# Phenylethyl Glycosides from *Globularia alypum* Growing in Turkey

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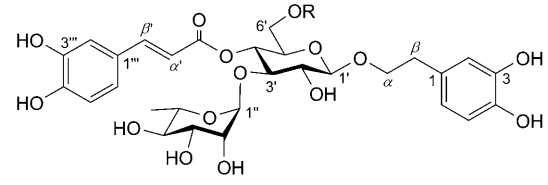
From the leaves of *Globularia alypum*, three new phenylethyl glycosides, namely galypumosides A (=2-(3,4-dihydroxyphenyl)ethyl *O*-$\alpha$-rhamnopyranosyl-(1→3)-4-*O*-[(*E*)-caffeoyl]-6-*O*-[(*E*)-*p*-coumaroyl]-$\beta$-glucopyranoside; **1**), B (=2-(3,4-dihydroxyphenyl)ethyl *O*-$\alpha$-rhamnopyranosyl-(1→3)-4-*O*-[(*E*)-caffeoyl]-6-*O*-[(*E*)-feruloyl]-$\beta$-glucopyranoside; **2**), and C (=2-(3,4-dihydroxyphenyl)ethyl *O*-$\alpha$-rhamnopyranosyl-(1→3)-4-*O*-[(*E*)-caffeoyl]-6-*O*-menthiafoloyl-$\beta$-glucopyranoside; **3**), were isolated, together with two known phenylethyl glycosides, calceolarioside A and verbascoside. Eight iridoid glucosides, catalpol, globularicisin, globularin, globularidin, globularinin, globularimin, lytanthosalin, and alpinoside, a flavon glycoside, 6-hydroxyluteolin 7-*O*-sophoroside, a lignan glycoside, syringaresinol 4′-*O*-$\beta$-glucopyranoside, and a phenylpropanoid glycoside, syringin, were also obtained and characterized. The structures of the isolates were elucidated on the basis of 1D- and 2D-NMR experiments as well as HR-MALDI-MS.

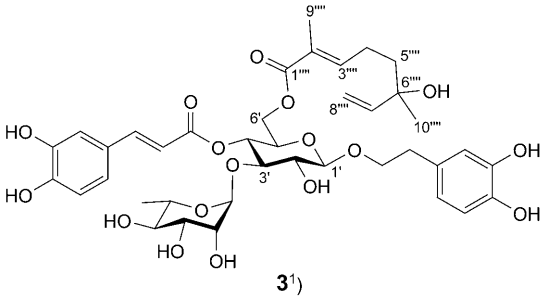
**1. Introduction.** – *Globularia alypum* L. (formerly Globulariaceae, now Plantaginaceae) is a shrub distributed in the Mediterranean area. It is one of the nine species of the genus *Globularia* growing in Turkey [1][2]. *G. alypum* is widely utilized in indigenous systems of medicine in some Mediterranean countries, as a hypoglycaemic agent, laxative, cholagogue, and stomachic [3]. In Moroccan folk medicine, *G. alypum* is one of the mostly used medicinal plants for the treatment of diabetes and hypertension [4]. Several biological studies on this plant showed its significant hypoglycaemic activity in rats [5–7]. Antioxidant effects of *G. alypum* extract and its constituents have also been reported [8]. Recent phytochemical researches on this plant have resulted in the isolation of various types of iridoid glucosides [9] as well as phenylethyl and flavonoid glycosides [10]. As a part of our continuing phytochemical investigations on the members of the genus *Globularia* from the flora of Turkey [11–15], we have investigated the chemical constituents of *G. alypum*. In this article, we describe the isolation and structural elucidation of the new phenylethyl glycosides **1**–**3**[1]).

**2. Results and Discussion.** – The air-dried leaves of *G. alypum* were extracted with MeOH. The $H_2O$-soluble part of the MeOH extract was separated by a combination of

[1]) Trivial atom numbering; for systematic names, see *Exper. Part.*



**1** R = (*E*)-*p*-coumaroyl[1])[2])
**2** R = (*E*)-feruloyl[1])[2])



**3**[1])

various chromatographic methods to obtain the three new phenylethyl glycosides **1**–**3**, in addition to 13 known metabolites: two phenylethyl glycosides, calceolarioside A [16] and verbascoside [17], eight iridoid glucosides, catalpol, globularicisin, globularin, globularidin, globularinin, globularimin [18], lytanthosalin [19], and alpinoside [20], a flavon glycoside, 6-hydroxyluteolin 7-*O*-sophoroside [21], a lignan glycoside, syringaresinol 4′-*O*-*β*-D-glucopyranoside [22], and a phenylpropanoid glycoside, syringin [18]. All these known compounds were characterized by comparison of their spectroscopic data (1D- and 2D-NMR and MS) with previously published data.

