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Abbes Benmerache, Abdulmagid Alabdul Magid, Ahmed Kabouche, Dominique Harakat, Laurence Voutquenne-Nazabadioko, et al. 6'''- O -acetylisopinosin, a new C -glycosylflavone and known compounds from the aerial parts of *Cladanthus mixtus*. *Natural Product Research*, 2019, 34 (20), pp.2887-2893. 10.1080/14786419.2019.1596100 . hal-02430902

HAL Id: hal-02430902

<https://hal.univ-reims.fr/hal-02430902v1>

Submitted on 29 Oct 2021

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6'''-O-acetylisopinosin, a new C-glycosylflavone and known compounds from the aerial parts of *Cladanthus mixtus*

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Abstract

A new C-glycosylflavone, identified as 6''-O-acetylisopinosin (**1**), was isolated from the aerial parts of *Cladanthus mixtus* together with 30 known compounds. The structures of these compounds were established by interpretation of their spectral data, mainly UV, 1D and 2D NMR spectroscopic methods including (¹H and ¹³C NMR, COSY, ROESY, HSQC and HMBC experiments), ESI-MS, and by comparison with the literature data.

Keywords : *Cladanthus mixtus*, flavonoid, phenolic acid, quinic acid derivative.

1. Introduction

Cladanthus genus (Asteraceae), comprising about five species, is native to Europe in South-West Europe and the Mediterranean region (Oberprieler, 2002). *Cladanthus* species are used in traditional medicine for their expectorant, antitussive, antibroncholytic, antispasmodic, anthelmintic, carminative and diuretic properties (Bellakhdar, 1997). The major constituents reported from this genus are essential oils (Satrani et al. 2007; Aghraz et al. 2017), sesquiterpenoid lactones (Daniewski et al. 1993), coumarins and flavonoids (Aghraz et al. 2017). According to literature survey, the essential oil of *Cladanthus* has exhibited various biological properties, such as antioxidant, antimicrobial and insecticidal activities (Aghraz et al. 2017). *Cladanthus mixtus* (L.) Oberpr. & Vogt., synonym (*Ormenis mixta* subsp. *mixta*), is a traditional herbal medicine, widespread in Algeria, Morocco, Northern and Eastern part of the Mediterranean basin (Zrira et al. 2007; Hajjaj et al. 2016). This species is a tall spontaneous annual plant, 10–40 cm in height, with fragrant white-yellow flowers. It has been used in folk medicine to treat different ailments as a mild sedative, spasmolytic and antibacterial agent. Leaves and flowers infusion is used in Morocco as an anxiolytic and to rebalance the central nervous system. The essential oil showed significant antimicrobial (Satrani et al. 2007), anti-inflammatory, antinociceptive, antioxidant and antibacterial effects (Zrira et al. 2007; Hajjaj et al. 2016).

In the present study, we report, for the first time, the isolation and structural elucidation of a new *C*-glycosylflavone (**1**) and 30 known compounds from the aerial parts of *Cladanthus mixtus*.

2. Results and discussion

The phytochemical investigation from the *n*-butanol extract from the aerial parts of *C. mixtus* led to the isolation of a new *C*-glycosylflavone (**1**) together with 30 known compounds from which three *C*-glycosyl flavones, isospinosin (**2**) (Chen et al. 2000), apigenin-8-*C*- β -D-glucopyranosyl-(1 \rightarrow 2)- β -D-glucopyranoside (vitexin-2'''-*O*- β -D-glucopyranoside) (**3**) (Ferrerres et al. 2007), genkwanin-8-*C*- β -D-apiofuranosyl-(1 \rightarrow 2)- β -D-glucopyranoside (**4**) (Alabdul Magid et al. 2017), 14 flavonoids namely, apigenin (**5**) (Nacer et al. 2006), kaempferol (**6**) (Gangwal et al. 2010), luteolin (**7**) (Ode and Asuzu, 2014), luteolin 3'-*O*-methylether (chrysoeriol) (**8**) (Lagoune et al. 2008), luteolin 4'-*O*- β -D-glucopyranoside (**9**) (Krenn et al. 2003), luteolin 3'-*O*- β -D-glucopyranoside (**10**) (Jiang et al. 2010), quercetin 3-*O*- β -D-

