Phytochemistry, 1978, Vol. 17, pp. 729–734. Pergamon Press Printed in England.

THE SYSTEMATIC IMPLICATIONS OF THE COMPLEXITY OF LEAF FLAVONOIDS IN THE BROMELIACEAE

CHRISTINE A. WILLIAMS

Botany Department, Plant Science Laboratories, The University, Whiteknights, Reading, Berks., RG6 2AS, England

(*Received* 30 *November* 1977)

Key Word Index—Bromeliaceae; 6-hydroxyflavones; gossypetin; patuletin; methylated 6-hydroxymyricetin derivatives; 6,3′,5′-trimethoxy-3,5,7,4′-tetrahydroxyflavone 3-glucoside; patuletin 3-rhamnoside; biochemical systematics.

Abstract—In a leaf survey of 61 species of the Bromeliaceae, an unexpectedly wide spectrum of flavonoid constituents was encountered. The family is unique amongst the monocotyledons in the frequency and variety of flavonoids with extra hydroxylation or methoxylation at the 6-position. More common flavonols (in 43% of species) and flavones (in 13%) are distributed throughout the family whereas the rarer flavonoid classes are restricted to one or two of the three subfamilies. Thus 6-hydroxyflavones were found in both the Pitcairnioideae (in 50%) and the Tillandsioideae (in 14%) but patuletin (in 19%), gossypetin (in 1 species) and methylated 6-hydroxymyricetin derivatives (in 24%) were detected only in the Tillandsioideae. A new flavonol, 6,3′,5′-trimethoxy-3,5,7,4′-tetrahydroxyflavone, was identified as the 3-glucoside in *Tillandsia usneoides* and a new glycoside, patuletin 3-rhamnoside, in *Vriesea regina*. Myricetin glycosides were found only in the Bromelioideae and their presence here and the concomitant absence of 6-hydroxyflavonoids could indicate the primitive condition of this subfamily. The flavonoid results, *in toto*, confirm the view based on morphology, that the Bromeliaceae occupies an isolated position in relation to other monocot families.

INTRODUCTION

The Bromeliaceae is a large, predominantly tropical family with some 45 genera and 1900 taxa, all restricted to the New World except for one West African *Pitcairnia* species. The family is both morphologically and anatomically [1] distinct and is treated by most taxonomists as an isolated group within the monocotyledons, which alone comprises the Order Bromeliales [2–4].

The present flavonoid study follows a recent investigation of another isolated order, the Zingiberales [5], and forms part of a larger flavonoid survey of monocotyledonous families [6–15], which is now in its final stages. The results are arranged according to the system of Mez [16], which is one of the most recent comprehensive treatments of the Bromeliaceae.

Probably, because it is not of any commercial importance and of the inaccessibility of many of its species, the Bromeliaceae has rarely been screened for its chemical constituents (cf. Hegnauer [17]). Bate-Smith [18] examined 14 taxa, in his leaf survey of the monocots, and was unable to identify any flavonoids, recording only the presence of the four common hydroxycinnamic acids. A more detailed study of the stem tissue of the pineapple, *Ananas comosus* var. *cayenne* has shown some unusual combined forms of these cinnamic acids: a quinyl-di-*p*-coumarate [19], which acts as a modifier of pineapple IAA oxidase [20, 21] and glyceryl esters of both caffeic and *p*-coumaric acid, which are known antioxidants [22]. More recently a new 6-hydroxyflavonol, 3,6,3′,5′-tetramethoxy-5,7,4′-trihydroxyflavone has been characterised from *Tillandsia usneoides* [23], commonly called Spanish moss.

RESULTS

The results of the leaf flavonoid survey are given in Table 1. Fresh leaf material was collected from plants growing at the Royal Botanic Gardens, Kew. The data in Table 1 refer to flavonoid aglycones detected in leaf tissue after acid hydrolysis. The aglycones were identified by means of R_f values and colour reactions in UV light, when compared with standard markers. The results of the aglycone survey were confirmed by 2D PC of direct leaf extracts and by characterisation of flavonoid glycosides in 14 species (Table 2). Flavone *C*-glycosides were confirmed by their resistance to 4 hr acid hydrolysis.