Compound **1** was obtained as an amorphous powder. Its molecular formula was determined to be $C_{38}H_{42}O_{17}$ by the $[M+Na]^+$ ion peak at *m/z* 793.2327 in the HR-MALDI-MS. Based on further spectroscopic data ($^1$H- and $^{13}$C-NMR (*Table 1*), HSQC, and HMBC spectra), the structure of **1** was identified as 2-(3,4-dihydroxyphenyl)ethyl *O*-*α*-rhamnopyranosyl-(1 → 3)-4-*O*-[(*E*)-caffeoyl]-6-*O*-[(*E*)-*p*-coumaroyl]-*β*-glucopyranoside[1])[2]), and named galypumoside A.

The $^1$H-NMR spectrum of **1** showed the characteristic signals arising from an (*E*)-caffeoyl moiety[2]), *i.e.*, three aromatic H-atoms as an *ABX* system at *δ* 7.08 (*d*, *J* = 2.0 Hz), 6.97 (*dd*, *J* = 8.1 and 2.0 Hz), and 6.78 (*d*, *J* = 8.1 Hz) and two *trans*-olefinic H-atoms as an *AB* system at *δ* 7.63 and 6.30 (*d*, *J* = 15.9 Hz), from an (*E*)-*p*-coumaroyl moiety[2]), *i.e.*, four aromatic resonances as an *AA′BB′* system at *δ* 7.33 and 6.76 (*d*, *J* = 8.5 Hz) and two *trans*-olefinic H-atoms as an *AB* system at *δ* 7.62 and 6.29 (*d*, *J* = 15.7 Hz), and from a 2-(3,4-dihydroxyphenyl)ethoxy group, *i.e.*, three aromatic H-atoms as an *ABX* system at *δ* 6.72 (*d*, *J* = 1.6 Hz), 6.68 (*d*, *J* = 8.1 Hz), and 6.58 (*dd*, *J* = 8.1 and 1.6 Hz), two geminal

[2]) Caffeic acid = 3-(3,4-dihydroxyphenyl)prop-2-enoic acid, *p*-coumaric acid = 3-(4-hydroxyphenyl)-prop-2-enoic acid, ferulic acid = 3-(4-hydroxy-3-methoxyphenyl)prop-2-enoic acid, and menthiafolic acid = 6-hydroxy-2,6-dimethylocta-2,7-dienoic acid.

Table 1. *$^1$H- and $^{13}$C-NMR Data*[a]) ($CD_3OD$) *of* **1** *and* **2**[1]). $\delta$ in ppm, $J$ in Hz.