glucopyranoside (isoquercitrin) (**11**) (Touafek et al. 2011), quercetin 3-*O*- β -D-rutinoside (**12**) (Bencheraiet et al. 2011), quercetin 3-*O*-methylether (isorhamnetin) (**13**) (Krenn et al. 2003), 6-hydroxyquercitrin-3-*O*- β -D-glucopyranoside (**14**), patulitrin (quercetagetin-6-methylether-7-*O*- β -D-glucopyranoside) (**15**) (Shahzadi and Shah, 2015), 6-hydroxykaempferol-7-*O*- β -D-glucopyranoside (**16**) (Valant-Vetschera et al. 2003), (2R,3R)-taxifolin 3-*O*- β -D-galactopyranoside (**17**), (2S,3S) taxifolin 3-*O*- β -D-glucopyranoside (isoglucodistylin) (**18**) (Sakushima et al. 2002), 5 phenolic acids namely *trans*-caffeic acid (**19**) (Bhatt, 2011), *trans*-ferulic acid (**20**) (Liao et al. 2014), *trans-p*-coumaric acid (**21**) (Liao et al. 2014), 4-hydroxybenzoic acid (*p*-hydroxybenzoic acid) (**22**) (Dhakai et al. 2008), protocatechuic acid (**23**) (Liao et al. 2014), one phenol glycoside, 2-hydroxy-5-(2-hydroxyethyl)phenyl- β -D-glucopyranoside (**24**) (Lu and Foo, 1999), a monoterpene glycoside, chamolol (**25**) (Zaiter et al. 2007), and 6 quinic acids (3-*O*-caffeoylquinic acid methylester (neochlorogenic acid methyl ester) (**26**) (Kozawa et al. 1983), 5-*O*-caffeoylquinic acid (**27**) (Liu et al. 2013), 1,5-di-*O*-caffeoylquinic acid (**28**) (Islam et al. 2002), 3,5-di-*O*-caffeoylquinic acid (**29**) (Liu et al. 2013), 3,5-di-*O*-caffeoylquinic acid methyl ester (**30**) (Liu et al. 2013), and 4,5-di-*O*-caffeoylquinic acid (**31**) (Liu et al. 2013).

Compound **1** was isolated as a yellow amorphous powder. The positive HR-ESI-MS spectrum (Fig. S2) of **1** gave a quasi-molecular ion peak $[M+Na]^+$ at m/z 673.1734 compatible with the molecular formula of $C_{30}H_{34}O_{16}$ (calcd for $C_{30}H_{34}O_{16}Na$, 673.1745). In the UV spectral analyses (Fig. S5), compound **1** gave a typical MeOH spectrum $\lambda_{max} = 208, 270, 326$ nm, of a flavone derivative. The 1H -NMR spectrum of **1** showed an A_2B_2 spin system at δ_H 6.98 (2H, *d*, $J = 8.7$ Hz, H-3',H-5') and 8.03 (2H, *d*, $J = 8.7$ Hz, H-2',H-6'), together with two singlets at δ_H 6.60 (H-3) and 6.48 (H-6), a sugar chain [two anomeric protons at δ_H 5.08 (*d*, $J = 7.7$ Hz) and 4.37 (*d*, $J = 7.8$ Hz)], a methoxy group at δ_H 3.93 (3H, *s*), and an acetyl group at δ_H 1.96 (3H, *s*). Analysis of ^{13}C -NMR spectrum (Table S1) and a combination of 1H - 1H -COSY, HSQC, and HMBC experiments suggested that **1** was an isospinosin (**2**) [5,4'-dihydroxy-7-methoxyflavone-8-*C*- β -D-glucopyranosyl(1 \rightarrow 2)- β -D-glucopyranoside] with an additional acetyl moiety (Chen et al. 2000). The exact location of the sugars, methoxy, and acetyl was revealed by the HMBC correlations observed between δ_H 3.93 (*s*, OMe) and δ_C 164.0 (C-7), the anomeric proton signal at δ_H 4.37 (H-1_{glcII}) and C-2_{glcI} (δ_C 78.1), the anomeric proton signal at δ_H 5.08 (H-1_{glcI}) and C-8 (δ_C 105.0), and the H₂-6_{glcII} (δ_H 3.94 and 3.64) with the carbon of the acetyl group (δ_C 171.4). Thus, compound **1** was identified as 5,4'-dihydroxy-7-methoxyflavone-8-*C*-(6'''-*O*-acetyl)- β -D-glucopyranosyl(1 \rightarrow 2)- β -D-glucopyranoside or 6'''-*O*-acetylisospinosin.