In the course of the survey a new 6-hydroxymyricetin derivative was discovered in stem and leaf tissue of *Tillandsia usneoides*: 6,3′,5′-trimethoxy-3,5,7,4′-tetrahydroxyflavone as the 3-glucoside. Its structure was confirmed by comparison of MS, UV spectra and R_f data with an authentic sample of the closely related 3,6,3′,5′-tetramethoxy-5,7,4′-trihydroxyflavone recently characterised from the same plant [23], which was also found in the free state and as the 7-glucoside in the present investigation. A further 16 *Tillandsia* species were screened for the new flavonol, but its presence was confirmed in only one other taxon, *T. fasciculata*. However, in *T. caput-medusae*, *T. streptocarpa* and *T. tricolor*, another possible highly methylated 6-hydroxyflavonol was detected, which was not present in sufficient amount to allow full characterisation.

Another methylated flavonol, patuletin (6-methoxyquercetin), was identified as the 3-rhamnoside and the 3-glucoside in *Vriesia regina* and as the 3-glucoside in *Tillandsia bulbosa* (see Table 2). This represents the first

Table 1. The distribution of flavonoids in leaves of the Bromeliaceae

Subfamily,* genus, species	Flavonols		Flavones			Accession number†
	common	6- or 8-hydroxy	common	6-hydroxy-	C-glycosides	
Bromelioideae						
Acanthostachys strobilacea (Schult. f.) Klotzsch	Qu	–	–	–	–	118-58.11804
Aechmea bromeliifolia (Rudge) Baker	Qu	–	–	–	–	059-74.00733
Ananas ananassoides (Baker) L. B. Smith var. *ananassoides*	–	–	–	–	–	331-68.3315
Araeococcus parviflorus (Mart.) Lindm.	–	–	–	–	–	131-74.015
Billbergia zebrina (Herb.) Lindl.	Qu	–	Lu	–	–	237-66.23704
Bromelia karatas L.	–	–	–	–	–	321-64.32101
Canistrum lindenii (Regel) Mez var. *lindenii*	Qu	–	–	–	–	060-71.00768
Gravisia aquilega (Salisb.) Mez	Qu	–	–	–	–	250-71.02283
Hohenbergia penduliflora (A. Rich.) Mez	Qu Isorh?	–	–	–	–	152-73.0-1467
Neoglaziovia variegata (Arr. da Cam.) Mez	Qu?	–	–	–	–	087-72.00655
Neoregalia spectabilis (Moore) L. B. Smith	–	–	Lu	–	–	000-69.50932
Nidularium billbergioides (Schult. f.) L. B. Smith	–	–	–	–	–	444-72.04020
N. fulgens Lem.	Qu	–	–	–	–	000-73.13310
N. innocentii Lem. var. *innocentii*	Qu My, Isorh	–	–	–	–	000-69.50934
N. innocentii Lem. var. *lineatum* (Mez) L. B. Smith	–	–	–	–	–	197-73.01785
N. procerum Lindm. var. *procerum*	Qu	–	–	–	–	703-62.70303
N aff *rutilans* E. Morr.	Qu	–	–	–	–	552-66.55214
Portea kermesiana K. Koch	–	–	–	–	–	000-69.50935
P petropolitana (Wawra) Mez	Qu My	–	–	–	–	518-67.51804
Quesnelia liboniana (De Jonghe) Mez	Qu	–	–	–	–	406-66.40609
Q. testudo Lindm.	–	–	–	–	–	552-66.55219
Streptocalyx longifolius (Rudge) Baker	Qu					386-71.03774
Pitcairnioideae						
Abromeitiella brevifolia (Griseb.) Castell	–	–	–	–	–	753-63.75312
Brocchinia reducta Baker	Qu	–	–	–	–	406-71.03933
Deuterocohnia longipetala (Baker) Mez	–	–	–	–	–	362-66.36218
Dyckia brevifolia Baker	–	–	–	–	–	362-66.36222
Hechtia argentea Baker ex Hemsl.	–	–	–	–	–	000-69.12356
Pitcairnia albucifolia Schrad.	–	–	–	–	–	000-69.51146
P. atrorubens (Beer) Baker	Qu	–	–	–	–	395.54
P. corallina Linden et André	–	–	Ap	6OH Lu Scut	–	000-73.13633
P. darblayana André	–	–	Ap	6OH Lu Scut	–	000-73.13317
P. imbricata (Brongn.) Regel	–	–	–	6OH Lu	–	152-73.01479
P. integrifolia Ker Gawl	Qu	–	–	6OH Lu	–	407-74.03199
P. poortmanii André	–	–	–	6OH Lu Scut	–	000-69.12852
P. punicea Scheidw	Qu	–	–	Scut	–	361-72.03429
P. 'rubiflora'	Qu Km	–	Ap	6OH Lu	–	000-73.13320
P. sprucei Baker	Qu Km	–	–	6OH Lu	–	393-74.03084
P. uaupensis Baker	–	–	–	–	–	518-67
P xanthocalyx Mart.	–	–	Lu	6OH Lu Scut?	+	248-65.24804
Puya sp.	–	–	–	6OH Lu + 2 methylated derivs.	–	085-74.01022
Tillandsioideae						
Catopsis berteroniana (Schult. fil,) Mez	Qu	Goss	–	–	–	235-75.02159
Guzmania lingulata (L.) Mez§	–	–	–	–	–	281-73 02738

