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FLAVONE *O*- AND *C*-GLYCOSIDES FROM *SETARIA ITALICA*

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Key Word Index—*Setaria italica*; Poaceae; green foxtail millet; flavone glycosides; *C*-glycosylflavones; acyl *C*-glycosylflavones.

Abstract—A chemical investigation of the leaves of *Setaria italica* yielded six known *O*-glycosylflavones and 10 *C*-glycosylflavones including the new compounds scoparin 2″-*O*-xyloside and scoparin 2″-*O*-glucoside, and six new acylated *C*-glycosylflavones, five of which were at least partly elucidated: orientin 6″-*O*-(*E*)-feruloyl-2″-*O*-xyloside, orientin X″-*O*-(*E*)-feruloyl-2″-*O*-glucoside, vitexin X″-*O*-(*E*)-feruloyl-2″-*O*-xyloside, vitexin X″-*O*-(*E*)-feruloyl-2″-*O*-glucoside and vitexin X″-*O*-(*E*)-sinapoyl-2″-*O*-xyloside.

INTRODUCTION

The use of micromolecular biology to understand the natural organization of the graminean specific complex of *Setaria italica* [1] led us to isolate and identify 22 out of the 40 flavonoid compounds present in this species. Previous investigations in the genus *Setaria* [2–4], mainly on the basis of R_f values and spots colours, mentioned the occurrence of vitexin, orientin, luteolin, apigenin and tricin as 'the most characteristic flavones' [4], of isoorientin [2], of 'flavone *C*-glycosides as the major flavonoids' [3], and of flavonols 'like kaempferol, quercetin and myricetin' [4]. The present work deals with the structural elucidation of six *O*-glycosylflavones and 16 *C*-glycosylflavones.

RESULTS AND DISCUSSION

The 22 flavonoid glycosides isolated from *Setaria italica* are listed in Table 1.

Apigenin derivatives

Compounds **7b**, **10**, **11** and **16** showed the same UV spectra and diagnostic shifts [5] as apigenin with free 5, 7 and 4′-hydroxyl groups for the three former and only 5 and 4′-hydroxyl groups in the last one. In the three former compounds acid hydrolysis yielded vitexin (identified by co-chromatography with an authentic specimen) accompanied by small amounts of its Wessely–Moser isomer. Comparison of R_f values before and after hydrolysis led to the identification of **7b** as vitexin, **10** and **11** as its *O*-glycosylderivatives and GC of the TMS derivatives of the sugar moieties from **10** and **11** characterized these as xylose and glucose, respectively. Acid hydrolysis of **16** gave besides apigenin (co-chromatography with an authentic standard), a blue fluorescent compound identified as *p*-coumaric acid by GC of its TMS derivative and two sugars identified in the same way as glucose and rhamnose. The chromatographic behaviour of **16** suggested a di-*O*-glycosidic structure. A kinetic study using mild acid hydrolysis (with 0.1 M HCl) showed rhamnose to be the terminal sugar. Compound **16** is therefore apigenin 7-(*p*-coumarylrutinoside).

The mass spectra of their PM derivatives confirmed **7b** to be vitexin, and showed **10** and **11** to be a vitexin X″-*O*-pentoside and a vitexin X″-*O*-hexoside, respectively (according to the observed fragmentation pattern which was characteristic for an apigenin 8-*C*-hexosyl-X″-*O*-glycoside: $[SO]^+ > [S]^+$ and $[j]^+$ as base peak in both cases [6]). Moreover, the striking similarity between the fragmentation patterns of permethylated **10** and vitexin 2″-*O*-xyloside [6] suggested a 2″-*O*-pentosyl linkage in **10**.

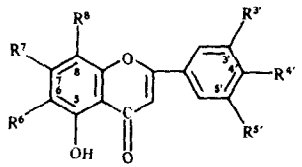
EIMS of hydrolysed permethylated **11** gave a i, j, k and l peak pattern, which was in agreement with the hypothesis of a free 2″-hydroxyl group and eliminated any suggestion of a free 6″-hydroxyl group [7]. FAB-CAD-MIKE analysis [8] of underivatized **10** and **11** gave a $[M-H-120]^-$ daughter ion characteristic of the 2″-linkage of the second sugar residue. These data proved **10** and **11** to be vitexin 2″-*O*-xyloside and vitexin 2″-*O*-glucoside, respectively. Final confirmation of the structure of **10** was obtained from its ^{13}C NMR spectrum.