| | **1** | | **2** | |
|---|---|---|---|---|
| | $\delta$(H)[b]) | $\delta$(C)[c]) | $\delta$(H)[b]) | $\delta$(C)[c]) |
| Aglycone: | | | | |
| C(1) | – | 131.0 | – | 131.0 |
| H–C(2) | 6.72 (*d*, *J* = 1.6) | 117.0 | 6.72 (*d*, *J* = 1.8) | 117.0 |
| C(3) | – | 145.6 | – | 145.6 |
| C(4) | – | 144.3 | – | 144.3 |
| H–C(5) | 6.68 (*d*, *J* = 8.1) | 116.2 | 6.67 (*d*, *J* = 8.1) | 116.2 |
| H–C(6) | 6.58 (*dd*, *J* = 1.6, 8.1) | 121.2 | 6.58 (*dd*, *J* = 8.1, 1.8) | 121.2 |
| $CH_2(\alpha)$ | 3.78–4.02 (*m*) | 72.5 | 3.78–4.01 (*m*) | 72.5 |
| $CH_2(\beta)$ | 2.83 (*t*, *J* = 8.3) | 36.5 | 2.83 (*t*, *J* = 8.3) | 36.5 |
| Glc: | | | | |
| H–C(1′) | 4.46 (*d*, *J* = 8.1) | 104.1 | 4.46 (*d*, *J* = 7.9) | 104.5 |
| H–C(2′) | 3.47 (*dd*, *J* = 8.1, 8.9) | 76.0 | 3.46 (*dd*, *J* = 9.1, 7.9) | 76.2 |
| H–C(3′) | 3.89 (*t*, *J* = 8.9) | 81.4 | 3.88 (*t*, *J* = 9.1) | 81.4 |
| H–C(4′) | 5.08 (*t*, *J* = 9.3) | 70.8 | 5.08 (*t*, *J* = 9.3) | 70.8 |
| H–C(5′) | 3.83–3.88 (*m*) | 72.7 | 3.83–3.88 (*m*) | 73.0 |
| $CH_2$(6′) | 4.31 (*dd*, *J* = 12.1, 2.0), 4.27 (*dd*, *J* = 12.1, 4.0) | 63.9 | 4.29[d]) | 64.2 |
| Rha: | | | | |
| H–C(1″) | 5.23 (*d*, *J* = 1.6) | 102.9 | 5.23 (*d*, *J* = 1.6) | 102.9 |
| H–C(2″) | 3.96 (*dd*, *J* = 3.0, 1.6) | 72.2 | 3.96 (*dd*, *J* = 3.0, 1.6) | 72.2 |
| H–C(3″) | 3.61 (*dd*, *J* = 9.5, 3.0) | 71.5 | 3.61 (*dd*, *J* = 9.5, 3.0) | 71.5 |
| H–C(4″) | 3.32 (*t*, *J* = 9.5) | 73.5 | 3.32 (*t*, *J* = 9.5) | 73.5 |
| H–C(5″) | 3.57–3.62 (*m*) | 70.7 | 3.57–3.62 (*m*) | 70.7 |
| Me(6″) | 1.12 (*d*, *J* = 6.1) | 18.5 | 1.12 (*d*, *J* = 6.1) | 18.5 |
| Caffeoyl[2]): | | | | |
| C(1‴) | – | 127.4 | – | 127.3 |
| H–C(2‴) | 7.08 (*d*, *J* = 2.0) | 115.1 | 7.07 (*d*, *J* = 2.0) | 115.2 |
| C(3‴) | – | 146.5 | – | 146.5 |
| C(4‴) | – | 149.5 | – | 149.5 |
| H–C(5‴) | 6.78 (*d*, *J* = 8.1) | 116.5 | 6.78 (*d*, *J* = 8.1) | 116.4 |
| H–C(6‴) | 6.97 (*dd*, *J* = 8.1, 2.0) | 123.2 | 6.95 (*dd*, *J* = 8.1, 2.0) | 122.9 |
| H–C(α′) | 6.30 (*d*, *J* = 15.9) | 114.8 | 6.31 (*d*, *J* = 15.9) | 114.7 |
| H–C(β′) | 7.63 (*d*, *J* = 15.9) | 147.0 | 7.62 (*d*, *J* = 15.9) | 147.8 |
| C=O | – | 168.0 | – | 167.7 |
| *p*-Coumaroyl/ Feruloyl[2]): | | | | |
| C(1⁗) | – | 126.9 | – | 127.3 |
| H–C(2⁗) | 7.33 (*d*, *J* = 8.5) | 131.0 | 7.11 (*d*, *J* = 2.0) | 111.4 |
| H–C(3⁗) | 6.76 (*d*, *J* = 8.5) | 116.5 | – | 150.2 |
| C(4⁗) | – | 161.1 | – | 149.0 |
| H–C(5⁗) | 6.76 (*d*, *J* = 8.5) | 116.5 | 6.77 (*d*, *J* = 8.1) | 116.4 |
| H–C(6⁗) | 7.33 (*d*, *J* = 8.5) | 131.0 | 6.97 (*dd*, *J* = 8.1, 2.0) | 124.1 |
| H–C(α″) | 6.29 (*d*, *J* = 15.7) | 114.5 | 6.35 (*d*, *J* = 15.9) | 114.4 |
| H–C(β″) | 7.62 (*d*, *J* = 15.7) | 147.0 | 7.62 (*d*, *J* = 15.9) | 147.1 |
| C=O | – | 168.6 | – | 168.3 |
| MeO | – | – | 3.84 (*s*) | 56.3 |

[a]) All $\delta$(H) and $\delta$(C) assignments are based on 2D-NMR (COSY, HSQC, HMBC) spectral data. [b]) Recorded at 600 MHz. [c]) Recorded at 150 MHz). [d]) Signal pattern unclear due to overlapping.