Table 1 – *continued*

Subfamily,* genus, species	Flavonols		Flavones			Accession number†
	common	6- or 8-hydroxy	common	6-hydroxy-	*C*-glycosides	
Tillandsia anceps Lodd.	–	–	–	–	+	235-75.02147
T araujei Mez	–	–	–	–	–	196-73.01727
T. brachycaulos Schlechtend.	Qu?	Pat?	–	–	–	720-64.72003
T. bulbosa Hooker	–	Pat	–	–	–	466-69.03807
T. caput-medusae E. Morr.	–	–‡	–	–	–	347-61.34703
T. cyanea Linden ex K. Koch var. *cyanea*	–	–	–	6OH Lu	–	061-74.00900
T. aeranthos (Loisel) L. B. Smith	Qu	–	–	–	+	196-73.01724
T. fasciculata Swartz	Qu	6,3′,5′-triOMe-3,5,7,4′ tetraOH flavone	–	–	–	062-73.00394
T. juncea (Ruiz et Pav.) Poir	–	–	–	–	–	473-69.03885
T. lindeniana Regel	Qu	–	Lu	6OH Lu	–	406-66.40605
T. morreniana Regel	–	–	Lu	6OH Lu	–	516-58.51601
T. recurvata (L.) L.	–	–	–	–	+	087-72.00666
T. streptocarpa Baker	–	–	–	–	+	100-74.01083
T. streptophylla Scheidw.	–	–‡	–	–	–	196-73.01758
T. tricolor Cham. et Schlectend.	–	–‡	–	–	–	000-69.51149
T. triglochinoides Presl.	–	–	–	–	+	196-73.017600
T. usneoides (L.) L.	Qu	3,6,3′,5′ tetraOMe-5,7,4′-triOHflavone 6,3′,5′-triOMe-3,5,7,4′-tetraOHflavone	–	–	–	053-75.00674
Vriesea imperialis E. Morr.	Qu	Pat	–	–	–	118-58.11803
V. regina Beer	Qu	Pat	–	–	–	655-68.00268

* Classification according to Mez (1965). † Fresh plant material collected from the Royal Botanic Gardens, Kew. ‡ These plants contained possible methylated derivatives of 6-hydroxyflavonols. § This plant contained two unidentified dark to dark (in UV + NH_3) aglycones.

Key: Km = kaempferol, Qu = quercetin, My = myricetin, Isorh = isorhamnetin, Pat = patuletin, Goss = gossypetin, Ap = apigenin, Lu = luteolin, 6OH Lu = 6-hydroxyluteolin, Scut = scutellarein.

report of patuletin 3-rhamnoside in plant tissue. Patuletin was also detected in *V. imperialis* and *T. brachycaulos* and gossypetin, 8-hydroxyquercetin, in *Catopsis berteroniana*, another member of the subfamily Tillandsioideae.