Luteolin derivatives

Compounds **3a**, **3b**, **4**, **5** and **8** [1] were shown to be luteolin derivatives from UV spectral data [5]; **3a**, **3b**, **4** and **5** gave evidence for free hydroxyl groups in the 5, 7, 3′ and 4′ positions but **8** lacked a free 7-hydroxyl. Acid hydrolysis of **3a**, **3b**, **4** and **5** gave a mixture of orientin and isoorientin (identified by co-chromatography with authentic samples). Comparison of R_f values before and after hydrolysis showed **3a** and **3b** to be orientin and isoorientin respectively, **4** and **5** being their *O*-glycosylderivatives. GC of TMS derivatives of the sugar residues from **4** and **5** identified them as xylose and glucose, respectively. Compound **8** was characterized by complete and partial acid hydrolysis as luteolin 7-rutinoside.

Mass spectra (EIMS) of their permethylated derivatives confirmed **3a** and **3b** as orientin and isoorientin, and showed **4** and **5** to be an orientin X″-*O*-pentoside and an

Table 1. Free and acylated flavone glycosides and C-glycosylflavones from *Setaria italica*



Compound	Number on TLC (1)	$R^{3'}$	$R^{4'}$	$R^{5'}$	R^{6}	R^{7}	R^{8}
Apigenin 7-(*p*-coumarylrutinoside)	**16**	H	OH	H	H	*O*rut*p*coum	H
Luteolin 7-rutinoside	**8**	OH	OH	H	H	*O*rut	H
Chrysoeriol 7-glucoside	**22**	OMe	OH	H	H	*O*glc	H
Chrysoeriol 7-rutinoside	**20**	OMe	OH	H	H	*O*rut	H
Tricin 7-glucoside	**23**	OMe	OH	OMe	H	*O*glc	H
Tricin 7-rutinoside	**21**	OMe	OH	OMe	H	*O*rut	H
Vitexin	**7b**	H	OH	H	H	OH	β-D-glc
Vitexin 2″-*O*-xyloside	**10**	H	OH	H	H	OH	xyl(1‴→2″)glc
Vitexin 2″-*O*-glucoside	**11**	H	OH	H	H	OH	glc(1‴→2″)glc
Vitexin X″-*O*(*E*)-ferulyl-2″-*O*-xyloside	**12**	H	OH	H	H	OH	xyl(1‴→2″)glc-X″-fer
Vitexin X″-*O*(*E*)-sinapyl-2″-*O*-xyloside	**15**	H	OH	H	H	OH	xyl(1‴→2″)glc-X″-sin
Vitexin 2″-*O*-xyloside polyacylated	**1**	H	OH	H	H	OH	xyl(1‴→2″)glc + acyls
Vitexin X″-*O*(*E*)-ferulyl-2″-*O*-glucoside	**13a**	H	OH	H	H	OH	glc(1‴→2″)glc-X″-fer
Isoorientin	**3b**	OH	OH	H	β-D-glc	OH	H
Orientin	**3a**	OH	OH	H	H	OH	β-D-glc
Orientin 2″-*O*-xyloside	**4**	OH	OH	H	H	OH	xyl(1‴→2″)glc
Orientin 2″-*O*-glucoside	**5**	OH	OH	H	H	OH	glc(1‴→2″)glc
Orientin 6″-*O*(*E*)-ferulyl-2″-*O*-xyloside	**6**	OH	OH	H	H	OH	xyl(1‴→2″)glc-6″-fer
Orientin X″-*O*(*E*)-ferulyl-2″-*O*-glucoside	**9**	OH	OH	H	H	OH	glc(1‴→2″)glc-X″-fer
Scoparin	**13b**	OMe	OH	H	H	OH	β-D-glc
Scoparin 2″-*O*-xyloside	**17**	OMe	OH	H	H	OH	xyl(1‴→2″)glc
Scoparin 2″-*O*-glucoside	**18**	OMe	OH	H	H	OH	glc(1‴→2″)glc

rut, Rutinose; glc, glucose; xyl, xylose; fer, ferulyl; sin, sinapyl; *p*-coum, *p*-coumaryl.

orientin X''-*O*-hexoside, respectively (according to the MS pattern: $[SO]^+ > [S]^+$ and $[j]^+$ as base peak in both cases [6]). EIMS of the hydrolysis product of permethylated **5** gave a i, j, k and l peak pattern superimposable on a similar derivative from **11** suggesting the presence of a free 2''-hydroxyl. Finally, the characteristic features of their MIKE spectra allowed the assignment of the structures orientin 2''-*O*-xyloside and orientin 2''-*O*-glucoside to **4** and **5**, respectively.