3. Experimental

3.1. General experimental procedures

1D and 2D NMR spectra were carried in CD₃OD on Bruker Avance DRX III 500 instruments (¹H at 500 MHz and ¹³C-*J*mod at 125 MHz), using standard Bruker microprograms. HR-ESI-MS experiments were performed using a Micromass Q-TOF micro-instrument (Manchester, UK). Optical rotations were determined in CD₃OD with a Perkin-Elmer 341 polarimeter. The UV spectra were obtained in methanol on a Shimadzu UV-2450 spectrophotometer, TLC were performed on pre-coated silica-gel 60 F₂₅₄ Merck and were visualized under UV light at 254 and 366 nm and by spraying the dried plates with 50% H₂SO₄, followed by heating. Column chromatography was carried out on Kieselgel 60 (63–200 mesh) or LiChroprep RP-18 (40–63 mm) Merck. Centrifugal Partition Extraction (CPE) was performed on a lab-scale column of 303 mL capacity (FCPE300, Rousselet Robatel Kromaton, Annonay, France). The CPE column was filled with the stationary phase at 200 rpm by using a KNAUER Preparative 1800 V7115 pump (Berlin, Germany), the rotating at 1200 rpm and a flow rate of 20 mL/min in the ascending mode. High Performance Flash chromatography was performed on a Grace Reveleris system equipped with dual UV and ELSD detection using Grace1 cartridges (Silica gel or RP-C18) and a flow rate of 30 mL/min. The chromatograms were monitored at 205, 225, 250, and 360 nm. An Armen instrument equipped with an AP 250/500 pump, ACC 250/500 sampler, and a Merck UV-detector K-2501 was used for preparative HPLC. A Lichrospher RP18 prepacked column (Merck 250x50mm, 12μm) was used with binary gradient eluent (H₂O and CH₃CN) and a flow rate of 40 mL/min; the chromatogram was monitored at λ 250 nm. Semi-prep. HPLC was performed on a Dionex apparatus equipped with an ASI-100 autosampler, an Ultimate 3000 pump, a diode array detector UVD 340S and a Chromeleon software. RP- C18 column (Phenomenex 250 -15 mm, Luna 5 m) was used for semi preparative HPLC with a binary gradient eluent (H₂O (pH 2.4 with TFA); MeCN) and a flow rate of 5 mL/min; the chromatogram was monitored at 205, 225, 250, and 360 nm.

3.2. Plant material

The aerial parts of *Cladanthus mixtus* (L.) Oberpr. & Vogt., were collected in April 2014 from Constantine (North Eastern Algerian). A voucher specimen (LOST.Cm.04.14) was deposited in the Herbarium of the laboratory of Therapeutic Substances (LOST), University freres

Mentouri-Constantine 1 and authenticated by Prof. Gérard De Bélair (University of Annaba, Algeria).