Neither 6-hydroxy- nor 8-hydroxyflavonols were found in species of the other two subfamilies recognised by Mez [16]: the Bromelioideae and the Pitcairnioideae. However, 6-hydroxyflavones were detected in both the Pitcairnioideae (in 6 *Pitcairnia* and 1 *Puya* species) and the Tillandsioideae (in 3 *Tillandsia* species). 6-Hydroxyluteolin was identified as the 7-glucoside and 7-rutinoside in *P. poortmanii* and as the 7-glucoside in five other *Pitcairnia* species. Scutellarein (6-hydroxyapigenin) was also present as the 7-rutinoside in *P. poortmanii* and the aglycone was detected in hydrolysed extracts of four other *Pitcairnia* species, where except in *P. punicea* it always co-occurred with 6-hydroxyluteolin. From one *Puya* species, in addition to 6-hydroxyluteolin 7-glucoside, two partially characterised methylated derivatives were isolated and provisionally identified as 6,7-dimethoxy-5,3′,4′-trihydroxyflavone 4′-glucoside and 6,7,3′-trimethoxy-5,4′-dihydroxyflavone 4′-glucoside (see Experimental).

Quercetin (in 46%) was the most common leaf constituent and was identified as the 3-rutinoside or the 3-glucoside in species from all three subfamilies (see Table 2). Other common flavonols, myricetin (in 3%) and isorhamnetin (in 3%) occurred only in the Bromelioideae; myricetin was identified as the 3-galactoside and 3-glucoside in *Portea petropolitana*. Kaempferol was found as the 3-rutinoside in only two taxa, *Pitcairnia rubiflora* and *P. sprucei*.

Flavone *C*-glycosides (in 10%) are rare constituents confined to the sub-families Pitcairnioideae and Tillandsioideae. They were found with 6-hydroxyflavones and luteolin in *Pitcairnia xanthocalyx* and alone in five *Tillandsia* species. Common flavones, apigenin (in 5%) and luteolin (in 5%), are also minor flavonoid components in leaf tissue. However, luteolin glycosides were found in species representing all three subfamilies. Luteolin 7-glucoside was characterised in *P. rubiflora* and the 7-rutinoside in *P. xanthocalyx*. Apigenin derivatives, on the other hand, are restricted in their distribution to three *Pitcairnia* species; the 7-glucoside was identified in *P. rubiflora* and a 7-diglucuronide isolated from *P. corallina*. Common flavones were always found in association with 6-hydroxyflavones in *Pitcairnia* species.

In all three subfamilies, some species were found to be depauperate in flavonoids although cinnamic acid derivatives were present in considerable amount. These plants were re-examined at least once but with the same result. It could be that unnatural growing conditions of glasshouses at Kew may have affected the flavonoid/cinnamic acid balance in these plants and it would be valuable to repeat the survey of these species with plant material from natural stands.

DISCUSSION

Although most major flavonoid classes are represented in the Bromeliaceae, the most distinctive constituents

Table 2. Flavonoid glycosides identified in the leaves of some Bromeliaceae species

Species	Flavonoid glycosides identified
Canistrum lindenii	Quercetin 3-rutinoside
Catopsis berteroniana	Gossypetin glucoside*
Pitcairnia corallina	6-Hydroxyluteolin 7-glucoside, apigenin 7-diglucuronide
P. integrifolia	6-Hydroxyluteolin 7-glucoside, quercetin diglucoside*, apigenin *C*-glycoside*
P. poortmanii	6-Hydroxyluteolin 7-glucoside and 7-rutinoside, scutellarein 7-rutinoside
P. rubiflora	6-Hydroxyluteolin, luteolin and apigenin 7-glucosides, quercetin 3-glucoside and 3-rutinoside, kaempferol 3-rutinoside
P sprucei	6-Hydroxyluteolin and luteolin 7-glucosides, quercetin 3-glucoside and 3-rutinoside, kaempferol 3-rutinoside
P. xanthocalyx	6-Hydroxyluteolin 7-glucoside, luteolin 7-rutinoside, two apigenin 6,8-di-*C*-glycosides*
Puya sp.	6-Hydroxyluteolin 7-glucoside, quercetin 3-rutinoside, 6,7-dimethoxy-5,3′,4′-trihydroxyflavone and 6,7,3′-trimethoxy-5,4′-dihydroxyflavone 4′-glucosides
Portea petropolitana	Myricetin 3-galactoside and 3-glucoside, quercetin 3-glucoside
Tillandsia bulbosa	Patuletin 3-glucoside
T fasciculata	6,3′,5′-trimethoxy-3,5,7,4′-tetrahydroxyflavone 3-glucoside
T. usneoides	Quercetin 3-rutinoside, 6,3′,5′-trimethoxy-3,5,7,4′-tetrahydroxyflavone3-glucoside, 3,6,3′,5′-tetramethoxy-5,7,4′-trihydroxyflavone 7-glucoside
Vriesea regina	Quercetin and patuletin 3-glucosides, patuletin 3-rhamnoside