Methylated luteolin derivatives

The UV spectral data of compounds **13b**, **17**, **18**, **20** and **22** agreed with a *C*-substituted chrysoeriol for the first three and a 7-*O*-substituted chrysoeriol for **20** and **22**. Repeating the same stages of experimental procedure as for the previous compounds [comparison of chromatographic behaviour between the natural compound and its acid hydrolysis product(s), co-chromatographic identification with authentic samples and sugar analysis by GC] led us to the following conclusions: **20** and **22** were chrysoeriol 7-rutinoside and chrysoeriol 7-glucoside, respectively; **13b** was identical with scoparin, **17** and **18** being its X''-*O*-xylosyl and X''-*O*-glucosyl derivatives, respectively.

The data available from FAB-MIKE analysis of **13b**, **17** and **18** corroborated the 8-linkage of the *C*-sugar on the aglycone and suggested a 2''-linkage of the *O*-sugar: therefore **17** and **18** are scoparin 2''-*O*-xyloside and scoparin 2''-*O*-glucoside, respectively, which are two new compounds for the literature [9].

Tricin derivatives

Compounds **21** and **23** showed the UV spectrum and diagnostic shifts of a 7-*O*-substituted tricin [5]. From chromatographic and acid hydrolysis data, the structures of tricin 7-rutinoside and tricin 7-glucoside could be assigned to **21** and **23**, respectively.

Acylated C-glycosylflavones

The distribution of flavonoids among individuals of *S. italica* showed a striking correlation between the yields of **6** and **4**, **9** and **5**, **12** and **10**, **13a** and **11** [1]: this suggested a derivative relationship between members of each pair. The increase (of *ca* 50%) of band I in the UV spectra of the first number of each couple indicated the presence of an additional chromophore; indeed the slight increase of R_f in aqueous acetic acid, when compared to that of the supposed basic compound, favours the hypothesis of an esterification by a phenolic acid. In all cases alkaline hydrolysis yielded ferulic acid and the expected basic flavonoid. According to the diagnostic shifts, this acylation took place on the sugar residue.

FAB-MIKE MS of the free compounds **6**, **9**, **12** and **13a**, all gave a daughter ion $[M-H-(O-\text{sugar}-H_2O)-H_2O]^-$ which indicated that the ferulic acid was ester linked to the *C*-sugar and corroborated their identification as orientin X''-*O*-(*E*)-feruloyl-2''-*O*-xyloside, orientin X''-*O*-(*E*)-feruloyl-2''-*O*-glucoside, vitexin X''-*O*-(*E*)-feruloyl-2''-*O*-xyloside and vitexin X''-*O*-(*E*)-feruloyl-2''-*O*-glucoside, respectively. Further, ^{13}C NMR of **6** confirmed the 2''-*O*-linkage of the xylosyl residue and indicated, from the typical shifts of the C-5'' and C-6'' signals [10], acylation at the C-6'' of the glucosyl residue.

Similarly **15** was identified as vitexin X''-*O*-(*E*)-sinapyl-2''-*O*-xyloside. On the same basis, compound **1** is esterified by sinapic and ferulic acids and a third *p*-nitraniline reacting substituent not yet identified [11]. When permethylated, **15** and **1** gave derivatives which showed the same R_f and gave the same EIMS as that obtained from **10**, a fact which corroborates the proposed structures. These six acyl *C*-glycosylflavonoids are new in the literature [9].

EXPERIMENTAL

Plant material. Setaria italica subsp. *viridis* (L.) Thellung was collected in August 1984 in several spontaneous stations near 73-Ruffieux, France.