benzylic $CH_2$ H-atoms at $\delta$ 2.83 ($t$, $J = 8.3$ Hz), and two nonequivalent oxygenated $CH_2$ H-atoms at $\delta$ 4.02 and 3.78 (each 1 H, $m$). Furthermore, two anomeric-H-atom resonances appeared at $\delta$ 4.46 ($d$, $J =$ 8.1 Hz, H−C(1′) of $\beta$-glucose) and 5.23 ($d$, $J = 1.6$ Hz, H−C(1″) of $\alpha$-rhamnose), indicating a diglycosidic structure, which was confirmed by the corresponding anomeric C-atom resonances at $\delta$ 104.1 and 102.9 in the $^{13}$C-NMR spectrum (*Table 1*). The remaining C-signals of the sugar units indicated that $\beta$-glucose is glycosylated at C(3′) ($\delta$ 81.4), while $\alpha$-rhamnose is terminal. The NMR data of **1** were found to be similar to those of verbascoside [17], except for the additional (*E*)-*p*-coumaroyl signals of **1**. The downfield shifts for $CH_2$(6′) ($\delta$ 4.31 and 4.27) and C(6′) ($\delta$ 63.9) and the upfield shift for C(5′) ($\delta$ 72.7) established the esterification site of the (*E*)-*p*-coumaroyl unit to be at OH−C(6′) of the glucose. Interpretation of the HMBC spectrum permitted the determination of all the interfragmental connectivities. Thus, cross-peaks were observed between H−C(1′) ($\delta$ 4.46) of glucose and C($\alpha$) ($\delta$ 72.5) of the (dihydroxyphenyl)ethyl moiety, between H−C(1″) ($\delta$ 5.23) of rhamnose and C(3′) ($\delta$ 81.4) of glucose, between H−C(4′) ($\delta$ 5.08) of glucose and the carbonyl C-atom ($\delta$ 168.0) of the caffeoyl unit, and between $CH_2$(6′) ($\delta$ 4.31 and 4.27) of glucose and the carbonyl C-atom ($\delta$ 168.6) of the *p*-coumaroyl moiety. Consequently, compound **1** was found to be a new verbascoside derivative.

Compound **2** was obtained as an amorphous powder. Its molecular formula was determined to be $C_{39}H_{44}O_{18}$ by the $[M+Na]^+$ ion peak at $m/z$ 823.2431 in the HR-MALDI-MS. The $^1$H- and $^{13}$C-NMR spectrum (*Table 1*) revealed the presence of signals indicative of an (*E*)-caffeoyl moiety, a 2-(3,4-dihydroxyphenyl)ethoxy group, and two anomeric H-atoms. These findings suggested the presence of a verbascoside skeleton in **2** as in **1**. The complete analysis of the remaining $^1$H- and $^{13}$C-NMR signals assigned by 1D-TOCSY and 2D-NMR techniques (COSY, HSQC, and HMBC) indicated that **2** possesses an (*E*)-feruloyl moiety[2]) instead of the (*E*)-*p*-coumaroyl unit attached to OH−C(6′) of $\beta$-glucose. Thus, the structure of **2** was determined to be 2-(3,4-dihydroxyphenyl)ethyl *O*-$\alpha$-rhamnopyranosyl-(1 → 3)-4-*O*-[(*E*)-caffeoyl]-6-*O*-[(*E*)-feruloyl]-$\beta$-glucopyranoside[1])[2]), and named galypumoside B.

The final structure proof came from a HMBC experiment with **2**, in which cross-peaks between H−C(1′) ($\delta$ 4.46) of glucose and C($\alpha$) ($\delta$ 72.5) of the (dihydroxyphenyl)ethyl moiety, between H−C(1″) ($\delta$ 5.23) of rhamnose and C(3′) ($\delta$ 81.4) of glucose, between H−C(4′) ($\delta$ 5.08) of glucose and the C=O C-atom ($\delta$ 167.7) of the caffeoyl unit, and between $CH_2$(6′) ($\delta$ 4.29) of glucose and the C=O C-atom ($\delta$ 168.3) of the (*E*)-feruloyl moiety[2]) were observed.

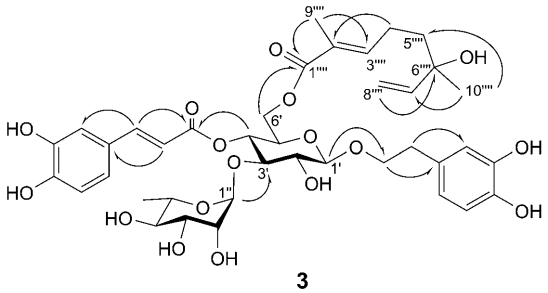
Compound **3** was obtained as amorphous powder. The molecular formula, $C_{39}H_{50}O_{17}$, requiring 15 degrees of unsaturation, was deduced by a combination of HR-MALDI-MS ($[M+Na]^+$ at $m/z$ 813.3056) and $^{13}$C-NMR data (*Table 2*). The $^1$H- and $^{13}$C-NMR, COSY, HSQC, and HMBC (*Table 2* and *Figure*) experiments established the structure of compound **3** as 2-(3,4-dihydroxyphenyl)ethyl *O*-$\alpha$-rhamnopyranosyl-(1 → 3)-4-*O*-[(*E*)-caffeoyl]-6-*O*-menthiafoloyl-$\beta$-glucopyranoside[1])[2]) which was named galypumoside C.