* These compounds were not completely characterised.

are undoubtedly those flavonoids with hydroxylation or methoxylation at the 6- or 8-position, i.e. scutellarein, 6-hydroxyluteolin, gossypetin, patuletin and methylated 6-hydroxymyricetin derivatives. Indeed, in the predominance and variety of these compounds the Bromeliaceae is unique amongst the monocotyledons. Similar constituents have been reported previously from isolated genera of only four other monocot families: 6-hydroxyluteolin from *Lagenocarpus* (Cyperaceae) [15] and *Tradescantia* (Commelinaceae) [24], gossypetin from *Restio* and hypolaetin from *Hypolaena* (Restionaceae) [25] and quercetagetin from *Eriocaulon* (Eriocaulaceae) [7]. The Bromeliaceae has never been considered to be closely allied with the Cyperaceae, Restionaceae or Eriocaulaceae but a number of taxonomists [2–4] have suggested some association with the Commelinaceae. However, as the main flavonoid components of the Commelinaceae [24], flavone *C*-glycosides, are rare constituents of the Bromeliaceae, a close alliance between the two families seems improbable. A preliminary study of anthocyanin pigmentation in leaves of the family [26] indicates again structural complexity. Methylated pigments i.e. peonidin and malvidin predominate, together with pelargonidin, and these are probably present as 3,5-diglucoside and/or 3-rutinoside-5-glucoside. The pattern is clearly different from that in the Commelinaceae, which have delphinidin and cyanidin 3-monoglucosides in leaf tissue [26]. Some workers [3] have suggested a possible association of the Bromeliales with another isolated order, the Zingiberales, but apart from the occurrence of myricetin glycosides in both there is no strong chemical evidence to support this view. The Bromeliales are probably still best considered as an isolated order, in which 6-hydroxylation and methoxylation have evolved independently of other monocot groups.

Table 3 The distribution of flavonoids at subfamily level in the Bromeliaceae

Subfamily*	Bromelioideae	Pitcairnioideae	Tillandsioideae
Common flavonols	+	+	+
Common flavones	–	+	+
Flavone *C*-glycosides	–	+	+
6-Hydroxyflavones	–	+	+
Gossypetin or patuletin	–	–	+
Methylated 6-hydroxymyricetin derivatives	–	–	+

* Classification according to Mez (1965).

The present results appear to be most meaningful at the subfamily level (see Table 3). From this table, it may be seen that the distribution of the different flavonoid classes may be used to distinguish the three subfamilies. Thus, in the Bromelioideae common flavonols and flavones are the only flavonoid constituents, in the Pitcairnioideae common flavonols and flavones, flavone *C*-glycosides and 6-hydroxyflavones are all represented whiles in the Tiallandsioideae gossypetin, patuletin and methylated 6-hydroxymyricetin derivatives also occur. Chemically, 6- or 8-hydroxy- or methoxyflavonoids are considered as advanced characters in that at least one extra biosynthetic step is needed for synthesis from the parent compound. Thus from the chemical data the Bromelioideae appear as the most primitive group with increasing complexity through the Pitcairnioideae to the most advanced Tillandsioideae. The occurrence of myricetin derivatives in two members of the Bromelioideae further support the chemically primitive condition of this subfamily. Taxonomically and anatomically [1], the Tillandsioideae is regarded as the most advanced group in the Bromeliaceae but there is still uncertainty which is the most primitive. Cronquist [3] considers the Pitcairnioideae to be the least advanced subfamily because most of its species are terrestrial zerophytes in contrast to the high proportion of epiphytic species in the other two subfamilies. However, anatomically [1] the Bromelioideae and Pitcairnioideae are hardly distinguishable apart from those characters which reflect the epiphytic nature of the former so that the chemical evidence could be regarded as decisive in this case.

