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# FLAVONOL GLYCOSIDES OF *PHLOMIS SPECTABILIS*

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**Key Word Index**—*Phlomis spectabilis*, Labiatae, flavonol glucosides, tiliroside, astragalin

**Abstract**—Four flavonol glucosides, one new, have been isolated from a methanolic extract of *Phlomis spectabilis* Their structures were established as the 3-glucosides and 3-(6″-(*E*)-*p*-coumaroyl)glucosides of kaempferol and of kaempferol 7,4′-dimethyl ether

*Phlomis spectabilis* Falc ex Benth is a temperate subalpine perennial herb growing in Kashmir (2400–2600 m) The plant has not been chemically investigated previously, although some members of the genus are reported to contain iridoids [1–3], alkaloids [4], flavonoids [5] and essential oils [6], and some subtropical *Phlomis* species are reported to possess medicinal and insecticidal properties [7, 8] The present communication deals with the isolation and characterization of the 3-*O*-[6″-*O*-(*E*)-*p*-coumaroyl]-β-D-glucopyranoside of kaempferol 7,4′-dimethyl ether (**1**), tiliroside (**2**) [9, 10], kaempferol 7,4′-dimethyl ether 3-glucoside (**3**) [11] and astragalin (**4**) [12] These flavonoids are reported for the first time from genus *Phlomis* and to our knowledge **1** has not been reported previously from any natural source

A defatted methanol extract of whole plant on repeated CC over silica gel furnished compounds **1**–**4** Compound **1**, mp 133–135°, $C_{32}H_{30}O_{13}$, showed signals for 12 protons in the aromatic region of the $^1$H NMR spectrum Out of these, eight protons were accounted for by the two aromatic AA′BB′ systems, one of these being attributed to the flavonoid B-ring Two doublets with large coupling constants (16 Hz), which excluded their aromatic origin, could be assigned to two *trans* olefinic protons This, along with the other AA′BB′ system and an α,β-unsaturated ester carbonyl band in the IR spectrum, showed an (*E*)-*p*-coumaroyl moiety in **1** This was confirmed by mild alkaline hydrolysis of **1** when (*E*)-*p*-hydroxycinnamic acid was obtained as one of the two products formed The remaining two signals in the aromatic region appeared as *meta*-coupled doublets and their chemical shifts allocated them to H-6 and H-8 in the A-ring $^1$H NMR also allowed the identification of characteristic H-1″ of a glucose moiety, two methoxyls and two phenolic hydroxyls (one chelated due to its downfield position) The other alkaline hydrolysis product of **1** was found to be identical in all respects with **3** Compound **3**, mp 224–226°, showed a similar $^1$H NMR pattern to **1**, but lacked signals due to (*E*)-*p*-coumaroyl moiety On acid hydrolysis it gave glucose and an aglycone which showed similar physico-chemical properties as those reported for 3,5-dihydroxy-7,4′-dimethoxyflavone [13] Compound **2**, mp 264–266°, was identified as tiliroside by comparing its spectral data with the reported data [10] $^{13}$C NMR of sugar moiety of **2** shows a downfield shift of C-6″ ($\Delta\delta$ +2) and an upfield shift of C-5″ ($\Delta\delta$ −2 9) from the chemical shift values reported for the corresponding carbon resonances of flavonol 3-*O*-glucopyranosides [14] These shifts are expected from the substituent effects of C-6″ acylation [15] This evidence excludes other possible acylation sites in the glucose moiety of **2** and fixes the (*E*)-*p*-coumaroyl group to C-6″ $^1$H NMR of **2** was almost identical to that of **1**, but lacked two methoxy group signals and exhibited two extra hydroxyl singlets Thus **1** appears to be a dimethyl ether of **2** This assumption was confirmed by treatment of **1** with $CH_2N_2$ to give a monomethyl ether, which was identical with the methylation product of **2**, obtained by similar treatment (mp, mmp, co-TLC, superimposable IR and $^1$H NMR) Presence of a chelated hydroxyl signal in the $^1$H NMR spectrum of this methylated product, and formation of (*E*)-*p*-hydroxycinnamic acid by **1**, leaves only two positions, viz C-7 and C-4′, in **1** for placing the methoxyls Thus **1** is the 7,4′-dimethyl ether of tiliroside and **3** is therefore kaempferol 7,4′-dimethyl ether 3-glucoside Compound **4**, mp 175–176°, was found to be identical with the alkaline hydrolysis product of **2** This and its spectral data established its structure as astragalin

