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## Flavonoid profiles of certain species of Rhynchosia of the family Leguminosae (Fabaceae)

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Summary. A survey of seventy naturally occurring populations of Rhynchosia representing seven species of different subgenera revealed the occurrence of C-glycosides, O-glycosides, prenylated flavonoids and aglycones. Rhynchosia albiflora (R. cyanosperma) differs in gross morphological features and seed coat color and in its flavonoid composition from the other representative species of Rhynchosia.

Key words. Rhynchosia gen.; flavonoids, leaf.

The genus *Rhynchosia* is widely distributed with several species occurring in mountainous regions in the tropics. The wide range of morphological features selected to classify the genus has often resulted in anomalies in the systematic position of certain species, as is evident in the regional floras of Hooker<sup>2</sup>, Cooke<sup>3</sup> and Duthie<sup>4</sup> and in the classification of Roxburgh<sup>5</sup> and Bentham and Hooker<sup>6</sup>. As described in the floras, the basic similarities of various species of *Rhynchosia* are, pinnate leaves, compressed pods and calyx not accrescent. The division of the genus *Rhynchosia* into various subgenera involves differences in enlargement of the fruiting calyx in size and breadth, and the presence or absence of the arillus on the seed.

Cylista tomentosa has been re-classified as a member of the genus Rhynchosia. It was originally called R. cyanosperma but is now called R. albiflora. We therefore considered it would be valuable to compare the flavonoid profiles of seven indigenous species of five subgenera (Phyllomatia, Eurhynchosia, Cyanospermum, Ptychocentrum and Nomismia), to see whether Rhynchosia albiflora would fit comfortably into the genus with regard to its flavonoid composition or whether it differs from the other representatives, as shown in morphological details. Though the members of Fabaceae have been widely surveyed for morphological and chemical characteristics, the details at the level of the species included in various genera of the family have not been adequately enough worked out to arrive at a logical phylogenetic arrangement of the species at generic level. Earlier studies have shown the occurrence of flavonoids in three different species of Rhynchosia7. The present survey encompasses three more species of Rhynchosia (R. heynei, R. capitata and R. sericea) and compares the results with those for R. albiflora (table).

Flavonoids which were reported in earlier studies and in three species in the present study are different from those of *R. albiflora*. A minimum of 9 populations of 7 species of *Rhynchosia* (80 g dry wt per population of each species) were examined for flavonoid constituents of the leaves. Isolation of flavonoids and their glycosides, hydrolytic procedures<sup>8</sup> and spectral analysis were carried out using standard methods<sup>9</sup>. Flavonoids were

exhaustively extracted using both acetone and methanol, reduced to dryness and loaded on column, paper and thin-layer chromatograms<sup>10</sup>. They were then reisolated and characterized using normal analytical procedures (UV, IR, NMR and MS)9. Sixteen different flavonoid compounds have been identified from the selected species (table). Of these, there are eight Cglycosides, three O-glycosides, two prenylated flavonoids (dihydroflavonol and the corresponding flavonol), a flavone, a flavanone and a 5-deoxyflavonol. The C-glycosides vitexin (8-C-β-D-glucopyranosyl apigenin), isovitexin (6-C-β-D-glucopyranosyl apigenin), orientin (8-C- $\beta$ -D-glucopyranosyl luteolin), isoorientin (6-C-β-D-glucopyranosyl luteolin) and vicenin-2 (6,8-di-C-β-D-glucopyranosyl apigenin) are invariably present in R.rufescens, R.heynei, R. capitata, R.beddomei, R.minima<sup>11,12</sup> and R. sericea, while lucenin-2 (6,8-di-C-β-D-glucopyranosyl luteolin) is common to R. rufescens, R. beddomei and R. minima. In the disputed species Rhynchosia albiflora none of the C-glycosides is present, instead, it was found to possess two O-glycosides namely rutin (quercetin-3-rutinoside) and kaempferol-3-rutinoside and two prenylated flavonoids namely tirumalin<sup>13</sup> (8-C-prenyltaxifolin 7,4'-dimethyl ether) and rhynchospermin<sup>14</sup> (8-C-prenylquercetin 7,4'-dimethyl ether), and is thus very different in its flavonoid composition. However, R. beddomei showed the occurrence of the maximum number of flavonoid constituents, which included apigenin, naringenin and rhynchosin (5-deoxyquercetagetin) and it is the only species which possessed O-glycosides close to those of R. albiflora. Rhynchosin<sup>15</sup>, a compound devoid of 5-hydroxyl groups occurring in R. beddomei, is considered to be an isolated charac $ter^{16}$ .