Analysis of the $^1$H- and $^{13}$C-NMR spectra of **3** (*Table 2*) showed the presence of an (*E*)-caffeoyl, a 2-(3,4-dihydroxyphenyl)ethoxy, and a disaccharide moiety, similar to compounds **1** and **2**, indicating the existence of a verbascoside skeleton. The $^{13}$C-NMR spectrum of **3** displayed 39 resonances, eight of which could be ascribed to the aglycone (2-(3,4-dihydroxyphenyl)ethyl unit), and nine of which were attributed to an aromatic acyl unit ((*E*)-caffeoyl moiety), while 12 of which were ascribed to the sugar portion ($\beta$-glucose and $\alpha$-rhamnose). All the remaining $^{13}$C signals (169.1, 145.8, 144.9, 127.9, 112.2, 73.3, 41.6, 27.5, 24.2, and 12.0) established by HSQC and HMBC experiments (*Figure*) were assignable to a $C_{10}$ acyclic monoterpenoid acid with three unsaturations. The $^1$H-NMR spectrum of **3** exhibited signals of an

Table 2. *$^1$H- and $^{13}$C-NMR Data*[a]) ($CD_3OD$) *and Significant HMBCs for* **3**. $\delta$ in ppm, $J$ in Hz.

| | $\delta$(H)[b]) | $\delta$(C)[c]) | HMBC (H → C) |
|---|---|---|---|
| Aglycone: | | | |
| C(1) | – | 131.1 | |
| H–C(2) | 6.70 (*d*, *J* = 1.8) | 116.7 | C(3), C(4), C(6) |
| C(3) | – | 146.0 | |
| C(4) | – | 144.5 | |
| H–C(5) | 6.69 (*d*, *J* = 8.1) | 116.2 | C(1), C(3), C(4) |
| H–C(6) | 6.58 (*dd*, *J* = 8.1, 1.8) | 120.8 | C(2), C(4) |
| $CH_2(\alpha)$ | 3.75–3.98 (*m*) | 72.4 | C(1′), C(1) |
| $CH_2(\beta)$ | 2.81 (*t*, *J* = 8.3) | 36.4 | C($\alpha$), C(1), C(2), C(6) |
| Glc: | | | |
| H–C(1′) | 4.43 (*d*, *J* = 7.9) | 104.2 | C($\alpha$) |
| H–C(2′) | 3.42 (*dd*, *J* = 9.1, 7.9) | 76.0 | |
| H–C(3′) | 3.86 (*t*, *J* = 9.1) | 81.2 | |
| H–C(4′) | 5.04 (*t*, *J* = 9.3) | 70.6 | C=O (caffeoyl) |
| H–C(5′) | 3.80–3.86 (*m*) | 73.0 | |
| $CH_2$(6′) | 4.26[d]) | 64.1 | C=O (menthiafoloyl) |
| Rha: | | | |
| H–C(1″) | 5.22 (*d*, *J* = 1.6) | 102.9 | C(3′) |
| H–C(2″) | 3.94 (*dd*, *J* = 3.0, 1.6) | 72.1 | |
| H–C(3″) | 3.61 (*dd*, *J* = 9.5, 3.0) | 71.5 | |
| H–C(4″) | 3.31 (*t*, *J* = 9.5) | 73.6 | |
| H–C(5″) | 3.57–3.60 (*m*) | 70.6 | |
| Me(6″) | 1.12 (*d*, *J* = 6.1) | 18.2 | |
| Caffeoyl[1]): | | | |
| C(1‴) | – | 127.2 | |
| H–C(2‴) | 7.07 (*d*, *J* = 2.0) | 114.9 | C(4‴), C(6‴) |
| C(3‴) | – | 146.7 | |
| C(4‴) | – | 149.6 | |
| H–C(5‴) | 6.80 (*d*, *J* = 8.1) | 116.3 | C(1‴) |
| H–C(6‴) | 6.98 (*dd*, *J* = 8.1, 2.0) | 123.1 | C(2‴), C(5‴), C(β′) |
| H–C($\alpha'$) | 6.28 (*d*, *J* = 15.9) | 114.3 | C(1‴), C=O, C($\beta'$) |
| H–C($\beta'$) | 7.60 (*d*, *J* = 15.9) | 147.8 | C(2‴), C(6‴), C(1‴), C=O |
| C=O | – | 168.0 | |
| Menthiafoloyl[1]): | | | |
| C(1⁗) | – | 169.1 | |
| C(2⁗) | – | 127.9 | |
| H–C(3⁗) | 6.80[d]) | 144.9 | C(1⁗), C(9⁗) |
| $CH_2$(4⁗) | 2.13–2.20 (*m*) | 24.2 | C(2⁗), C(3⁗), C(5⁗) |
| $CH_2$(5⁗) | 1.52–1.56 (*m*) | 41.6 | C(3⁗), C(6⁗), C(7⁗), C(10⁗) |
| C(6⁗) | – | 73.3 | |
| H–C(7⁗) | 5.86 (*dd*, *J* = 17.6, 10.9) | 145.8 | C(6⁗) |
| $CH_2$(8⁗) | 5.21 (*dd*, *J* = 17.6, 1.4) 5.06 (*dd*, *J* = 10.9, 1.4) | 112.2 | C(6⁗) |
| Me(9⁗) | 1.80 (*s*) | 12.0 | C(1⁗), C(2⁗), C(3⁗) |
| Me(10⁗) | 1.23 (*s*) | 27.5 | C(5⁗), C(6⁗), C(7⁗) |