3.3. Extraction and isolation

The powdered dried aerial parts (1.5 kg) of *Cladanthus mixtus* were macerated in a hydromethanol solution (80%) (3×7 L, 24 h). The residue was filtered, concentrated, and then successively extracted with dichloromethane, ethyl acetate and *n*-butanol. The *n*-BuOH extract (14 g) was subjected to a column chromatography on polyamid SC6 (90 × 5.5 cm), using toluene–MeOH (100:0 to 0:100) as the eluting solvent to give 22 fractions (FI-FXXII), based on the TLC profiles. Fraction FV (53 mg) was further purified by semi-prep HPLC (20-60% MeCN) to produce compound **25** (*R*_t 16.66 min, 3 mg). Fractions FVI-IX (1.7 g) were combined and subjected to Flash chromatography over silica gel, eluted with a gradient system of CHCl₃-MeOH (20-80 % MeOH) to obtain 12 sub-fractions. Compound **19** (*R*_t 13.73 min, 1 mg) was obtained from sub-fraction VI-IX-3 (37 mg) by semi-prep HPLC (isocratic elution 30% MeCN), whereas compounds **21** (*R*_t 10.26 min, 5 mg) and **22** (*R*_t 9.28 min, 5 mg) were isolated from sub-fraction VI-IX-7 (150 mg) by semi-prep HPLC (isocratic elution 25% MeCN). Fractions X-XI (1.0 g) were combined and subjected to Flash chromatography over silica gel, eluted with a gradient system of CHCl₃-MeOH (10-50 % MeOH) to give 17 sub-fractions. Sub-fraction X-XI-3 (32 mg) was submitted to semi-prep HPLC (40-45% MeCN, 20 min) affording two compounds, **13** (*R*_t 13.84 min, 5 mg) and **6** (*R*_t 18.74 min, 3 mg). Sub-fraction X-XI-7 (18 mg) was further purified by semi-prep HPLC (20-50% MeCN, 20 min) affording three compounds, **20** (*R*_t 9.20 min, 2 mg), **5** (*R*_t 17.57 min, 2 mg), and **7** (*R*_t 18.52 min, 1 mg). Sub-fraction X-XI-15 (35 mg) was purified by semi-prep HPLC (40-45% MeCN, 20 min) leading two compounds, **23** (*R*_t 11.87 min, 1 mg) and **24** (*R*_t 13.73 min, 1 mg). Fraction XVIII (0.4 g) was subjected to preparative HPLC (15-35% CH₃CN in 60 min) to give 14 compounds, **1** (3 mg), **2** (3 mg), **3** (2 mg), **4** (6 mg), **9** (20 mg), **10** (2 mg), **11** (7 mg), **12** (3 mg), **14** (7 mg), **15** (3 mg), **16** (3 mg), **17** (10 mg), and **18** (4 mg). and **26** (3.1 mg). Fractions XXI-XXII (0.4 g) were combined and subjected to CPE fractionation using the biphasic solvent system composed of *n*-hexane-MeOH-EtOAc-H₂O (3:1:3:1, v/v) in the ascending mode (i.e. the upper *n*-hexane -MeOH-EtOAc phase was used as the mobile phase) affording three compounds, **8** (24 mg), **29** (2 mg), and **30** (2 mg), and 4 sub-fractions. Compounds **27** (5 mg), **28** (4 mg), and **31** (7 mg) were isolated from sub-fraction XXI-XXII-2 by semi-prep HPLC (isocratic elution 25% MeCN).

5,4'-dihydroxy-7-methoxyflavone-8-C-(6'''-O-acetyl)-β-D-glucopyranosyl(1→2)-β-D-glucopyranoside or 6'''-O-acetylisopinosin.