It is now clear that *R. albiflora* (Sims.) Alston, (*R. cyanosperma*) which has undergone a name change and a generic transfer from *Cylista tomentosa* Roxb., to *Rhynochosia* is quite distinct from other species of *Rhynchosia* not only in its general morphology and blue seeds but also in its flavonoid compounds. The other species of *Rhynchosia* which were studied here shared many morphological features as well as

Flavonoid constituents of Rhynchosia species (Leguminosae)

	C-glycosides								O-glycosides			Prenylated flavonoids			
	Orientin	Isoorientin	Vitexin	Isovitexin	Vicenin-2	Vicenin-3	Schaftoside	Lucenin-2	Rutin	Kaempferol 3-rutinoside	3',4'-di-O-methyl luteolin 7-glucuronide	Tirumalin	Rhynchospermin	Apigenin	Naringenin
Rhynchosia rufescens	+	+	+	+	+		_	+			_				
Rhynchosia heynei	+	+	+	+	+	_	_	-	_	****		_	_	_	_
Rhynchosia capitata	+	+	+	+	+		-		_	_	_	_	***	***	-
Rhynchosia beddomei	+	+ .	+	+	+	_	_	+	+		+	_	_	+	+
Rhynchosia minima	+	+	+	+	+	+	+	+	_			_	_	_	_
Rhynchosia sericea	+	+	+	+	+	_	_	-		-	_	_	_	_	_
Rhynchosia albiflora (R. cyanosperma)	_		_	_	_	-	-	sales and the sa	+	+	_	+	+	-	-

<sup>-,</sup> indicates absence; +, indicates presence and isolated.

having a similar flavonoid composition. The flavonoid composition even of R. minima, which occupies shady habitats and of R. beddomei, which is predominantly adapted to dry hilly tracts, is like that of the other species. The elaboration of the flavonoids in these two species might reflect biochemical adaptation to reflect the structural elaboration of the species in different environments.

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In conclusion it appears that a negative correlation exists between the leaf flavonoid profiles of R. albiflora and those of other species investigated. However, the situation with regard to morphological features and flavonoid content is so complex that a decision about whether this species fits into the genus Rhynchosia or not must await further information about other species.

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## Intracellular injection of cAMP and cGMP into snail neurones induces an increase in Na+conductance

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Summary. Injection of cAMP and more rarely cGMP into the neurones of the snail Helix pomatia induces an increase in membrane conductance, membrane depolarization and excitation. The effect is theophylline-dependent and has a reversal potential near -10mV.

Key words. Helix pomatia; snail neurones; Na+conductance; cAMP-injection; cGMP-injection.

Both an increase and a decrease of K+-channel conductance induced by intracellular cAMP injection into Aplysia and Helix neurones are well documented 1-6. However, studies of cAMP and cGMP influence on the Na<sup>+</sup>-conductance are rather contradictory<sup>7-9</sup>. The present study shows that pressure intracellular injections of cAMP and more rarely cGMP into

snail neurones induce membrane depolarization and a membrane conductance increase which is associated, at least partly, with an increase in Na+-conductance.

Methods. Identified neurones of the viscero-abdominal ganglionic mass of Helix pomatia (cells RPa<sub>1</sub>, RPa<sub>2</sub>, LPa<sub>2</sub>, and V<sub>6</sub> of Sakharov and Salanki<sup>10</sup>) were studied. The snail ganglia