A number of genera have distinct flavonoid profiles, e.g. *Pitcairnia* and *Tillandsia*, but even in the small sample of species available of these large genera there is obvious inter-specific variation. Thus *Pitcairnia* species may be divided into those taxa with quercetin alone (2 species), with 6-hydroxyflavones alone (5 species), with quercetin and 6-hydroxyflavones (4 species), with flavones and 6-hydroxyflavones (2 species) or all three flavonoid classes present (1 species). Similarly, *Tillandsia* species

show every variation from no flavonoid constituents to methylated 6-hydroxymyricetin derivatives. However it is not possible to comment on subgeneric limits with the present small sample.

EXPERIMENTAL

Plant material. Verified fr. plant material was received from the Royal Botanic Gardens, Kew and accession numbers are given in Table 1.

Identification of flavonoids. Flavonoid aglycones were identified in acid hydrolysed leaf extracts using standard procedures and by comparison with authentic markers. Direct 80% methanolic extracts of leaf tissue were chromatographed two-dimensionally in BAW and 15% HOAc. Known glycosides, isolated and purified by standard procedures were identified on the basis of R_f, UV spectral analysis, acid hydrolysis to aglycone and sugar and by direct comparison with authentic markers. Flavone *C*-glycosides were confirmed by 4 hr acid treatment, extraction into amyl alcohol and PC against authentic markers in BAW and H_2O.

Identification of 6,3′,5′-trimethoxy-3,5,7,4′-tetrahydroxyflavone-3-glucoside from Tillandsia usneoides. The glycoside was isolated from an 80% methanolic leaf and stem extract by prep PC in 15% HOAc, BAW and H_2O. R_f data are given in Table 4. Acid hydrolysis with 2N HCl for 40 min gave glucose and a new yellow (in UV) aglycone. λ_{max} for the glycoside are: MeOH 253, 267, 356, + NaOAc 273, 382, + H_3BO_3 253, 269′, 360, + alk 264, 340, 430 nm. A positive NaOAc shift indicates that the 7-position is free and the colour change, from a glycoside which is dark to yellow in UV + NH_3 to an aglycone which is yellow in UV without fuming with NH_3, suggests that the glucose is attached at the 3-position. R_f data for the aglycone are given in Table 4, where it is compared with 3,6,3′,5′-tetramethoxy-5,7,4′-trihydroxyflavone previously identified in *T. usneoides* [23]. λ_{max} for the aglycone are: MeOH 253, 267, 356, + NaOAc 273, 385, + H_3BO_3 253, 267, 360 nm. The positive NaOAc shift indicates that the 7-position is free and the yellow colour, in UV without fuming with NH_3, that the 3- and 4′-hydroxyls are free and that the 6-hydroxyl is not. The lack of a borate shift indicates that either the 3′ or both 3′ and 5′ positions are blocked. Although no product could be detected on complete demethylation, the MS data confirms that there are three methoxyl groups present at positions 6,3′ and 5′. MS: (*m/e*) 376 (100%) (Found: 376.0795 $C_{18}H_{16}O_9$ requires 376.0794) 375 (0%) 361 (4.8%) 359 (4.5%). This fragmentation pattern is characteristic of compounds with free hydroxyls at 5 and 7 and a methoxyl at 6 [27]. All these data indicate that the new aglycone is 6,3′,5′-trimethoxy-3,5,7,4′-tetrahydroxyflavone and this was confirmed when it was obtained by partial demethylation of 3,6,3′,5′-tetramethoxy-5,7,4′-trihydroxyflavone. The sugar can only be attached to the 3-position (see above) so that the new aglycone occurs in this plant as the 3-glucoside.

Identification of 3,6,3′,5′-tetramethoxy-5,7,4′-trihydroxyflavone-7-glucoside from Tillandsia usneoides. The glycoside was isolated and purified as above. Acid hydrolysis gave glucose and 3,6,3′,5′-tetramethoxy-5,7,4′-trihydroxyflavone. R_f data for glycoside and aglycone are given in Table 4. The identity of the aglycone was confirmed by co-PC in 5 solvents and from a comparison of UV spectral data: λ_{max} MeOH 255, 264′, 357, + NaOAc 267, 372, + H_3BO_3 257, 370, + alk 265, 415 nm. As the glycoside appeared dark to yellow (in UV + NH_3) the 5 and 4′ hydroxyls must be free and the sugar is therefore attached at the 7-position.