Isolation procedure. 1 kg air-dried leaf material was extracted ×3 with EtOH–H_2O (7:3) and the concd extract taken up in boiling H_2O. Lipids were removed by PE and the flavonoids transferred from the H_2O extract to *n*-BuOH, which was separated on polyamide CC 6.6 with increasing concns of MeOH in toluene into 4 large fractions broadly corresponding to flavonoid aglycones, methylated flavone glycosides, glycosides of apigenin and luteolin respectively. By PC on Whatman no 3 (of the two last fractions) and polyamide TLC 6.6, 22 compounds were isolated. Purity of these compounds was monitored on TLC and HPLC. Chromatographic systems: PC, 5% HOAc; cellulose TLC, 15% HOAc (system **a**); polyamide TLC 6.6, toluene–MeOH–MeCOEt–*n*-BuOH (300:200:150:3) (system **b**) and H_2O–*n*-BuOH–Me_2CO–dioxan (22:3:2:1); reversed phase (nucleosil 5 μ C18) HPLC, solvents A 2% aq. HOAc, B 80% MeCN and 2% HOAc in H_2O-stepwise gradient: from 15 to 20% of B in A in 15 min, isocratic 20% B for 15 min, from 20 to 27% in 15 min, isocratic 27% B for 15 min, from 27 to 34% in 15 min, from 34 to 40% in 25 min, flow rate 0.7 ml/min, detection 340 nm.

Acid hydrolysis. The pure compounds were treated with 2 M HCl at 100° for 1 hr. Hydrolysates were extracted with EtOAc (aglycones), then *n*-BuOH (*C*-glycosides). Sugars were identified in aqueous residue by GC after silylation with MeCN and BSTFA+1% TMCS and separation on capillary column CPSil 5.

Alkaline hydrolysis. The acylated glycosides were added to 2 M NaOH and the mixture left under N_2 for 2 hr at room temp. in darkness then neutralized on ice with 2 M HCl. Hydrolysates were extracted with Et_2O (acids), then *n*-BuOH (deacylated glycosides). Phenolic acids were identified by GC as described for sugars above.

Permethylation. Preparation of PM derivatives was achieved as previously described [7]. Purification was carried out with the chromatographic systems silica gel TLC, $CHCl_3$–EtOAc–Me_2CO [5:4:1 (system **c**) and 5:1:4 (system **d**)].

Apigenin derivatives

Compound **7b** (vitexin). R_f 0.13 (system **a**), 0.34 (system **b**). R_t min: 26.5. EIMS of PM ether, 70 eV, *m/z* (rel. int.): 530 $[M]^+$ (66), 501 $[M-29]^+$ (1), 397 $[M-133]^+$ (2), 369 $[M-161]^+$ (8), 367 $[M-163]^+$ (2), 355 $[M-175]^+$ (100), 341 $[M-189]^+$ (16), 339 $[M-191]^+$ (13), 325 $[M-205]^+$ (8), 311 $[M-219]^+$ (8). PM vitexin, R_f 0.26 (system **c**).

Compound **10** (*vitexin 2''-O-xyloside*). R_f 0.62 (system a), 0.30 (system **b**). R_t min: 34.2. UV λ_{max}^{MeOH} nm: 269, 300sh, 329; +NaOAc 282, 300sh, 381; +NaOAc+H_3BO_3 280, 300sh, 380; +$AlCl_3$ 279, 303, 346, 382; +$AlCl_3$+HCl 278, 302, 342, 380; +NaOH 279, 328, 395. EIMS of PM ether, 70 eV, *m/z* (rel. int.): 690 $[M]^+$ (5), 544 $[SOj]^+$ (4), 515 $[SO]^+$ (40), 499 $[S]^+$ (5), 467

$[S-32]^+$ (8), 355 $[i]^+$ (11), 341 $[j]^+$ (100), 325 $[k]^+$ (40), 311 (22), 309 (5). PM vitexin 2″-*O*-xyloside, R_f 0.35 (system **d**). FAB^-MS 563. FAB-MIKE MS of *m/z* 563, *m/z* (rel. int.): 443 $[M-H-120]^-$ (10), 413 $[M-H-(\text{pentose}-H_2O)-H_2O]^-$ (100). ^{13}C NMR (DMSO-d_6), δ ppm: aglycone: 181.9 (C-4), 163.6 (C-2), 163.1 (C-7), 161.1 (C-5), 160.4 (C-4′), 156.5 (C-9), 128.7 (C-2′, C-6′), 121.4 (C-1′), 115.8 (C-3′, C-5′), 103.6 (C-10), 103.4 (C-8), 102.2 (C-3), 98.1 (C-6); *C*-glucosyl: 81.6 (C-2″), 80.7 (C-5″), 78.2 (C-3″), 71.4 (C-1″), 70.0 (C-4″), 60.9 (C-6″); *O*-xylosyl: 105.6 (C-1‴), 75.7 (C-3‴), 73.5 (C-2‴), 69.2 (C-4‴), 65.3 (C-5‴).