[a]) All $\delta$(H) and $\delta$(C) assignments are based on 2D-NMR (COSY, HSQC, HMBC) spectral data. [b]) Recorded at 600 MHz. [c]) Recorded at 150 MHz). [d]) Signal pattern unclear due to overlapping.




**3**

Figure. *Significant HMBCs* (H → C) *for* **3**

olefinic H-atom at $\delta$ 6.80, three olefinic-H-atom signals as an *ABX* system at $\delta$ 5.86 (*dd*, $J = 17.6$, 10.9 Hz), 5.21 (*dd*, $J = 17.6$, 1.4 Hz), and 5.06 (*dd*, $J = 10.9$, 1.4 Hz), a pair of nonequivalent $CH_2$ resonances at $\delta$ 2.13–2.20 (*m*, 2 H) and 1.52–1.56 (*m*, 2 H), as well as two Me signals at $\delta$ 1.80 (*s*) and 1.23 (*s*), corresponding to a menthiafoloyl substituent[2]) [23][24]. The configuration at the quaternary C(6'''') atom was not determined due to the low yield of compound **3**. The location of the menthiafoloyl unit was found be the OH–C(6′) of glucose due to the downfield shifts for C(6′) ($\delta$ 64.1) and $CH_2$(6′) ($\delta$ 4.26) and upfield shift for C(5′) ($\delta$ 73.0). Finally, the HMBC experiment permitted to clarify the interfragmental connectivities (*Figure*). Thus, cross-peaks were observed between H–C(1′) ($\delta$ 4.43) of glucose and C($\alpha$) ($\delta$ 72.4) of the (dihydroxyphenyl)ethyl moiety, between H–C(1″) ($\delta$ 5.22) of rhamnose and C(3′) ($\delta$ 81.2) of glucose, between H–C(4′) ($\delta$ 5.04) of glucose and the C=O C-atom ($\delta$ 168.0) of the caffeoyl unit, and between $CH_2$(6′) ($\delta$ 4.26) of glucose and the C=O C-atom (C(1''''), $\delta$ 169.1) of the menthiafoloyl moiety.

Galypumoside A and B are the second and third examples of a phenylethyl glycoside obtained from the genus *Globularia*, bearing two aromatic acyl units; the first example was the globusintenoside isolated from *G. sintenisii* [15]. Galypumoside C is an unusual phenylethyl glycoside containing a monoterpenoid acid in its structure. To the best of our knowledge, it is the first representative of this type of phenylethyl glycoside derivatives. Alpinoside, lytanthosalin, syringaresinol 4′-*O*-$\beta$-glucopyranoside and 6-hydroxyluteolin 7-*O*-sophoroside are also being reported for the first time from *G. alypum*. Recent studies have reported the occurrence of phenylethyl, flavonoid, and iridoid glycosides in Moroccan *G. alypum* [9][10]. A new 7-chlorinated iridoid glucoside, a new phenylethyl glycoside (6′-*O*-coumaroyl-1′-*O*-[2-(3,4-dihydroxyphenyl)ethyl]-$\beta$-D-glucopyranoside), as well as new flavon glycosides have been reported in these studies. Regarding iridoid glucosides, no chlorinated iridoids were isolated from the eight *Globularia* species studied up to date by our group [11–15]. On the other hand, the 8,9-unsaturated iridoid glucoside, alpinoside, which was isolated from most of the *Globularia* species growing in the flora of Turkey including *G. alypum*, has not been reported from Moroccan species. Concerning the phenylethyl glycosides, although the phenylethyl glycoside composition of both Turkish and Moroccan *G. alypum* seems to be different, it is interesting to note that the phenylethyl glycosides obtained from both materials contain a *p*-coumaroyl unit, which has not been encountered in other *Globularia* species investigated so far. Thus, the occurrence of the *p*-coumaroyl-bearing phenylethyl glycosides could be of taxonomic significance for the title plant. The

differences of the secondary metabolite profiles of *G. alypum* collected from different countries may arise from the geographical conditions.