Yellow amorphous powder; $[\alpha]_{\text{D}}^{20}$ -15.8 (*c* 0.25, MeOH). UV (MeOH) λ_{max} : 208 nm ($\log \epsilon = 3.6$), 270 nm ($\log \epsilon = 1.5$), 326 nm ($\log \epsilon = 1.5$). ^1H NMR (500 MHz, CD_3OD): δ_{H} 8.03 (d, $J = 8.7$ Hz, H-6', H-2'), 6.98 (d, $J = 8.7$ Hz, H-5', H-3'), 6.60 (s, H-3), 6.48 (s, H-6), 5.08 (d, $J = 7.7$ Hz, H-1''), 4.37 (d, $J = 7.8$, H-1'''), 4.35 (t, $J = 9.9$, Hz, H-2''), 3.97 (dd, $J = 12.6$ -1.29 Hz, H-6''a), 3.94 (dd, $J = 13.0$ -3.3 Hz, H-6''b), 3.94 (s, 7-OCH₃), 3.80 (dd, $J = 12.6$ -5.8 Hz, H-6''b), 3.76 (t, $J = 9.9$ Hz, H-3''), 3.69 (t, $J = 9.4$, Hz, H-4''), 3.64 (dd, $J = 13.0$ -1.50 Hz, H-6'''a), 3.47 (m, H-5'''), 3.17 (t, $J = 9.1$ Hz, H-3'''), 3.08 (t, $J = 9.1$ Hz, H-4'''), 2.95 (m, H-5'''), 2.94 (t, $J = 9.1$ Hz, H-2'''), 1.96 (s, H-2'''). ^{13}C NMR (125 MHz, CD_3OD): δ_{C} 171.4 (C-1'''), 182.9 (C-4), 165.2 (C-2), 164.0 (C-7), 161.9 (C-5), 161.5 (C-4'), 156.0 (C-9), 128.8 (C-2', C-6'), 122.1 (C-1'), 115.7 (C-3', C-5'), 105.0 (C-8), 104.5 (C-10), 103.4 (C-1'''), 102.3 (C-3), 94.6 (C-6), 81.6 (C-5''), 79.0 (C-3''), 78.1 (C-2''), 76.2 (C-3'''), 74.2 (C-2'''), 73.4 (C-5'''), 72.1 (C-1''), 70.6 (C-4''), 69.0 (C-4'''), 62.5 (C-6'''), 61.5 (C-6''), 55.6 (7-OCH₃), 19.3 (C-2'''). HR-ESI-MS m/z 673.1734 [$\text{M}+\text{Na}$]⁺, (calcd for $\text{C}_{30}\text{H}_{34}\text{O}_{16}\text{Na}$; 673.1745).

4. Conclusions

The present phytochemical investigation resulted in the isolation of a new C-glycosylflavone and 30 known compounds from which 17 flavonoids, 6 quinic acids, and 5 phenol acids, from *Cladanthus* genus for the first time. Compound **25** (Chamolol) has already been isolated from *Matricaria chamomilla* L. (Zaiter et al. 2007). Compound **24** is reported for the first time in the Asteraceae, it was first isolated from *Osmanthus asiaticus* Nakai (Sugiyama et al. 1992) and grape pomace (Lu and Foo, 1999). Various flavonoids of *C. mixtus* have been characterized in the family Asteraceae, They are mostly represented by flavones, flavonols, 6-hydroxyflavonol derivatives. It is noteworthy that C-glycosylflavones, substituted at position 8, and the dihydroflavonols (**17** and **18**) are rarely isolated from plants, their occurrence being hitherto confined to a small number of species. Compound **4** is reported for the first time in the family Asteraceae, it was only isolated from *Secamone afzelii* (Alabdul Magid et al. 2017). The phenolic acids (**19-23**) and quinic acids (**26-31**) are all ubiquitous and have already been isolated in Asteraceae (Lin and Wang, 2002; Gobbo-Neto and Lopes, 2008).

Supplementary Information

HR-ESI-MS, UV, ¹H NMR, ¹³C NMR, COSY and HMBC spectra of compound **1**

Acknowledgements

The authors are grateful to the MESRS, Algeria for the Profas grant (836958E) to Mr Abbes Benmerache, to CNRS, Conseil Regional Champagne-Ardenne, Conseil General de la Marne, Ministry of Higher Education and Research (MESR, France) and to the PLANET CPER Project.