Compound **11** (*vitexin 2″-O-glucoside*). R_f 0.62 (system a), 0.26 (system **b**). R_t min: 30.3; UV λ_{max}^{MeOH} nm: 269, 300sh, 330; +NaOAc 279, 300sh, 372; +NaOAc+H_3BO_3 279, 300sh, 375; +$AlCl_3$ 275, 302, 344, 384; +$AlCl_3$+HCl 276, 301, 342, 384; +NaOH 279, 330, 398. EIMS of PM ether, 70 eV, *m/z* (rel. int.): 734 $[M]^+$ (2), 719 $[M-15]^+$ (2), 703 $[M-31]^+$ (5), 544 $[SOj]^+$ (36), 515 $[SO]^+$ (90), 499 $[S]^+$ (29), 467 $[S-32]^+$ (20), 341 $[j]^+$ (100), 325 $[k]^+$ (45), 311 (40), 309 (4). PM vitexin 2″-*O*-glucoside, R_f 0.30 (system **d**). EIMS of hydrolysis product of PM **11**, 70 eV, *m/z* (rel. int.): 516 $[M]^+$ (100), 485 $[M-31]^+$ (3), 397 $[f]^+$ (8), 367 $[h]^+$ (12), 355 $[i]^+$ (49), 341 $[j]^+$ (69), 325 $[k]^+$ (32), 311 $[l]^+$ (22). FAB^-MS 593. FAB-MIKE MS of *m/z* 593, *m/z* (rel. int.): 575 $[M-H-H_2O]^-$ (30), 473 $[M-H-120]^-$ (10), 413 $[M-H-(\text{hexose}-H_2O)-H_2O]^-$ (100).

Compound **16** [*apigenin 7-(p-coumarylrutinoside)*]. R_f 0.35 (system **a**), 0.54 (system **b**). UV λ_{max}^{MeOH} nm: 268, 333; +$AlCl_3$ 273, 297, 346, 377; +$AlCl_3$+HCl 273, 297, 340, 379; +NaOH 270, 389.

Luteolin derivatives

Compound **3a** (*orientin*). R_f 0.08 (system **a**), 0.21 (system **b**). R_t min: 25.9. EIMS of PM ether, 70 eV, *m/z* (rel. int.): 560 $[M]^+$ (55), 427 $[M-133]^+$ (3), 399 $[M-161]^+$ (8), 397 $[M-163]^+$ (2), 385 $[M-175]^+$ (100), 371 $[M-189]^+$ (17), 369 $[M-191]^+$ (14), 355 $[M-205]^+$ (7), 341 $[M-219]^+$ (10). PM orientin, R_f 0.24 (system **c**).

Compound **3b** (*isoorientin*). R_f 0.24 (system **a**), 0.21 (system **b**). R_t min: 26.5. EIMS of PM ether, 70 eV, *m/z* (rel. int.): 560 $[M]^+$ (8), 546 $[M-14]^+$ (5), 545 $[M-15]^+$ (19), 531 $[M-29]^+$ (6), 530 $[M-30]^+$ (22), 529 $[M-31]^+$ (77), 513 $[M-47]^+$ (16), 457 $[M-103]^+$ (17), 427 $[M-133]^+$ (8), 399 $[M-161]^+$ (14), 397 $[M-163]^+$ (16), 385 $[M-175]^+$ (100), 371 $[M-189]^+$ (19), 369 $[M-191]^+$ (22), 355 $[M-205]^+$ (32), 341 $[M-219]^+$ (23). PM isoorientin, R_f 0.48 (system **c**).

