New Phenolics from *Baccharis* Leaf Exudate

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Several lipophilic phenolics were found in leaf resins of *Baccharis* species, reported earlier to produce terpenoids and flavonoid aglycones. They were identified by NMR and GC/MS studies, respectively. The structure of 2',4'β-trihydroxy-6'-methoxychalcone from *B. salicifolia* was confirmed by synthesis. Phenylethyl caffenate and benzyl caffenate were found in *B. sarothroides*.

**Introduction**

A previous study on the leaf resin composition of some *Baccharis* species revealed that the resinous material consists mostly of terpenoids, in which more or less lipophilic flavonoid aglycones are dissolved [1]. In *B. salicifolia*, a major terpenoid was found to be the rare triterpene maniladiol [2]. The major flavonoids were penduletin (6-hydroxykaempferol-3,6,7-trimethyl ether) and pinocembrin, accompanied by five flavones and seven flavonols [1]. One further compound remained unidentified, due to the paucity of material, and was isolated only in a partially purified state (mentioned as compound 6 in [1].

The leaf resin of *B. sarothroides* A. Gray had been found to consist mostly of the trivial triterpene oleanolic acid and the rare diterpene hauatriwaic acid and its 2-β-hydroxy derivative [2]. The flavonoid aglycones found in this species were exclusively flavonols, most of them substituted at C-6 and/or C-8, with 5,7,4'-trihydroxy-3,6,8-trimethoxy flavone prevailing [1]. In this material, a blue fluorescent spot (polyamide, UV366) remained unidentified in partially purified state (mentioned as compound 6 in [1].

In this material, a blue fluorescent spot (polyamide, UV366) remained unidentified in the earlier study. The two unidentified components from *B. salicifolia* and *B. sarothroides* have now been elucidated by spectroscopic studies. One of these structures was further confirmed by synthesis.

**Materials and Methods**

Plant collection data and general procedures for isolation and detection of flavonoids have been reported earlier [1].

Mass spectra were run at 70 eV on a Hewlett-Packard 5985. NMR spectra were recorded on a Bruker WP 200 SY. Derivatized samples were separated and analyzed in a Finnigan 1020 automated GC/MS (incorporating a Data General Nova 3 computer) [3].

Synthesis of I. Phloracetophenone-4-O-neohesperidoside (II) (1 g, 0.002 mol) was dissolved in dry DMF (10 ml), anhydrous K2CO3 (550 mg, 0.004 mol) and JMe (710 mg, 0.005 mol) were added and the mixture was stirred at 60–70 °C for 6 h. After usual work-up the crude product was found, by 1H NMR, to be a mixture of III, dimethylated II, and II in a relative ratio of approximately 3:1:5:1. This mixture was dissolved in 10% HCl (75 ml) and kept at 80–90 °C for 1 h. After cooling, a white powder was collected and purified by CC in order to eliminate undesired hydrolysis products. We thus obtained 2,4-dihydroxy-6-methoxy-acetophenone IV (105 mg), m.p. 195–197 °C (EtOAc–hexane). MS (DIP) m/z (rel. int.): 182 (M+, 37), 167 (M+–CH3, 100), 152 (M+–2×CH3, 13), 124 (9), 69 (31). 1H NMR...
To a solution of IV (100 mg) in 1.5 m NaOH (2.5 ml) benzoyl chloride (210 mg) was added and the mixture was stirred at room temperature for 2 h. After cooling, it was neutralized with 1 n HCl, extracted with ether and the crude product was purified by CC. 2,4-Dibenzoyloxy-6-methoxy-acetophenone V was obtained as a colourless oil (40 mg). MS m/z (rel. int.): 390 (M+, 28), 375 (M+-CH₃, 4), 359 (M+-OCH₃, 2), 105 (Ph-C=O, 100), 77 (Ph, 53).