References

- Aghraz A, Wanner J, Schmidt E, Aitdra L, Aitsidibrahim M, Tabanca N, Ali A, Nafis A, Hassani L, Markouk M, Jirovetz L, Larhsini M. 2017. Chemical Composition, *in vitro* antioxidant, antimicrobial and insecticidal activities of essential oil from *Cladanthus arabicus*. J Essent Oil Bear Plants. 20:601-609.
- Alabdul Magid A, Yao-Kouassi PA, Apie GDP, Mairot C, Voutquenne-Nazabadioko L. 2016. New antioxidant flavonoids from the aerial parts of *Secamone afzelii*. Journal of antioxidant activity .1(2): 8-16.
- Bellakhdar J. 1997. La pharmacopée traditionnelle marocaine: Médecine arabe ancienne et savoirs populaires, Paris, Ibis Press, 766.
- Bencheraiet R, Kabouche A, Kabouche Z, Jay M. 2011. Flavonols of *Bupleurum plantagineum*. Chem Nat Compds. 47:814-815.
- Bhatt B. 2011. Chemical constituents of *Solanum xanthocarpum*. J Chem Pharma Res. 3:176-181.
- Cheng G, Bai Y, Zhao Y, Tao J, Liu Y, Tu G, Ma L, Liao N, Xu X. 2000. Flavonoids from *Ziziphus jujuba* Mill var. *spinosa*. Tetrahedron. 56:8915-8920.
- Daniewski W M, Danikiewicz W, Gumuka M, Pankowska E, Krajewski J, Grabarczyk H, Wichacz M. 1993. Sesquiterpenes of *Cladanthus arabicus*. Phytochemistry. 34:1639-1641,
- Dhakal RC, Rajbhandari M, Kalauni SK, Awale S, Gewali I MB. 2008/2009. Phytochemical Constituents of the Bark of *Vitex negundo* L. J Nepal Chem Soc. 23:89-92.
- Ferreres F, Gil-Izquierdo A, Andrade PB, Valentao P, Tomas-Barberan FA. 2007. Characterization of C-glycosyl flavones O-glycosylated by liquid chromatography tandem mass spectrometry. J Chromato A. 1161:214-223.
- Gangwal A, Parmar SK, Sheth NR. 2010. Triterpenoid, flavonoids and sterols from *Lagenaria siceraria* fruits. Pharm Lett. 2(1):307-317.