Compound **4** (*orientin 2″-O-xyloside*). R_f 0.54 (system **a**), 0.19 (system **b**). R_t min: 26.5. UV λ_{max}^{MeOH} nm: 256, 269, 290sh, 348; +NaOAc 272, 324sh, 400; +NaOAc+H_3BO_3 268, 390; +$AlCl_3$ 276, 300sh, 336sh, 424; +$AlCl_3$+HCl 262sh, 272, 296sh, 356, 384; +NaOH 268, 278sh, 332sh, 404. EIMS of PM ether, 70 eV, *m/z* (rel. int.): 720 $[M]^+$ (4), 574 $[SOj]^+$ (5), 545 $[SO]^+$ (34), 529 $[S]^+$ (4), 497 $[S-32]^+$ (5), 385 $[i]^+$ (12), 371 $[j]^+$ (100), 355 $[k]^+$ (45), 341 (29), 339 (9), 311 (5). PM orientin X″-*O*-xyloside, R_f 0.33 (system **d**). FAB^-MS 579. FAB-MIKE MS of *m/z* 579, *m/z* (rel. int.): 561 $[M-H-H_2O]^-$ (15), 459 $[M-H-120]^-$ (100), 429 $[M-H-(\text{pentose}-H_2O)-H_2O]^-$ (60).

Compound **5** (*orientin 2″-O-glucoside*). R_f 0.54 (system **a**), 0.15 (system **b**). R_t min: 24.5. UV λ_{max}^{MeOH} nm: 256, 269, 292sh, 348; +NaOAc 272, 408; +NaOAc+H_3BO_3 269, 390; +$AlCl_3$ 273, 301sh, 332sh, 424; +$AlCl_3$+HCl 263sh, 275, 296sh, 355, 384; +NaOH 268, 274sh, 336sh, 407. EIMS of PM ether, 70 eV, *m/z* (rel. int.): 764 $[M]^+$ (1), 750 $[M-14]^+$ (1), 749 $[M-15]^+$ (1), 733 $[M-31]^+$ (2), 574 $[SOj]^+$ (18), 545 $[SO]^+$ (44), 529 $[S]^+$ (14), 497 $[S-32]^+$ (10), 371 $[j]^+$ (100), 355 $[k]^+$ (58), 341 (53), 339 (9), 311 (6). PM orientin 2″-*O*-glucoside, R_f 0.27 (system **d**). MS of hydrolysis product of PM **5**, 70 eV, *m/z* (rel. int.): 546 $[M]^+$ (100), 515 $[M-31]^+$ (4), 427 $[f]^+$ (6), 397 $[h]^+$ (10), 385 $[i]^+$ (50), 371 $[j]^+$ (59), 355 $[k]^+$ (27), 339 $[l]^+$ (20). FAB^-MS 609. FAB-MIKE MS of *m/z* 609, *m/z* (rel. int.): 591 $[M-H-H_2O]^-$ (55), 489 $[M-H-120]^-$ (100), 429 $[M-H-(\text{hexose}-H_2O)-H_2O]^-$ (40).

Compound **8** (*luteolin 7-rutinoside*). R_f 0.18 (system **a**), 0.32 (system **b**). R_t min: 43.6.

Chrysoeriol derivatives

Compound **13b** (*scoparin*). R_f 0.08 (system **a**), 0.55 (system **b**). R_t min: 43.1.

Compound **17** (*scoparin 2″-O-xyloside*). R_f 0.70 (system **a**), 0.57 (system **b**). R_t min: 43.5. UV λ_{max}^{MeOH} nm: 250sh, 253, 269, 342; +NaOAc 275, 320, 359; +NaOAc+H_3BO_3 254sh, 270, 346; +$AlCl_3$ 262sh, 274, 299sh, 361, 385; +$AlCl_3$+HCl 261, 274sh, 297sh, 353, 383sh; +NaOH 268, 279, 332sh, 405. FAB^-MS 593. FAB-MIKE MS of *m/z* 593, *m/z* (rel. int.): 575 $[M-H-H_2O]^-$ (20), 473 $[M-H-120]^-$ (20), 443 $[M-H-(\text{pentose}-H_2O)-H_2O]^-$ (100), 461 $[M-H-(\text{pentose}-H_2O)]^-$ (10).

