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FLOWER FLAVONOIDS OF *OPUNTIA* SUBGENUS *CYLINDROPUNTIA**

W. D. CLARK, G. K. BROWN and R. L. MAYS

Department of Botany and Microbiology, Arizona State University, Tempe, AZ 85281, U.S.A.

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Key Word Index—*Opuntia*; Cactaceae; high-performance liquid chromatography; quercetin and kaempferol glycosides.

Abstract—Flavonoids were identified by PC and HPLC from the flowers of three cholla species and their known diploid and triploid hybrids. All the individuals examined produce quercetin 3-glucoside, quercetin 3-rutinoside and kaempferol 3-glucoside, which vary quantitatively among taxa.

INTRODUCTION

As part of an investigation of the structure of a hybrid swarm among *Opuntia fulgida* Engelmann, *O. spinosior* (Engelmann) Toumey and *O. acanthocarpa* Engelmann & Bigelow, we undertook the identification [1] of the flower flavonoids from these species and their known diploid and triploid hybrids. The flavonoid extract from each individual was also analyzed quantitatively by HPLC. Prior to this report, the only complete studies of *Opuntia* flavonoids have been limited to species of the subgenus *Opuntia* [2, 3].

RESULTS AND DISCUSSION

The flowers of all taxa examined in this study produce the same common flavonoids: quercetin 3-rutinoside (**1**), quercetin 3-glucoside (**2**) and kaempferol 3-glucoside (**3**). However, these compounds vary quantitatively as determined by peak heights of HPL-chromatograms (Table 1). Each diploid species and color form exhibits a distinct quantitative profile, except *O. acanthocarpa*, which is very similar to the purple-flowered *O. spinosior*. The relative per cent flavonoid compositions in the diploid hybrids do not show intermediacy between parents, although each hybrid is morphologically intermediate [4]. On the other hand, the chemical profiles of the triploid hybrids are intermediate between each parent and, moreover, tend toward the relative flavonoid quantities found in the respective donors of the double genomes (i.e. *O. fulgida* in FFS and *O. spinosior* in FSS). Quantitative trends in the triploids are consistent with expected dosages from each genome, but the apparently transgressive variation in the diploid hybrids cannot as yet be explained.

Table 1. Relative percent composition of flavonoids in *Opuntia* species and hybrids

Genome designation*	Voucher number†	% Composition‡		
		Q-3-R	Q-3-G	K-3-G
SySy	P13815	15	63	22
SpSp	M2446	11	83	6
FF	P13819	40	30	30
AA	P13810	11	80	10
FS	P13809	44	24	32
FS	P13809A	3	94	3
AS	M2451	2	93	5
FFS	M1363	24	40	36
FFS	P13804	22	56	22
FSS	P13808	17	61	22

* SySy and SpSp = *O. spinosior*, yellow and purple flowers, respectively; FF = *O. fulgida*; AA = *O. acanthocarpa*; FS and AS = diploid hybrids; FFS and FSS = triploid hybrids. All genome designations are based on morphological and cytological evidence (see ref. [4]).

† P = collections by D. J. Pinkava; M = collections by L. A. McGill.

‡ Q-3-R = quercetin-3-rutinoside; Q-3-G = quercetin-3-glucoside; K-3-G = kaempferol-3-glucoside.

EXPERIMENTAL

Voucher specimens of all plant material are deposited in ASU. The locality for all collections is in the vicinity east of Florence, Arizona.

Flower flavonoids were isolated from 85% aq. MeOH extracts by standard preparative two-dimensional PC and characterized by UV spectral analyses [1]. Flavonoid glycosides were oxidized [5] to yield sugars, which were identified by HPLC [6]. Crude 85% aq. MeOH extracts of petals from each individual were filtered through a Fluoropore filter (Millipore Corp., Bedford, MA) of 0.5 μm pore size and a μ Bondapak C_{18} Sep-Pak cartridge (Waters Associates, Milford, MA) to remove fine particulates and nonpolar contaminants, respectively. These solutions were then applied to HPLC. The equipment used was a Waters Model 244 Liquid Chromatograph equipped with a gradient elution system

* Part 2 in the series "The Flavonoids of the Cactaceae". For Part 1 see ref. [2].