- Gobbo-Neto L, Lopes NP. 2008. Online identification of chlorogenic acids, sesquiterpene lactones, and flavonoids in the Brazilian *arnica* *Lychnophora ericoides* Mart. (Asteraceae) leaves by HPLC-DAD-MS and HPLC-DAD-MS/MS and a validated HPLC-DAD method for their simultaneous analysis, J Agric Food Chem. 56: 1193-1204.
- Hajjaj G, Bahlouli A, Tajani M, Cherrah Y, Zellou A. 2016. *In vivo* anti-nociceptive and anti-inflammatory properties of *Ormenis mixta* L. fom Morocco. Int J Pharm Pharma Sci. 8:49-52.
- Islam MS, Yoshimoto M, Yahara S, Okuno S, Ishiguro K, Yamakawa O. 2002. Identification and characterization of foliar polyphenolic composition in Sweetpotato (*Ipomoea batatas* L.) Genotypes [J]. J Agric Food Chem. 50:3718-3722.
- Jiang X, Shi S, Zhang Y, Chen X. 2010. Excellent combination of HPLC-RSD-DAD-ESI/MS and HSCCC experiments to screen and identify radical scavengers from *Neo-Taraxacum siphonanthun*. J Braz Chem Soc. 21:1524-1529.
- Krenn L, Miron A, Pemp E, Petr U, Kopp B. 2003. Flavonoids from *Achillea nobilis* L., Z. Naturforsch. 58:11-16.
- Kozawa M, Matsuyama Y, Fukumoto M, Baba K. 1983. Chemical studies of *Coelopleurum gmelinii* (DC) LEDEB. I. constituents of the root. Chem Pharma Bull. 31:64-69.
- Laggoune S, Boutaghane N, Kabouche A, Kabouche Z, Ait-Kaki Z, Ait-Kaki B. 2008. Components and antimicrobial activity of *Lamium amplexicaules* from Algeria. Chem Nat Compds. 44:363-364.
- Liao CR, Kuo YH, Ho YL, Wang CY, Yang CS, Lin CW, Chang YS. 2014. Studies on cytotoxic constituents from the leaves of *Elaeagnus oldhamii* Maxim. in non-small cell lung cancer A549 cells. Molecules. 19:9515-9534.
- Lin, YL, Wang, WY. 2002. Chemica Constituents of *Vernonia patula*. Chin J Pharm. 54:, 187-192.
- Liu H, Zhang X, Wu C, Wu H, Guo P, Xu X. 2013. Anti-hyperlipidemic caffeoylquinic acids from the fruits of *Pandanus tectorius* Soland. J Appl Pharma Sci. 3:16-19.
- Lu, Y, Foo, L. Y. 1999. The polyphenol constituents of grape pomace. Food Chem. 65:1-8.
- Nacer A, Bernard A, Boustie J, Touzani R, Kabouche Z. 2006. Aglycone flavonoids of *Centaurea tougourensis* from Algeria. Chem Nat Compds. 42:230-231.
- Oberprieler C. 2002. A phylogenetic analysis of *Chamaemelum* Mill. (Compositae: anthemideae) and related genera based upon nrDNA ITS and cpDNA *trnL/trnF* IGS sequence variation. Bot J Linnean Soc. 138:255-273.

- Ode O, Asuzu I. 2014. Luteolin isolate from the methanol extract identified as the single-carbon compound responsible for broad antiulcer activities of *Cassia singueana* leaves. IOSR J Pharm. 4:17-23.
- Sakushima A, Ohno K, Coskun M, Seki KI, Ohkura K. 2002. Separation and identification of taxifolin 3-*O*-glucoside isomers from *Chamaecyparis obtusa* (Cupressaceae). Nat Prod Lett. 16:383-387.
- Satrani B, Ghanmi M, Farah A, Aafi A, Fougrach H, Bourkhis B, Bousta D, Talbi M. 2007. Composition chimique et activité antimicrobienne de l'huile essentielle de *Cladanthus mixtus*. Bull Soc Pharm Bordeaux. 146:85-96
- Shahzadi I, Shah MM. 2015. Acylated flavonol glycosides from *Tagetes minuta* with antibacterial activity. Front Pharmacol. 6:195.
- Sugiyama, M., Kikuchi, M. 1992. Studies on the constituents of *Osmanthus* species. X. Structures of phenolic glucosides from the leaves of *Osmanthus asiaticus* Nakai, Chem. Pharm. Bull. 40: 325-326.
- Valant-Vetschera KM, Wollenwebera E, Faure R, Gaydou E. 2003. New exudate flavonoids of species from the *Chrysanthemum* complex (Asteraceae-Anthemideae). Biochem Syst & Ecol. 31:545-548.
- Touafek O, Kabouche Z, Brouard I, Barrera-Bermejo J. 2011. Flavonoids of *Campanula alata* and their antioxidant activity. Chem Nat Compds. 46:968-970.
- Zaiter L, Bouheroum M, Benayache S, Benayache F, Leon F, Brouard I, Quintana J, Estévez F, Bermejo J. 2007. Sesquiterpene lactones and other constituents from *Matricaria chamomilla* L. Biochem Syst & Ecol. 35:533-538.
- Zrira S, Menut C, Bessiere J, Benjilalii B. 2007. Chemical composition of the essential oils of Moroccan *Ormenis mixta* (L.) Dumort. ssp. *multicaulis*. J Essent Oil Bear Plants. 10:378-385.