## EXPERIMENTAL

Mps are uncorr The air dried defatted plant material (10 kg) of *P spectabilis* (voucher No 1366 UD, deposited with the Herbarium of Botany Department, Kashmir University), collected in the flowering season, was extracted with MeOH The crude syrup obtained (180 g), after removal of MeOH under red pres, was percolated on a silica gel column (3 5 kg, 60–120 mesh) and eluted with $CHCl_3$, EtOAc and MeOH The EtOAc fraction was twice re-chromatographed over silica gel with $CHCl_3$–MeOH mixture in different proportions and afforded 140 mg **1** ($CHCl_3$–MeOH, 4 1), 435 mg **2** ($CHCl_3$–MeOH, 3 1), 65 mg **3** ($CHCl_3$–MeOH, 3 1) and 95 mg **4** ($CHCl_3$–MeOH, 2 1) Known compounds were identified by comparing their UV, IR and $^1$H NMR spectra with the reported data

Compound **1** (pale yellow crystals, MeOH), mp 133–135°, $C_{32}H_{30}O_{13}$ UV $\lambda_{max}^{MeOH}$ nm 270, 311, 352 inf, +$AlCl_3$ 280, 305 352 inf, +NaOAc 270, 311, 352 inf $^1$H NMR (90 MHz,

$Me_2CO$-$d_6$) $\delta$2 60–4 41 (glucose protons), 3 80 (3H, *s*, OMe), 3 89 (3H, *s*, OMe), 4 98 (1H, *bs*, OH), 5 39 (1H, *d*, $J$ = 7 Hz, H-1″), 6 19 (1H, *d*, $J$ = 16 Hz, CO–CH=C–Ph), 6 29 (1H, *d*, $J$ = 2 5 Hz, H-6), 6 61 (1H, *d*, $J$ = 2 5 Hz, H-8), 6 99 (2H, *d*, $J$ = 9 Hz, H-3‴, H-5‴), 7 00 (2H, *d*, $J$ = 9 Hz, H-3′, H-5′), 7 47 (1H, *d*, $J$ = 16 Hz, CO–C=CH–Ph), 7 52 (2H, *d*, $J$ = 9 Hz, H-2‴, H-6‴), 8 19 (2H, *d*, $J$ = 9 Hz, H-2′, H-6′), 12 81 (1H, *bs*, OH), signals at $\delta$4 98 and 12 81 exchanged with $D_2O$ EIMS (probe) 70 eV, $m/z$ (rel int) 314 $[M - \text{coumaroylglucose}]^+$ (100), 300 (11 7), 286 (8 08), 276 (13 9), 246 (7 3), 181 (19 8), 178 (6 6), 166 (51 4), 136 (20 5), 134 (13 2), 126 (6 6), 98 (16 9), 77 (13 9), 69 (27 9), etc (Found C, 61 54, H, 4 51 $C_{32}H_{30}O_{13}$ requires C, 61 73, H, 4 98%) Methylation ($CH_2N_2$–$Et_2O$) gave a monomethyl ether, mp 148–150°, $C_{33}H_{32}O_{13}$ IR $\nu_{max}^{KBr}$ $cm^{-1}$ 3460–3340 (OH), 1720 (C=C–$CO_2$), 1640 (>C=O) $^1H$ NMR (90 MHz, $CDCl_3$) $\delta$2 85–4 35 (glucose protons), 3 78 (9H, *s*, 3 × OMe), 5 45 (1H, *d*, $J$ = 7 Hz, H-1″), 6 07 (1H, *d*, $J$ = 16 Hz, CO–CH=C–Ph), 6 17 (1H, *d*, $J$ = 2 5 Hz, H-6), 6 30 (1H, *d*, $J$ = 2 5 Hz, H-8), 6 77 (4H, *d*, $J$ = 9 Hz, H-3′, H-5′ and H-3‴, H-5‴), 7 31 (2H, *d*, $J$ = 9 Hz, H-2‴, H-6‴), 7 43 (1H, *d*, $J$ = 16 Hz, CO–C=CH–Ph), 8 03 (2H, *d*, $J$ = 9 Hz, H-2′, H-6′), 12 46 (1H, *bs*, OH-exchanged with $D_2O$) Alkaline hydrolysis (10% methanolic KOH) of **1** gave an acid and **3** Acid, mp 205–206° (MeOH–$CHCl_3$) $^1H$ NMR (60 MHz, $CDCl_3$–DMSO-$d_6$), $\delta$5 2 (1H, *hump*, OH-exchanged with $D_2O$), 6 23 (1H, *d*, $J$ = 16 Hz, CH=C–Ph), 6 87 (2H, *d*, $J$ = 9 Hz, H-3, H-5), 7 40 (2H, *d*, $J$ = 9 Hz, H-2, H-6), 7 60 (1H, *d*, $J$ = 16 Hz, C=CH–Ph), identified as (*E*)-*p*-hydroxycinnamic acid