Identification of patuletin 3-rhamnoside in Vriesea regina. The glycoside was isolated and purified from leaf tissue as above. Acid hydrolysis for 40 min with 2N HCl gave rhamnose and patuletin only. The aglycone identity was confirmed by co-PC with an authentic marker. λ_{max} for glycoside: MeOH 259, 267′, 348, + NaOAc 268, 377, + H_3BO_3 262, 369 nm. The positive NaOAc and H_3BO_3 shifts indicate that the 7,3′ and 4′ hydroxyls are all free. This data together with the dark to yellow (in UV + NH_3) colour of the glycoside suggests that the sugar is attached to the 3-position. Comparison with the λ_{max}MeOH for patuletin 3-glucoside of 260, 270′, 356 shows a hypsochromic shift in the long wave band of 8 nm in the new glycoside, which confirms the sugar as rhamnose and not glucose (compare quercetin 3-glucoside λ_{max} 363 with its 3-rhamnoside λ_{max}353).

Partial characterisation of two methylated 6-hydroxyluteolin-δ-4′-glucosides in Puya *sp.* The glycosides were isolated and purified from leaf tissue as above. Acid hydrolysis for 40 min with 2N HCl gave glucose and two new dark to yellow (in UV + NH_3) aglycones. R_f data for glycosides and aglycones is given in Table 4. λ_{max} for the glycosides are (1) MeOH 282, 330, + NaOAc

Table 4. Chromatographic data for flavonoid aglycones and glycosides found in the Bromeliaceae

Compound	Colour in UV/+ NH_3	R_f (× 100) in							
		Forestal	50% HOAc	CAW	BAW	PhOH	15% HOAc	H_2O	BEW
6,3′,5′-trimethoxy-3,5,7,4′-tetrahydroxy-flavone*	Y	64	46	78	71	92	—	—	—
3-glucoside*	DK/Y	—	—	—	48	88	37	37	41
3,6,3′,5′--tetramethoxy-5,7,4′-trihydroxy-flavone*		85	71	89	83	—	—	—	—
7-glucoside*	DK/Y	—	—	—	54	55	27	11	36
Unknown aglycone†	Y	69	39	76	23	—	—	—	—
-glucoside	DK/DK	—	—	—	31	—	30	05	18
Methylated 6-hydroxy-luteolin (1)‡	DK/Y	74	70	72	76	—	—	—	—
4′-glucoside‡	DK/DK	—	—	—	49	85	29	05	53
Methylated 6-hydroxy-luteolin (2)‡	DK/Y	80	72	86	77	—	—	—	—
4′-glucoside‡	DK/DK	—	—	—	51	86	35	06	56
6-Hydroxyluteolin	DK/DK	42	31	12	50	—	—	—	—
Patuletin	Y	48	37	32	64	54	—	—	—
3-rhamnoside§	DK/Y	—	—	—	64	71	53	50	62
3-glucoside§	DK/Y	—	—	—	53	59	44	32	48

* Isolated from *Tillandsia usneoides*. † Isolated from *T. caput-medusae*. ‡ Isolated from *Puya* sp. § Isolated from *Vriesea regina*. Key: Y = yellow, DK = dark, CAW = $CHCl_3$–HOAc–H_2O (30:15:2).

275, 335, + H_3BO_3 275, 335 and (2) MeOH 285, 328 nm. Demethylation of both aglycones gave a mixture of 6-hydroxyluteolin and what appeared to be its 7-methyl ether. The lack of borate shift in (1) and the dark colour, in UV not changing with NH_3, of both glycosides suggests that the glucose is attached to the 4'-position in both cases. The dark to yellow colour of both aglycones suggests that the 6-hydroxyl is methylated and the lack of NaOAc shift in (1) that the 7-position is also blocked. R_f data suggests that there is one more methoxyl group in aglycone (2) (see Table 4). From the data available it is therefore suggested that aglycone (1) could be 6,7-dimethoxy-5,3',4'-trihydroxyflavone and aglycone (2) could be 6,7,3'-trimethoxy-5,4'-dihydroxyflavone.