Compound **18** (*scoparin 2″-O-glucoside*). R_f 0.70 (system **a**), 0.53 (system **b**). R_t min: 41.3. UV λ_{max}^{MeOH} nm: 250sh, 270, 342; +NaOAc 276, 320, 360; +NaOAc+H_3BO_3 256sh, 270, 344; +$AlCl_3$ 260sh, 275, 300sh, 363, 384; +$AlCl_3$+HCl 260, 276sh, 299sh, 353, 383sh; +NaOH 267, 279, 340sh, 408. FAB^-MS 623. FAB-MIKE MS of *m/z* 623, *m/z* (rel. int.): 605 $[M-H-H_2O]^-$ (28), 503 $[M-H-120]^-$ (28), 443 $[M-H-(\text{hexose}-H_2O)-H_2O]^-$ (100).

Compound **20** (*chrysoeriol 7-rutinoside*). R_f 0.27 (system **a**), 0.58 (system **b**). R_t min: 54.8.

Compound **22** (*chrysoeriol 7-glucoside*). R_f 0.09 (system **a**), 0.62 (system **b**).

Tricin derivatives

Compound **21** (*tricin 7-rutinoside*). R_f 0.18 (system **a**), 0.60 (system **b**). R_t min: 55.9.

Compound **23** (*tricin 7-glucoside*). R_f 0.05 (system **a**), 0.69 (system **b**). R_t min: 56.0.

Acylated derivatives

Compound **6** (*orientin 6″-O-(E)-ferulyl-2″-O-xyloside*). R_f 0.77 (system **a**), 0.47 (system **b**). R_t min: 84.9. UV λ_{max}^{MeOH} nm: 270, 300sh, 332; +NaOAc 273sh, 280, 326, 381sh; +NaOAc +H_3BO_3 262, 296, 332, 367sh; +$AlCl_3$ 276, 300sh, 327, 414; +$AlCl_3$+HCl 278, 298, 334, 383sh; +NaOH 266, 275sh, 308sh, 387. FAB^-MS 755. FAB-MIKE MS of *m/z* 755, *m/z* (rel. int.): 737 $[M-H-H_2O]^-$ (15), 623 $[M-H-\text{pentose}-H_2O]^-$ (20), 605 $[M-H-(\text{pentose}-H_2O)-H_2O]^-$ (100), 579 $[M-H-(\text{ferulyl}-H_2O)]^-$ (18), 561 $[M-H-\text{ferulyl}]^-$ (15), 459 $[M-H-120-(\text{ferulyl}-H_2O)]^-$ (18). ^{13}C NMR (DMSO-d_6), δ ppm: aglycone: 181.8 (C-4), 166.7 (C-2), 162.6 (C-7), 160.6 (C-5), 156.5 (C-9), 149.6 (C-4′), 145.9 (C-3′), 121.8 (C-1′), 118.7 (C-6′), 115.6 (C-5′), 113.9 (C-2′), 103.7 (C-10), 103.1 (C-8), 102.3 (C-3), 97.9 (C-6); *C*-glucosyl: 80.4 (C-2″), 78.3 (C-3″), 78.0 (C-5″), 71.5 (C-1″), 70.1 (C-4″), 63.9 (C-6″); *O*-xylosyl: 105.7 (C-1‴), 75.7 (C-3‴), 73.4 (C-2‴), 69.1 (C-4‴), 65.4 (C-5‴); ferulyl: 172.2 (C-9), 149.2 (C-4), 147.7 (C-3), 145.2 (C-8), 125.3 (C-1), 122.9 (C-6), 115.3 (C-7), 114.8 (C-5), 113.9 (C-2).

Compound **9** (*orientin X″-O-(E)-ferulyl-2″-O-glucoside*). R_f 0.75 (system **a**), 0.38 (system **b**). R_t min: 81.7. UV λ_{max}^{MeOH} nm: 270, 300sh, 332; +NaOAc 270sh, 280, 324, 385sh; +NaOAc +H_3BO_3 262, 296, 331, 368sh; +$AlCl_3$ 275, 296sh, 327, 424; +$AlCl_3$+HCl 278, 297, 334, 384sh; +NaOH 266, 280sh, 309sh, 387. FAB^-MS 785. FAB-MIKE MS of *m/z* 785, *m/z* (rel. int.): 767 $[M-H-H_2O]^-$ (10), 623 $[M-H-(\text{hexose}-H_2O)]^-$ (25), 605

$[M-H-(hexose-H_2O)-H_2O]^-$ (100), 591 $[M-H-$ferulyl$]^-$ (5), 489 $[M-H-120-(ferulyl-H_2O)]^-$ (30).

