

FLAVONOIDS OF *SALVIA TOMENTOSA* (LABIATAE)

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ABSTRACT.—The following seven flavonoids were obtained from the leaves of *Salvia tomentosa* Mill. (syn. *Salvia grandiflora* Etling) (Labiatae): 5-hydroxy-6,7,3',4'-tetramethoxyflavone, cirsimaritin, jaceosidin, luteolin, luteolin 7-glucoside, 6-methoxyluteolin and a new compound 6-methoxyluteolin 7-glucoside.

Salvia tomentosa Mill., widespread in the Mediterranean and Aegean regions of Turkey, has been used to reduce abdominal pains and to heal wounds. Since luteolin and its derivatives are known to have spasmolytic action (1, 2), their presence in this species, as reported here, may account for its use as a folk medicine. *Salvia* species have been studied for triterpene acids (3, 4), triterpene alcohols (5, 6), diterpenes (7-11), and flavonoids (12-14). This is the first chemical investigation of *Salvia tomentosa*.

EXPERIMENTAL¹

PLANT MATERIAL.—Leaves of *S. tomentosa* were collected from Antakya in southeast Turkey. A specimen of the plant, which was identified by Prof. Dr. A. Baytop (Istanbul), is deposited in the Herbarium of the Faculty of Pharmacy, University of Istanbul (voucher no. ISTE 35146).

EXTRACTION AND ISOLATION OF THE FLAVONOIDS.—Powdered leaves (0.5 kg) of *S. tomentosa* were extracted in a Soxhlet with petroleum ether (bp. 30-60°), benzene, chloroform and ethanol, successively.

The petroleum ether fractions were combined and concentrated to dryness to give a residue of 25 g. After suspension of the material in acetone, insoluble hydrocarbons were removed by centrifugation. Triterpene acids were precipitated from the acetone solution by the following procedure: concentration of the acetone solution to dryness, resuspension of the residue in benzene, treatment of the resultant solution with ammonia and, finally, centrifugation. The supernatant benzene solution was extracted with 60% aqueous ethanol. When the material obtained from the latter extract was chromatographed on polyamide, it yielded 5-hydroxy-6,7,3',4'-tetramethoxyflavone.

The second extract of the plant material (benzene) was concentrated to a small volume, and the resultant concentrate was extracted with 60% aqueous ethanol. The latter extract yielded 7.5 g of syrup and an ether extraction of this syrup gave 3 g of flavonoid material. The flavonoid mixture was separated on a polyamide column (3×75 cm) using a water-ethanol gradient elution system beginning with water; the fractions which eluted with 50% aqueous ethanol yielded 8 mg of 5,4'-dihydroxy-6,7-dimethoxyflavone (cirsimaritin). The fractions which eluted with 60% aqueous ethanol yielded 50 mg of 5,7,4'-trihydroxy-6,3'-dimethoxyflavone (jaceosidin).

The third extract of the plant material (chloroform) contained mainly terpenoids and only traces of flavonoids while the fourth (ethanol) afforded, after workup, 12.5 g of a flavonoid-rich syrup. The syrup was chromatographed on a polyamide column (5.5×30 cm). Elution with water yielded glucose, while elution with 40% aqueous ethanol yielded 6-methoxyluteolin 7-glucoside. Elution with 50% aqueous ethanol gave luteolin 7-glucoside, while 60% aqueous ethanol gave 6-methoxyluteolin and luteolin successively.

CHARACTERIZATION OF 5-HYDROXY-6,7,3',4'-TETRAMETHOXYFLAVONE.—5-Hydroxy-6,7,3',4'-tetramethoxyflavone, mp 189-190° (lit. 189-191°) (15), exhibited uv and pmr spectral properties closely comparable with those of an authentic sample: ms, m/e M^+ : 358 (100%), $M-15$: 343 (71), $M-18$: 341 (10), $M-43$: 315 (13), A_1 : 196 (6), A_1-15 : 181 (15) and B_1 : 162 (10). The identity

¹Spectra were recorded with the following instruments: UV: Varian Techtron model 635; IR: Perkin-Elmer 577 grating model; NMR: Varian A-60A and Varian HA-100; MS: Joel double focus instrument and DuPont 21-491. Melting points were recorded in a Reichert microscope instrument and are not corrected. Adsorbants used for tlc and cc were from Macherey-Nagel and E. Merck.

of the compound was confirmed by comparison with an authentic sample by tlc on polyamide (90% formic acid; R_f 0.38) and cellulose (Avicel) (60% AcOH; R_f 0.90).