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\begin{align*}
&\text{H NMR (CDCl₃) ppm: 8.20 (2H, dd; } J = 8.5 \text{ and } 1.5), 8.13 (2H, dd; } J = 8.5 \text{ and } 1.5), 7.58 (6H, m), 6.87 (1H, d; } J = 2 \text{ Hz), 6.81 (1H, d; } J = 2 \text{ Hz), 3.91 (3H, s; OCH₃), 2.54 (3H, s; COCH₃).}
\end{align*}
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V (40 mg) was dissolved in dry DMSO (1 ml) and powdered NaOH (320 mg) added. The mixture was stirred at room temperature for 5 min, then poured onto ice (25 g), extracted with EtOAc, washed, dried and purified by CC to yield 20 mg of a light-yellow product, m.p. 167—169 °C, which is identical with natural I in every respect.

GC/MS data indicated two phenolic compounds to be present in approximately equal quantities in the B. sarothroides fraction exhibiting the blue fluorescent spot on TLC. Comparison of their retention times and mass spectra with those of authentic reference compounds enabled their identification as benzyl trans-caffeate and phenylethyl trans-caffeate (as the respective bis-TMS derivatives, M~ m/z 414 and 428).

**Results**

The unknown constituent I from B. salicifolia was isolated in pure state after repeated preparative TLC.
ment of the reaction product allowed Baker-Venkataraman rearrangement and cleavage of the second benzoyl moiety without further cyclization. The final product is identical with natural \( I \) in every respect.

From some fractions of \( B. \) salicifolia, a colourless product crystallized, which was not detected in our usual TLC systems, i.e. it is not visible in \( \text{UV}_{366} \) and \( \text{UV}_{254} \) before and after spraying with Naturstoffreagenz A, nor does it react with \( \text{SbCl}_3 \) and \( \text{MnCl}_2 \) spray reagent. In the MS it exhibits \( \text{M}^+ \) at \( m/z \) 386 (\( \text{C}_{23}\text{H}_{46}\text{O}_4 \)). The \(^1\text{H} \) NMR spectrum is very clear, thus showing that the product is pure. Interpretation of the NMR signals allows its identification as a monoglyceride, namely \( 1\)-mono-eicosanyl-glycerol (glycerol ester of arachidic acid).

The product causing the blue fluorescent spot on TLC of \( B. \) sarothroides leaf exudate could not be isolated, due to the minute amount present; it was obtained only in a mixed fraction containing some flavonoids. In this case, structure elucidation was possible by GC/MS analysis and comparison of data with those obtained during recent research on phenolic acid esters from poplar bud exudates and from propolis or bee-glue (cf. [5]). It thus became evident that the fluorescent spot is caused by benzyl caffeate and phenylethyl caffeate.

It should be mentioned in this context that the rare 5,7,4'-trihydroxy-3,6,8-trimethoxyflavonol, reported earlier from \( B. \) sarothroides [1], has now been confirmed by direct comparison with an authentic marker, isolated from Gutierrezia microcephala [6].
**Discussion**

The β-hydroxychalcone or dibenzoylmethane I found in *Baccharis salicifolia* certainly is the most interesting of the phenolics now identified. To our knowledge it is a novel natural product. Further representatives of this unusual group of flavonoids have been reported previously from *Milletia ovatifolia* [7], *Glycyrrhiza echinata* [8], *Lonchocarpus costaricensis* [9], *Primula pulverulenta* [10], and *Tephrosia procumbens* [12]. A product that is closely related structurally to the β-hydroxychalcone I from *B. salicifolia*, has been found earlier in *B. bigelovii*, namely 2β,5,7-trihydroxyflavanone [13], which can be considered the cyclic form of 2',4',6'-trihydroxy-dibenzoylmethane. The 7-glucoside of this unusual flavanone had been reported from *Malus* leaves [14], and the 7-methyl ether was found in *Populus* bud exudates [15].

The two blue fluorescent products from *B. saro-throides*, phenylethyl caffeate and benzyl caffeate, are known as characteristic constituents of the bud exudate of poplar species [5] and they are hence encountered in propolis, too [16].

In a previous report and review on flavonoid aglycones from *Baccharis* [1] we emphasized that reinvestigations of the species studied earlier by various authors would probably yield numerous additional flavonoids if these investigations were done with leaf exudates. The present results prompt us to stress that such reinvestigations would certainly be well worthwhile as they would reveal new products and rare structures.

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