6.60 (*s*, H-2′, H-6′); δ 5.90 (slight fine structure on expansion) (H-6, H-8); δ 4.88 (*d*, J = 12 Hz, H-2); and δ 4.47 (*d*, J = 12 Hz, H-3). NMR assignments for its hexaacetate are as follows: δ 7.40 (*s*, H-2′, H-6′); 6.83 (*d*, J = 2 Hz, H-8); 6.63 (*d*, J = 2 Hz, H-6); 5.75 (*d*, J = 12 Hz, H-2); 5.59 (*d*, J = 12 Hz, H-3) and 2.22 (*s*, acetate methyl protons). The large coupling constants observed for H-2 and H-3 arise from their *trans*-diaxial disposition which is fully in accord with the diequatorial orientation of phenyl and hydroxyl substituents in dihydrokaempferol and dihydroquercetin [6]. Optical rotation data for the *Leptarrhena* dihydromyricetin hexaacetate gave $[\alpha]_D^{20}$ +43° in good agreement with the published value of +46° [5]. Since dihydroflavonols having positive rotations are considered to have 2*R*:3*R* stereochemistry [6], the natural compound from *Leptarrhena* must be (2*R*:3*R*)-dihydromyricetin.

DISCUSSION

Detailed studies of the flavonoid chemistry of members of Saxifragaceae subtribe Saxifraginae [7 and refs. cited therein] have revealed that the majority of taxa produce glycosides of the common flavonols: kaempferol, quercetin and myricetin, in considerable quantities. Flavones have been found in a few taxa and compounds having additional A- and/or B-ring hydroxylation are also known. *O*-Methylation is known in *Heuchera* [7, 8], but is a predominant structural feature only in *Chrysosplenium* [9].

In addition to the presence of dihydromyricetin, discussed below, *Leptarrhena* is characterized by the presence of glycosides of simple flavonols and the absence of flavones. While the substantial concentrations of kaempferol, quercetin and myricetin glycosides are expectable, *Leptarrhena* accumulated a larger amount and number of isorhamnetin derivatives than other genera, for example, *Heuchera* [7, 8]. The flavonols are present as mono-, di- and triglycosides with the sugar always linked at position 3, a pattern which is common in the family although other positions of glycosylation have been observed: 3,7- in *Heuchera* [7, 8], 4′- in *Tellima* [10] and 6-, 2′- or 4′- in *Chrysosplenium* [9]. Monoglycosides are based on glucose, galactose, arabinose and xylose, giving a combination of compounds similar to those in *Heuchera* [7, 8] and *Peltiphyllum* [11]. However, the diglycoside fraction is less complex than *Heuchera* [7, 8] having only diglucosides, rhamnosylgalactosides and rhamnosylglucosides.

Plant material for this study was obtained from seven populations representing a geographic separation of *ca* 250 km. Two dimensional chromatograms of these

* Part 13 in the series "Chemotaxonomic Studies in the Saxifragaceae s.l.". For Part 12 see Bohm, B. A. and Collins, F. W. (1979) *Biochem. Syst. Ecol.* 7, (in press).

collections were identical with respect to the flavonoid constituents. The apparent lack of variation of flavonoids as characters in *Leptarrhena* is in accord with the invariant flavonoid profiles seen in several other taxa studied in this series [12]. The major polyphenol in all populations of *Leptarrhena* studied is (+)-dihydromyricetin; this is its first reported occurrence in the Saxifragaceae.

EXPERIMENTAL

Plant material was obtained from the following: Canada: Mt. Baldy, B.C. (BAB-1273); Goldie Lake, Mt. Seymour Prov. Park, B.C. (BAB-1274); Strachan Meadow, Cypress Bowl Prov. Park, B.C., two specimens (BAB-1232 and BAB-1233); Yew Lake, Cypress Bowl Prov. Park (BAB-1159); Mt. Becher, Courtenay, Vancouver Island, B.C. (BAB-1153); and United States: Washington: Chelan Co., summit, North Cascades Highway (BAB-1103). Vouchers are deposited in UBC.

Isolation procedures and chromatographic procedures were described by Wilkins and Bohm [8]. NMR spectra were determined in Me_2CO-d_6 with TMS as internal standard. Optical rotation was measured in Me_2CO. Acetylation was carried out with Ac_2O/Py and the product recrystallized from MeOH.

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NEW PYRROLIZIDINE ALKALOIDS FROM *CROTALARIA CANDICANS**

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Key Word Index—*Crotalaria candicans*; Leguminosae; pyrrolizidine; crocandine; isocrocandine; turneforcidine; 3-hydroxy-2,3,4-trimethylglutaric acid.

Abstract—Crocandine and isocrocandine, isolated from the seeds of *C. candicans*, have been shown by spectroscopy and chemical evidence to be macrocyclic diesters of turneforcidine and 3-hydroxy-2,3,4-trimethylglutaric acids.

INTRODUCTION

Crotalaria candicans W. & A. 184 [1], which grows wild in Western ghats in the Nilgiris (India) at *ca* 2000 m, is characterized by broadly cordate, acute, persistent, shining viscous and black (when dry) bracts and bracteoles, considered to be synonymous with *C. madurensis* Wight in Wall and has been investigated for alkaloids. Isolated constituents have been characterized as two new macrocyclic diesters, named here as crocandine (CC-I) and isocrocandine (CC-II), which are different from the ones reported from *C. madurensis* [2–6]. These chemotaxonomical data are further justification for considering *C. candicans* a different species as proposed by W. & A. A survey of literature [7] shows that only two esters of turneforcidine, viz. turneforcine and retusine, have been described hitherto.

RESULTS AND DISCUSSION

Structure of crocandine (1)

The ^1H NMR spectrum ($CDCl_3$) of the alkaloid exhibits signals at δ 1.15 (*d*, J = 7 Hz, $C\underline{H}_3$—CH—COO), 1.23 (*d*, J = 7 Hz, $C\underline{H}_3$—CH—COO), 1.28 (*s*, $C\underline{H}_3$—C—OH), 3.2 (OH), 3.78 and 4.73 (H-9) and 5.05 (*m*, H-7). The unusual width of the H-7 multiplet, 24.0 cps, is characteristic of esters of hastanecine and turneforcidine

* Part 37 in the series "*Genus* Crotalaria". For Part 36 see Siddiqi, M. A., Suri, K. A., Suri, O. P. and Atal, C. K., Indian Pharmaceutical Congress (held at Calcutta, 26–28 December, 1978).

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