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## Experimental Part

*General.* TLC: precoated silica gel *60* $F_{254}$ (*Merck*) aluminium plates; eluents $CHCl_3$/MeOH/$H_2O$ 80:20:1, 70:30:3, and 61:32:7, and AcOEt/MeOH/$H_2O$ 20:2:1; visualization by spraying with 1% vanillin/$H_2SO_4$ soln. followed by heating at 105° for 2–3 min. Column chromatography (CC): silica gel *60* (0.063–0.200 mm; *Merck*, Darmstadt) and *Sephadex LH-20* (*Fluka*). Vacuum liquid chromatography (VLC): *LiChroprep* $C_{18}$ (*Merck*). Medium-pressure liquid chromatography (MPLC): *Lewa* M5 pump, *LKB-17000-Minirac* fraction collector, *Rheodyne* injector, *Büchi* columns (3 × 24 cm and 2.6 × 46 cm, *LiChroprep* $C_{18}$; *Merck*) and *Combi Flash Companion* (*Isco*), *Redi step* column (*LiChroprep* $C_{18}$, 4 × 23 cm; *Teledyne Isco*). Semi-prep. high-performance liquid chromatography (HPLC): *Agilent-1100* system (*Agilent*, Palo Alto, CA, USA), equipped with a $C_{18}$ *μ-Bondapack* column (7.8 × 300 mm, 10 μm; *Waters Spherisorb S5ODS2*), a binary pump (*G-1312*), a *Rheodyne* injector (20 μl loop; *G-1328A*), a degasser (*G-1322A*), and a DAD analyzer (*G-1315A*). Optical rotations: *Jasco-DIP-1000* polarimeter. UV Spectra: *HP-Agilent-8453* spectrophotometer; $\lambda_{max}$ in nm. IR Spectra (KBr): *Perkin-Elmer-2000* FT-IR spectrometer; in $cm^{-1}$. NMR Spectra: *Bruker-AMX-600* instruments (600 ($^1H$) and 150 MHz ($^{13}C$)) with XWIN NMR 3.6 software package; $\delta$ in ppm, $J$ in Hz. HR-MALDI-MS: *Voyager DE* in MeOH; positive-ion mode; in $m/z$.

*Plant Material. G. alypum* L. was collected from İzmir, between Çeşme and Urla, West Anatolia, Turkey, in April 2004. A voucher specimen (IZEF 830) has been deposited with the Herbarium of the Faculty of Pharmacy, Ege University, İzmir, Turkey.

*Extraction and Isolation.* The air-dried and powdered leaves of *G. alypum* (350 g) were extracted twice with MeOH (2 × 2 l, 4 h) at 45°. The combined MeOH extracts were concentrated (85 g, yield 24%). The MeOH extract was suspended in $H_2O$ (150 ml), and partitioned with $CHCl_3$ (4 × 150 ml). The $H_2O$ layer provided 75 g of an extract. An aliquot of the $H_2O$ extract (45 g) was separated by VLC (*LiChroprep* $C_{18}$ (120 g), $H_2O$ (300 ml), then 20 → 100% MeOH/$H_2O$ in steps of 20% of MeOH (each 300 ml)): *Frs. A–H*. *Fr. B* (2.44 g) was subjected to MPLC (*LiChroprep* $C_{18}$, 2.6 × 46 cm column, 0 → 60% MeOH/$H_2O$ in steps of 5% of MeOH (each 200 ml)): *catalpol* (5 mg), *alpinoside* (8 mg), *syringin* (49 mg), *globularin* (310 mg), and *Fr.* $B_4$ and $B_5$. Likewise *Fr.* $B_4$ (200 mg) was applied to MPLC ($C_{18}$, 3 × 24 cm column, 15 → 40% MeOH/$H_2O$ in steps of 5% of MeOH (each 200 ml)): *globularimin* (15 mg) and *Fr.* $B_{4b}$ and $B_{4c}$. Separation of *Fr.* $B_{4b}$ (60 mg) by CCs (*Sephadex LH-20* (20 g), MeOH (200 ml), then $SiO_2$ (5 g), $CH_2Cl_2$/MeOH/$H_2O$ 80:20:1 and 61:32:7 (both 100 ml)) gave *calceolarioside A* (9 mg) and *6-hydroxyluteolin 7*-O-*sophoroside* (3 mg). *Globularicisin* (9 mg) and *verbascoside* (43 mg) were isolated from *Fr.* $B_{4c}$ (90 mg) by CC ($SiO_2$ (13 g), $CH_2Cl_2$/MeOH/$H_2O$ 90:10:1, 80:20:1, and 75:25:2 (each 100 ml)). *Fr.* $B_5$ (384 mg) was further submitted to CC ($SiO_2$ (38 g), $CHCl_3$/MeOH/$H_2O$ 90:10:1, 85:15:1, 80:20:1, 80:20:2, and 70:30:3 (each 100 ml)): *syringaresinol 4′*-O-*β*-D-*glucopyranoside* (4 mg), *globularin* (170 mg), and *globularidin* (9 mg). *Globularinin* (8 mg) was isolated from *Fr. D* (2.0 g) by successive chromatographic methods: MPLC ($C_{18}$, 2.6 × 46 cm column, 15 → 70% MeOH/$H_2O$ in steps of 5% of MeOH (each 200 ml)) and CC ($SiO_2$ (12 g), $CHCl_3$/MeOH/$H_2O$ 85:15:1, 80:20:2, 70:30:3, and 61:32:7 (each 100 ml)). *Fr. F* (1.1 g) was separated by MPLC ($C_{18}$, 4 × 23 cm column, 30–75% MeOH/$H_2O$ gradient): *globularin* (30 mg) and *Frs.* $F_{2-7}$. *Fr.* $F_5$ (150 mg) was applied to CC ($SiO_2$ (18 g), $CH_2Cl_2$/MeOH/$H_2O$ 90:10:1, 80:20:2, and 70:30:3 (each 100 ml)): *lytanthosalin* (7 mg). *Fr.* $F_6$ (120 mg) likewise was subjected to CC ($SiO_2$ (15 g), $CH_2Cl_2$/MeOH/$H_2O$ 80:20:2 and 70:30:3 (both 100 ml)): mixture of *galypumosides A* and *B* (**1/2**; 10 mg) exhibiting a single spot on TLC ($CH_2Cl_2$/MeOH/$H_2O$ 80:20:2; $R_f$ 0.23). Separation of **1/2** (each 2 mg) was hardly achieved by HPLC ($C_{18}$, 7.8 × 300 mm column, 35 → 40% MeCN/$H_2O$ for 20 min): $t_R$ 12.39 (**1**) and 12.66 (**2**). *Fr.* $F_7$ (90 mg) was separated by CC ($SiO_2$ (12 g), $CH_2Cl_2$/MeOH/$H_2O$ 90:10:1 and 80:20:2 (both 100 ml)): *galypumoside C* (**3**; 7 mg).