Partial identification of a new flavonoid glycoside from Tillandsia caput-medusae. The glycoside was isolated and purified from leaf tissue as above. Acid hydrolysis gave glucose and a new yellow (in UV) aglycone. λ_{max} for the glycoside are: MeOH 285, 354, + NaOAc 284, 354, + H_3BO_3 287, 305, 348, + $AlCl_3$ 391, + $AlCl_3$ + HCl 382. R_f data for the glycoside and aglycone are given in Table 4. On demethylation of the aglycone no product was detected but the high R_f, 76 in CAW, suggests that is a methylated compound and the spectral data that it is a 6-hydroxylflavonol.

Acknowledgements—The author is grateful to the Director of the Royal Botanic Gardens, Kew for provision of plant material. Mass spectral measurements were kindly carried out by Mr R. W. Butters of Tate and Lyle Ltd. and an exact mass measurement by ICI (Plant Protection) Jealots Hill The author is also indebted to Professor J. B. Harborne for his help and encouragement and to Mr D Philcox of the Royal Botanic Gardens, Kew for help with the taxonomic aspects of the paper.

REFERENCES

1. Tomlinson, P. B. (1969) in *Anatomy of the Monocotyledons* Vol. III (Metcalfe, C. R. ed.). Clarendon Press, Oxford.
2. Takhtajan, A. (1969) *Flowering Plants, Origin and Dispersal* (trans. Jeffrey, C.) Oliver & Boyd, Edinburgh.
3. Cronquist, A. (1968) *The Evolution and Classification of Flowering Plants.* Thomas Nelson, London.
4. Hutchinson, J. (1959) *The Families of Flowering Plants*, Vol II. Clarendon Press, Oxford.
5. Williams, C. A. and Harborne, J. B. (1977) *Biochem. System. Ecol.* **5**, 221.
6. Harborne, J. B. and Clifford, H. T. (1969) *Phytochemistry* **8**, 2071.
7. Bate-Smith, E C. and Harborne, J. B. (1969) *Phytochemistry* **8**, 1035.
8. Williams, C. A., Harborne, J. B. and Clifford, H. T. (1971) *Phytochemistry* **10**, 1059.
9. Harborne, J. B. (1971) *Phytochemistry* **10**, 1569.
10. Williams, C. A. and Harborne, J B (1973) *Phytochemistry* **12**, 2417.
11. Williams, C. A. and Harborne, J. B. (1975) *Biochem System. Ecol.* **3**, 181.
12. Williams, C. A. (1975) *Biochem System Ecol.* **3**, 229.
13. Harborne, J. B. and Williams, C. A. (1976) *Biochem System. Ecol.* **4**, 37.
14. Harborne, J. B and Williams, C. A. (1976) *Biochem. System. Ecol* **4**, 267.
15. Williams, C. A. and Harborne, J B. (1977) *Biochem. System. Ecol.* **5**, 45.
16. Mez, C. (1965) in *Das Pflanzenreich* (Engler, A. ed.) **100** (IV.32.). Verlag von Engelmann (J. Cramer), Weinheim.
17. Hegnauer, R. (1963) *Chemotaxonomie der Pflanzen* Vol. 2 'Monocotyledoneae'. Birkhauser, Basle.
18. Bate-Smith, E. C. (1968) *J. Linn. Soc. (Botany)* **60**, 325
19. Sutherland, G. K. and Gortner, W A. (1959) *Australian J. Chem.* **12**, 240.
20. Gortner, W. A. and Kent, M. J (1958) *J Biol. Chem.* **233**, 731.
21. Gortner, W. A., Kent, M. J and Sutherland, G. K. (1958) *Nature* **81**, 630.
22. Takata, R. H. and Scheuer, P. J. (1976) *Lloydia* **39**, 409
23. Lewis, D. S. and Mabry, T. J. (1977) *Phytochemistry* **16**, 1114.
24. Martinez, M. A. D. P. (1977) unpublished results.
25. Harborne, J B and Clifford, H T. (1969) *Phytochemistry* **8**, 2071.
26. Harborne, J. B. (1977) personal communication
27. Goudard, M., Favre-Bonvin, J., Lebreton, P. and Chopin, J. (1978) *Phytochemistry* **17**, 145.