Compound **2**, mp 264–266° (MeOH), $C_{30}H_{26}O_{13}$ $^1H$ NMR (100 MHz, DMSO-$d_6$) $\delta$3 2–4 2 (glucose protons), 5 19 (1H, *bs*, OH), 5 45 (1H, *bs*, OH), 5 47 (1H, *d*, $J$ = 7 Hz, H-1″), 6 12 (1H, *d*, $J$ = 16 Hz, CO–CH=C–Ph), 6 19 (1H, *d*, $J$ = 2 Hz, H-6), 6 40 (1H, *d*, $J$ = 2 Hz, H-8), 6 81 (2H, *d*, $J$ = 8 Hz, H-3‴, H-5‴), 6 89 (2H, *d*, $J$ = 8 Hz, H-3′, H-5′), 7 37 (1H, *d*, $J$ = 16 Hz, CO–C=CH–Ph), 7 39 (2H, *d*, $J$ = 8 Hz, H-2‴, H-6‴), 8 01 (2H, *d*, $J$ = 8 Hz, H-2′, H-6′), 10 13 (1H, *bs*, OH), 12 6 (1H, *bs*, OH), signals at $\delta$5 19, 5 45, 10 13 and 12 6 exchanged with $D_2O$ $^{13}C$ NMR (25 2 MHz, DMSO-$d_6$) $\delta$63 0 (*t*, C-6″), 70 0 (*d*, C-4″), 74 3 (*d*, C-2″, C-5″), 76 3 (*d*, C-3″), 93 5 (*d*, C-8), 98 5 (*d*, C-6), 101 2 (*d*, C-1″), 103 7 (*s*, C-10), 113 7 (*d*, C-α), 115 2 (*d*, C-3‴, C-5‴), 115 7 (*d*, C-3′, C-5′), 120 7 (*s*, C-1′), 125 0 (*s*, C-1‴), 130 0 (*d*, C-2′, C-6′), 130 5 (*d*, C-2‴, C-6‴), 133 4 (*s*, C-3), 144 2 (*d*, C-β), 156 4 (*s*, C-2), 156 5 (*s*, C-9), 159 7 (*s*, C-4′, C-4‴), 161 3 (*s*, C-5), 164 2 (*s*, C-7), 165 8 (*s*, $\underline{C}O_2$), 177 5 (*s*, C-4) Methylation ($CH_2N_2$–$Et_2O$) gave a trimethyl ether, mp, mmp, co-TLC and superimposable IR and $^1H$ NMR with monomethyl ether of **1** Alkaline hydrolysis of **2** gave (*E*)-*p*-hydroxycinnamic acid and **4**

Compound **3**, mp 224–226° (MeOH), $C_{23}H_{24}O_{11}$ UV $\lambda_{max}^{MeOH}$ nm 265, 295, 340; + $AlCl_3$ 260, 300, 365 $^1H$ NMR (90 MHz, DMSO-$d_6$) $\delta$2 60–4 30 (glucose protons), 3 83 (3H, *s*, OMe), 3 86 (3H, *s*, OMe), 5 18 (1H, *d*, $J$ = 7 Hz, H-1″), 6 43 (1H, *d*, $J$ = 2 5 Hz, H-6), 6 70 (1H, *d*, $J$ = 2 5 Hz, H-8), 7 00 (2H, *d*, $J$ = 9 Hz, H-3′, H-5′), 8 11 (2H, *d*, $J$ = 9 Hz, H-2′, H-6′), 12 60 (1H, *bs*, OH-exchanged with $D_2O$) Acid hydrolysis of **3** gave glucose (PC) and 3,5-dihydroxy-7,4′-dimethoxyflavone identified by comparison of UV, IR and $^1H$ NMR data with the reported data [13]

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