CHARACTERIZATION OF 5,4'-DIHYDROXY-6,7-DIMETHOXYFLAVONE (CIRSIMARITIN).—5,4'-Dihydroxy-6,7-dimethoxyflavone, mp 257° (lit. 255–257°) (16), exhibited uv and pmr spectral values similar to literature values (16); ms, m/z M^+ : 314 (100%), M-15: 295 (75), M-43: 271 (22), A₁-15: 181 (20), B₁: 118 (20) and B₂: 131 (8). (The ms fragmentation corresponded to that of a standard sample².)

IDENTIFICATION OF 5,7,4'-TRIHYDROXY-6,3'-DIMETHOXYFLAVONE (JACEOSIDIN).—5,7,4'-Trihydroxy-6,3'-dimethoxyflavone, mp 223–224° (lit. 219–221°) (17), showed uv spectral properties identical to those for jaceosidin; identification was confirmed by pmr (DMSO- d_6) δ 3.8 (s, OCH₃), 3.92 (s, OCH₃), 6.65 (s, C₂-H), 6.89 (s, C₅-H), 7.05 (br. s, C₅-H), 7.4–7.6 (br. m, C₂-H and C₆-H); ms, m/z , M^+ : 330 (100%), M-15: 315 (56), M-18: 312 (45); M-43: 287 (29), A₁-15: 167 (11), B₁: 148 (12) and B₂: 151 (5).

STRUCTURE OF 6-METHOXYLUTEOLIN 7-GLUCOSIDE.—The new glycoside, mp 172–174°, afforded on hydrolysis (with 0.1N HCl for 30 min) 6-methoxyluteolin (tlc, uv, ms comparison with an authentic sample) and glucose (pc comparison with a standard sample). The uv spectra of the glucoside established a 3',4'-ortho-dihydroxyl system (uv spectra with AlCl₃ and H₃BO₃). In addition, the bathochromic shift of 20 nm of Band I with AlCl₃/HCl relative to Band I in MeOH indicated the presence of a 5-hydroxyl-6-methoxyl system (18). Thus, the glucose must be attached at the 7 position. The absence of a Band III peak in the NaOMe spectrum (19) and the lack of a Band II shift with NaOAc confirmed a 7-O-substituent. The complete spectral data for 6-methoxyluteolin 7-glucoside are as follows: uv λ max (MeOH), 346, 272, 255; NaOMe, 404, 269, 230 (sh); AlCl₃, 424, 335 (sh), 300 (sh), 276, 240 (sh); AlCl₃/HCl, 366, 295 (sh), 264, 235 (sh); NaOAc, 400, 300 (sh), 256; NaOAc/H₃BO₃, 375, 261 nm; pmr (as TMS ether in CCl₄) δ 3.2–3.6 (6H, br. m, glucose), 3.75 (s, OCH₃), 5.05 (d, glu-H₁), 6.3 (s, C₅-H), 6.65 (s, C₅-H), 6.85 (d, $J=8$ Hz, C₅-H), 7.28 (d, d, $J=2$ and 9 Hz, C₆-H) and 7.72 (d, $J=2$ Hz, C₂-H); ms (as PDM ether), m/z , M^+ : 597 (73%), M-230 (M-[PDM-glu]); 367 (74), M-248 (M-[PDM-glu-CD₃]): 349 (100); A₁-15: 184 (4), B₁: 168 (14) and B₂: 171 (19); the series of ions due to the PDM-glucosyl moiety were also present: 230 (52), 196 (80) and 161 (62).

IDENTIFICATION OF LUTEOLIN 7-GLUCOSIDE.—Luteolin 7-glucoside, mp 241° (lit. 258–260°) (20), gave uv and pmr spectral results identical to a standard sample; ms (as PDM ether), m/z M^+ : 567 (88%), M-230 (M-[PDM-glucose]): 337 (96), A₁: 169 (13), B₁: 168 (24), 230 (53), 196 (90) and 161 (68). Hydrolysis yielded luteolin and glucose.

IDENTIFICATION OF 6-METHOXYLUTEOLIN.—6-Methoxyluteolin, mp 262° (lit. 258–262°) (21), exhibited uv spectral properties similar to those of an authentic sample; identification was confirmed by pmr and ms: pmr (DMSO- d_6) δ 3.8 (s, OCH₃); 6.59 (s, C₅-H); 6.66 (s, C₅-H); 6.92 (d, $J=9$ Hz, C₅-H), 7.3–7.5 (br. m, C₂-H and C₆-H) ppm; ms, m/z M^+ : 316 (100%), M-15: 301 (86), M-18: M-43: 273 (73), A₁-15: 167 (26), B₁: 134 (7) and B₂: 137 (13).

IDENTIFICATION OF LUTEOLIN.—Luteolin exhibited uv, pmr, ms and tlc properties identical to those of a standard sample.

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