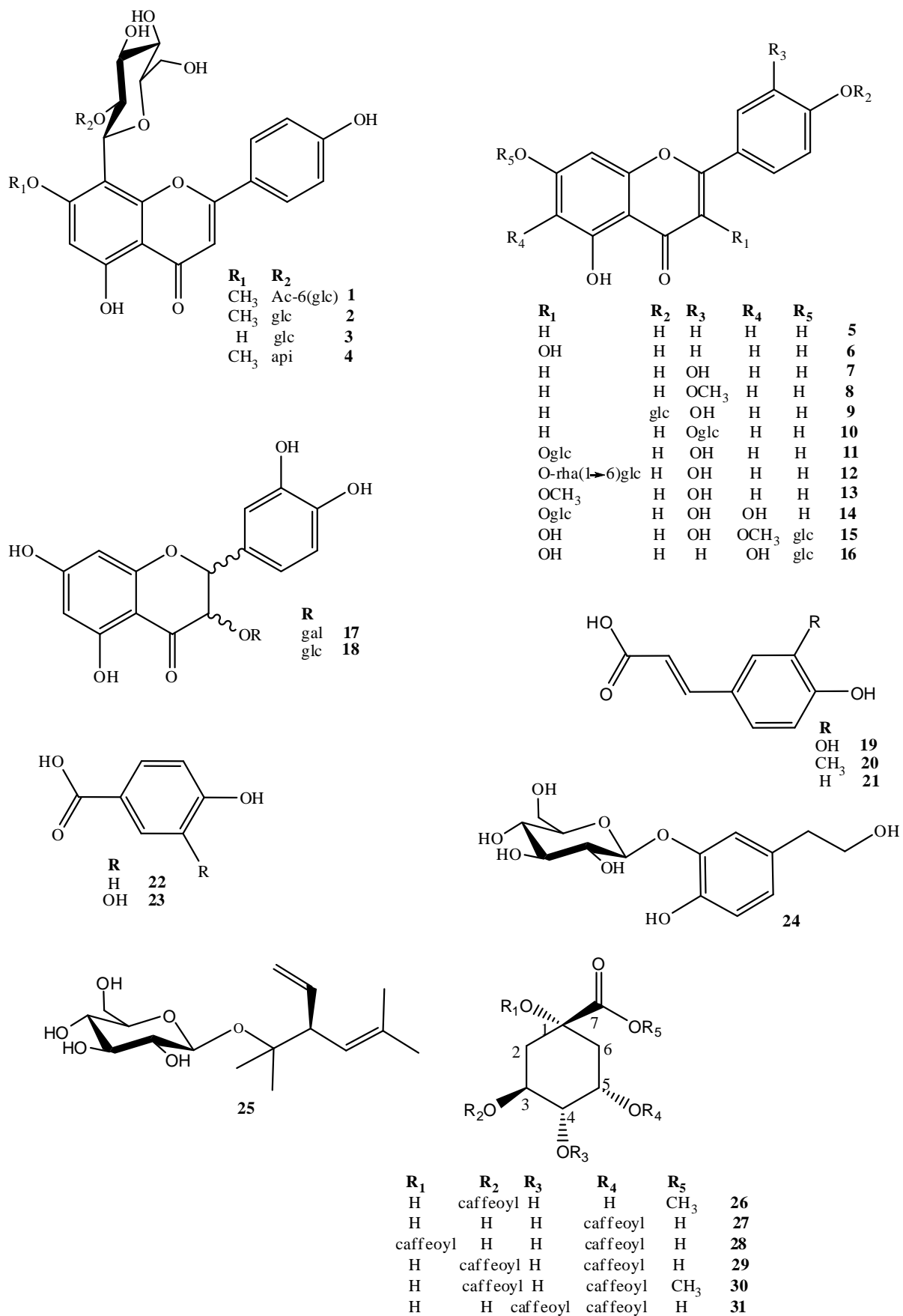


Fig. 1. Chemical structures of compounds 1-31 isolated from *Cladanthus mixtus*