*Galypumoside A* (=*2-(3,4-Dihydroxyphenyl)ethyl 3*-O-*(6-Deoxy-α*-L-*mannopyranosyl)-β*-D-*glucopyranoside 4-[(2*E*)-3-(3,4-Dihydroxyphenyl)prop-2-enoate] 6-[(2*E*)-3-(4-Hydroxyphenyl)prop-2-enoate]*; **1**): Amorphous powder. $[\alpha]_D^{24} = -72.0$ ($c = 0.10$, MeOH). UV (MeOH): 222 (*sh*, 4.36), 289 (*sh*, 4.22), 319 (4.35). IR (KBr): 3435, 2923, 2852, 1739, 1630, 1594, 1384. $^1$H- and $^{13}$C-NMR: *Table 1*. HR-MALDI-MS: 793.2327 ($[M+Na]^+$, $C_{38}H_{42}NaO_{17}^+$; calc. 793.2320).

*Galypumoside B* (=*2-(3,4-Dihydroxyphenyl)ethyl 3*-O-*(6-Deoxy-α*-L-*mannopyranosyl)-β*-D-*glucopyranoside 4-[(2*E*)-3-(3,4-Dihydroxyphenyl)prop-2-enoate] 6-[(2*E*)-3-(4-Hydroxy-3-methoxyphenyl)-prop-2-enoate]*; **2**): Amorphous powder. $[\alpha]_D^{24} = -75.0$ ($c = 0.10$, MeOH). UV (MeOH): 221 (*sh*, 4.43), 289 (*sh*, 4.27), 327 (4.37). IR (KBr): 3435, 2923, 2853, 1739, 1631, 1602, 1454, 1384. $^1$H- and $^{13}$C-NMR: *Table 1*. HR-MALDI-MS: 823.2431 ($[M+Na]^+$, $C_{39}H_{44}NaO_{18}^+$; calc. 823.2425).

*Galypumoside C* (=*2-(3,4-Dihydroxyphenyl)ethyl 3*-O-*(6-Deoxy-α*-L-*mannopyranosyl-β*-D-*glucopyranoside 4-[(2*E*)-3-(3,4-Dihydroxyphenyl)prop-2-enoate] 6-[6-Hydroxy-2,6-dimethylocta-2,7-dienoate]*; **3**): Amorphous powder. $[\alpha]_D^{24} = -85.0$ ($c = 0.30$, MeOH). UV (MeOH): 221 (*sh*, 4.70), 291 (*sh*, 4.13), 329 (4.22). IR (KBr): 3435, 2922, 2851, 1699, 1630, 1609, 1522, 1384. $^1$H- and $^{13}$C-NMR: *Table 2*. HR-MALDI-MS: 813.3056 ($[M+Na]^+$, $C_{39}H_{50}NaO_{17}^+$; calc. 813.3048).

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