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CHALCONE GLYCOSIDES OF *ANTIRRHINUM MAJUS*

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Key Word Index—*Antirrhinum majus*; Scrophulariaceae; chalcononaringenin 4′-glucoside; 3,4,2′,4′,6′-pentahydroxychalcone 4′-glucoside.

Abstract—Three chalcones have been found in yellow flowers of *A. majus*, two of which have been identified as chalcononaringenin 4′-glucoside and 3,4,2′,4′,6′-pentahydroxychalcone 4′-glucoside.

INTRODUCTION

THE NORMAL flower of the snapdragon exhibits a yellow palette spot on the lower lip of the mouth. This has been shown to be caused by the presence of two aurones, aureusin (aureusidin-6-glucoside) and bracteatin-6-glucoside.[1] These two aurones are present in all genotypes except the albino *niv/niv*; the mutation *eosinea*, when homozygous, removes all flavonoids with the 3,4-dihydroxy-grouping in the B-ring, replacing them with the corresponding 4-monohydroxyflavonoids, but in no way affects the aurones. Thus it may be argued that although the pathways of aurone and flavonoid biosynthesis share a common enzyme (the niv^+ gene product) at the first step, thereafter they diverge, sharing no common enzymes.

The mutation *sulfurea*, when homozygous, causes yellow pigment to be spread out so that it occurs in all cells of the front epidermis of the flower. It was decided to investigate the nature of this yellow pigmentation.

RESULTS AND DISCUSSION

Ethyl acetate extracts of flowers from plants with all homozygous combinations of *sulf* (C_1, C_2 and C_3 occur only in *sulf/sulf* niv^+/genotypes) with *eos* and *inc* (*incolorata*, when homozygous, removes anthocyanins and flavonols completely) were examined by two-dimensional TLC. Ethyl acetate was preferred as a neutral solvent which excludes anthocyanins. All chromatograms showed the presence of 3 spots, C_1, C_2 and C_3 having the colour reactions of chalcones, in addition to the well-known spots of flavones, flavonols, flavanones and aurones. C_1 was identified as the 4′-glucoside of chalcononaringenin and C_2 as the 4′-glucoside of 3,4,2′,4′,6′-pentahydroxychalcone. C_3 was extremely unstable and could not be prepared in sufficient quantity for analysis, but is most probably the chalcone corresponding to bracteatin-6-glucoside. This is the first reported occurrence of these compounds in nature.

These results explain the anomalous presence of naringenin 7-glucoside in *A. majus* as a result of acidic methanol extraction, C_1 cyclising to the corresponding flavanone. C_2 and C_3 presumably oxidize to the corresponding aurones under these conditions.

The fact that the chalcones occur as glucosides may indicate two possibilities: (1) that in normal biosynthesis of the aurones, glucosidation occurs at the chalcone stage, followed by cyclisation; or (2) that in *sulfurea* plants, where cells that do not usually form aurones

are constrained to do so, the enzyme system becomes 'overloaded', so that the 'aurone-6-glucosyltransferases' add glucose to chalcones without their being cyclised. Thus *sulf*, which appears to be a loss of control of the biosynthetic enzymes, allows the accumulation of intermediates, in much the same way as *Diluta* allows the accumulation of chrysanthemin[2] and pelargonidin 3-glucoside.[3]

TABLE 1. R_f (× 100) VALUES OF CHALCONE GLUCOSIDES

Solvent	C_1	C_2	C_3*	Solvent	C_1	C_2	C_3*
BAW	81	56	32	30% HOAc	44	27	21
15% HOAc	16	09	08	PhOH	66	40	17

* R_fs estimated from 2-D chromatograms of crude extracts.

EXPERIMENTAL

Whole flowers were ground in a mortar and pestle in EtOAc, spotted directly onto cellulose thin-layer plates (MN300HR) and run in two directions (1. BAW (6:1:2); 2. 15% HOAc; and 1. BAW; 2. PhOH). Flowers were also extracted (Soxhlet) in EtOAc, and pigments separated by PLC (MN300) (30% HOAc, BAW, 30% HOAc). R_fs are given in Table 1.

C_1. EtOH spectrum 233, 366 $\Delta\lambda$ ($AlCl_3$) + 36, $\Delta\lambda$ (EtONa) + 50, $\Delta\lambda$ (NaOAc–H_3BO_3) + 1 nm. The shift with EtONa also produced a large increase in intensity. Results are indicative of a free 2′-hydroxyl, free 4-hydroxyl and no *o*-dihydroxy B-ring grouping.[4] Hydrolysis yielded naringenin and glucose only. Acid cyclisation yielded naringenin 7-glucoside quantitatively.

C_2. EtOH spectrum 234, 376 $\Delta\lambda$ ($AlCl_3$) + 42, $\Delta\lambda$ (EtONa) + 51, $\Delta\lambda$ (NaOAc–H_3BO_3) + 32 nm. Conclusions as for C_1, but *o*-dihydroxy group present on B-ring. Acid hydrolysis yielded eriodictyol and glucose only, whereas acid cyclisation gave eriodictyol 7-glucoside. Treatment of C_2 with 6% H_2O_2 yielded aureusin. Identifications based on standard methods.[4,5]

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