Compound **12** (*vitexin X″-O-(E)-ferulyl-2″-O-xyloside*). R_f 0.72 (system **a**), 0.54 (system **b**). R_t min: 90.8. UV λ_{max}^{MeOH} nm: 271, 303sh, 326; +NaOAc 280, 297, 315, 328sh, 381sh; +NaOAc +H_3BO_3 272, 285, 300sh, 326; +$AlCl_3$ 278, 304, 335, 381sh; +$AlCl_3$ + HCl 279, 303, 333, 381sh; +NaOH 279, 310sh, 337sh, 386. FAB^-MS 739. FAB-MIKE MS of *m/z* 739, *m/z* (rel. int.): 721 $[M-H-H_2O]^-$ (15), 607 $[M-H-(pentose-H_2O)]^-$ (25), 589 $[M-H-(pentose-H_2O)-H_2O]^-$ (100), 563 $[M-H-(ferulyl-H_2O)]^-$ (15), 545 $[M-H-$ferulyl$]^-$ (20), 473 $[M-H-90-(ferulyl-H_2O)]^-$ (7), 443 $[M-H-120-$(ferulyl $-H_2O)]^-$ (5).

Compound **13a** (*vitexin X″-O-(E)-ferulyl-2″-O-glucoside*). R_f 0.72 (system **a**), 0.53 (system **b**). R_t min: 87.0. UV λ_{max}^{MeOH} nm: 271, 300sh, 327; +NaOAc 281, 300sh, 313sh, 326sh, 379sh; +NaOAc +H_3BO_3 272, 282, 325; +$AlCl_3$ 278, 304, 335, 383sh; +$AlCl_3$ + HCl 280, 303, 334, 380sh; +NaOH 279, 310sh, 331sh, 386. FAB^-MS 769. FAB-MIKE MS of *m/z* 769, *m/z* (rel. int.): 751 $[M-H-H_2O]^-$, 607 $[M-H-(hexose-H_2O)]^-$, 593 $[M-H-(ferulyl-H_2O)]^-$, 589 $[M-H-(hexose-H_2O)-H_2O]^-$ (100), 575 $[M-H-$ferulyl$]^-$, 473 $[M-H-120-$(ferulyl $-H_2O)]^-$.

Compound **15** (*vitexin X″-O-(E)-sinapyl-2″-O-xyloside*). R_f 0.75 (system **a**), 0.56 (system **b**). R_t min: 88.9. UV λ_{max}^{MeOH} nm: 270, 328; +NaOAc 279, 310, 328sh, 380sh; +NaOAc+H_3BO_3 278, 307, 332sh, 372sh; +$AlCl_3$ 278, 305, 338, 386sh; +$AlCl_3$+HCl 278, 303, 335, 386sh; +NaOH 279, 334sh, 394. EIMS of PM ether, 70 eV, *m/z* (rel. int.): 690 $[M]^+$ (4), 544 $[SOj]^+$ (3), 515 $[SO]^+$ (25), 499 $[S]^+$ (3), 467 $[S-32]^+$ (5), 355 $[i]^+$ (10), 341 $[j]^+$ (100), 325 $[k]^+$ (36), 311 (23), 309 (6). PM **15**, R_f 0.35 (system **d**). FAB^-MS 769, FAB^+MS 771.

Compound **1** (*polyacylated vitexin 2″-O-xyloside*). R_f 0.73 (system **a**), 0.10 (system **b**). R_t min: 81.3. UV λ_{max}^{MeOH} nm: 269, 286sh, 330; +NaOAc 278, 300sh, 333, 386sh; +NaOAc +H_3BO_3 278, 301sh, 340; +$AlCl_3$ 278, 302, 345, 382; +$AlCl_3$ + HCl 277, 300, 342, 380; +NaOH 279, 329, 395. EIMS of PM ether, 70 eV, *m/z* (rel. int.): 690 $[M]^+$ (4), 544 $[SOj]^+$ (4), 515 $[SO]^+$ (37), 499 $[S]^+$ (4), 467 $[S-32]^+$ (7), 355 $[i]^+$ (12), 341 $[j]^+$ (100), 325 $[k]^+$ (41), 311 (24), 309 (6). PM **1**, R_f 0.35 (system **d**).

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