and a μ Bondapak C_{18} column (30 cm × 3.9 mm i.d.), including a pre-column filled with C_{18} /Corasil. Solvent A was H_2O–HOAc (97.5:2.5) and solvent B was MeOH–HOAc (97.5:2.5). The mixture at injection was 90:10 (A:B), which was programmed to 50:50 through a linear gradient (solvent program #6) over 1 hr. Detection was at 365 nm. The flavonoids eluted in 42.0 min (**1**), 43.2 min (**2**) and 49.6 min (**3**) at 1.5 ml/min. Relative concentrations were calculated based on peak heights.

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DEUX FLAVONES EXTRAITES D'UN EXSUDAT DE FOUGÈRE (*PITYROGRAMMA CALOMELANOS* VAR. *AUREOFLAVA*)

JEAN FAVRE-BONVIN, MAURICE JAY, ECKHARD WOLLENWEBER* et VOLKER H. DIETZ*

Département de Biologie Végétale, Service de Phytochimie, Université Claude Bernard Lyon-I, 43 Boulevard du 11 Novembre, 69622 Villeurbanne, France; * Institut für Botanik, Technischen Hochschule, Schnittspahnstr. 3, 6100 Darmstadt, West Germany

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Key Word Index—*Pityrogramma calomelanos*; Polypodiaceae; farinose exudate; flavonoids; β-(5,7,4′-trihydroxy-flavon-8-yl)-β-phenylpropionic acid; methyl ester.

INTRODUCTION

Les frondes de *Pityrogramma calomelanos* var. *aureoflava* (Hook.) Domin produisent un exsudat farineux de couleur jaune. Une précédente étude a permis d'y déceler la présence de deux flavonoides majeurs: dihydroxy-2′,6′ diméthoxy-4,4′ chalcone, et son homologue dihydrochalcone [1]; plus récemment [2], l'existence d'un nouveau couple chalcone–dihydrochalcone a été rapportée, avec une substitution du type: dihydroxy-2′,6′ méthoxy-4′. Nous avons aujourd'hui isolé de ce même matériel deux nouveaux aglycones flavoniques, **1** et **2**, dont l'analyse structurale apporte la preuve de leur caractère original.

RÉSULTATS ET DISCUSSION

Les propriétés spectrales UV de **1** et de **2** orientent vers une structure flavone à noyau B monosubstitué; l'emploi des réactifs classiques [3] permet en outre de placer des hydroxyles libres en -5 (comportement avec $AlCl_3$ et fluorescence) et en -4′ (comportement avec NaOH). En présence de NaOAc, deux comportements différents sont enregistrés: pour **1** un déplacement bathochrome de 12 nm sur BII (= OH-7), pour **2** pas de déplacement significatif de BII mais l'apparition d'une déformation rendant cette bande très dissymétrique.

La SM réalisée sur les produits naturels révèle dans les deux cas un pic de plus haute masse à *m/e* 400 correspondant à la formule brute $C_{24}H_{16}O_6$; les ions-fragments les plus importants sont M − 43 et surtout M − 77 signifiant la perte d'un noyau phényle. Bien que différents chromatographiquement, ces deux produits paraissent donc construits sur le même thème de base $C_{15}C_6$ (et probablement) C_3, sans qu'il soit possible pour l'instant d'aller plus avant dans l'enchaînement de ces trois maillons.

Notre attention portera tout d'abord sur **1**; le cas de **2** sera envisagé ultérieurement et par différence à cette première analyse structurale.

Le dérivé TMSi de **1** montre un pic moléculaire *m/e* 648 (432 + 3 TMSi) qui conduit à reconnaitre pour la génine initiale une masse moléculaire égale a 432, c'est à dire supérieure de 32 uma à l'ion de plus haute masse détecté en SM du produit naturel. La réalité du pic moléculaire *m/e* 432 pour **1** est encore démontrée par SM en CI/D (NH_3 gaz reactant) où est révélé un ion M + H *m/e* 433, suivi d'un ion M − 32 + H *m/e* 401; en d'autres termes, tout porte à penser qu'en SM du produit naturel, il y a, avant ionisation, perte d'une molécule de MeOH.