Semipapposides A-M, triterpenoid bidesmosides saponins from the roots of Scabiosa semipapposa

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Abstract

Phytochemical investigations of the roots of *Scabiosa semipapposa* Salzem ex D.C. have led to the isolation of isolation of twelve undescribed triterpenoid saponins named semipapposides A-L, one undescribed saponin semipapposide M obtained as an inseparable mixture, together with three known oleanolic acid glycosides. Their structures were elucidated by analysis of 1D and 2D-NMR (^{1}H-^{1}H COSY, TOCSY, HSQC-TOCSY, ROESY, HSQC, and HMBC) spectroscopic data and mass spectrometry (HR-ESI-MS), and by comparison with those of related metabolites. These results represent a contribution to the chemotaxonomy of the genus *Scabiosa*, highlighting a partial sequence rhamnopyranosyl-(1→2)-xylopyranosyl or rhamnopyranosyl-(1→2)-arabinopyranosyl- at C-3 of the aglycon and a gentiobiose unit at C-28 as chemotaxonomic markers of this genus.

Keywords:

Scabiosa semipapposa Salzem ex D.C., Caprifoliaceae, Triterpenoid saponins, Semipapposides A-M

1. Introduction

The genus *Scabiosa*, belonging to the Caprifoliaceae family, is formed by around 100 species (Carlson et al., 2012) which grow as annual or perennial herbs, distributed in Europe and the Mediterranean Basin, southern Africa and eastern Asia (Reveal and Chase, 2011). It is well represented in Algeria by twelve species (Quezel and Santa, 1963).

Several *Scabiosa* species have been used in traditional medicine to treat measles and furuncles (Bonet et al., 2007), diphtheria (Rigat et al., 2007), respiratory infections including bronchitis, bronchial pneumonia, influenza and asthma, as well as high blood pressure, uterine disorders (Rigat et al., 2007; Kose et al., 2015; Moteetee et al., 2016), heel cracks (Bammi and Douira , 2002), dermatoses (herpes ringworm, scabies), ulcers (Girre, 1980) and liver diseases (Zhang et al., 2015).

Many *Scabiosa* species are characterized by an extremely rich chemical diversity (Pinto et al., 2018), in particular, pentacyclic triterpene saponins (Alimbaeva et al., 1977; Baykal et al., 1998; Zheng et al., 2004; Lehbili et al., 2018a; Kılınç et al., 2020) in addition to the flavonoids, coumarins (Garaev et al., 2008; Al-Qudah et al., 2017; Lehbili et al., 2018b), iridoid glycosides (Papalexandrou et al., 2003; Polat et al., 2010; Lehbili et al., 2018b) and lignan glycosides (Pasi et al., 2002).

In a continuation of our phytochemical investigations of *Scabiosa* species growing in Algeria (Lehbili et al., 2018a, Lehbili et al., 2018b), we are interested to *Scabiosa semipapposa* Salzem ex D.C. (*Scabiosa semipapposa f. albiflora* Faure & Maire is the synonyme), an herbaceous, hairy annual plant, rather canescent, with a lilac flowers and branched stems. The lower leaves are oval coarsely toothed. The petiolate superior ones are pinnatifid with oblong linear sub dentate lobes (Don, 1834; Quezel and Santa, 1963). As far as we know, no previous research work has investigated the chemical profile of *S. semipapposa* roots. In this study, the phytochemical composition of *S. semipapposa* roots has been explored and resulted in the isolation of twelve undescribed triterpene saponins, namely, semipapposides A-L (**1–12**), one undescribed saponin semipapposide M (**13**) obtained as an inseparable mixture with compound **4** (Fig.1), together with three known triterpene saponins (**14–16**).

2. Results and discussion

The 80% methanol extract of the roots of *S. semipapposa* was fractionated by vacuum-liquid chromatography (VLC) on RP-C_{18} and purified by successive chromatographic techniques, including flash chromatography as well as semi-preparative and preparative high performance liquid chromatography (HPLC) yielding sixteen compounds including thirteen undescribed

compounds (**1–13**) (Fig.1) and three known ones. Their structures were mainly elucidated by the use of 1D and 2D NMR techniques (^{1}H, *J*-modulated ^{13}C, DEPT, ^{1}H-^{1}H-COSY, TOCSY, *J*-modulated HSQC, HSQC-TOCSY, HMBC, ROESY, and NOESY) in combination with HR-ESI-MS and by comparison of their physical and spectral data with literature values. The known compounds were identified by comparison of their spectral data with literature values as scabiosaponine E (**14**) previously isolated from *Scabiosa tschiliensis* (Zheng et al., 2004), scabiostellatoside B (**15**) and D (**16**) previously isolated from *Scabiosa stellata* (Lehbili et al., 2018a).

Compounds **1**–**16** were isolated as white amorphous powders. The monosaccharides obtained by acid hydrolysis of an aliquot of the saponin containing fraction were identified as L-arabinose (Ara), L-rhamnose (Rha), D-glucose (Glc), D-xylose (Xyl), and D-galactose (Gal) by comparison on TLC with authentic samples followed by measurement of the optical rotations values of each purified sugar (see Experimental Section).

Compound **1** exhibited, in the positive ion-mode HR-ESI-MS, a quasi-molecular ion peak at *m/z* 1645.7848 $[M+H]^{+}$ (calcd for 1645.7849) compatible with the molecular formula $C_{76}H_{124}O_{38}$.

The ^{1}H and ^{13}C NMR spectra displayed resonances due to the triterpene part characteristic of oleanolic acid aglycone (Boutaghane et al., 2013, Mahato and Kundu 1994) (Table 1) with seven angular methyl groups at δ_H 1.07, 0.88, 0.97, 0.82, 1.17, 0.93 and 0.96 (s, each) showing correlations in the HSQC spectrum with their corresponding carbon at δ_C 27.1 (C-23), 15.9 (C-24), 14.8 (C-25), 16.4 (C-26), 24.9 (C-27), 32.1 (C-29) and 22.7 (C-30). Furthermore, other characteristic signals were observed such as one olefinic proton at δ_H 5.27 (1H, t, *J* = 3.7 Hz, H-12) and one oxygen-bearing methine protons at δ_H 3.14 (dd, *J* = 11.8, 4.3 Hz, H-3) showing HSQC correlation with δ_C 122.4 (C-12) and 88.8 (C-3), respectively. The ROESY correlations between H-3/H-5 (δ_H 0.80, d, *J* = 12.3 Hz) and H-5/H-9 (δ_H 1.59, m) indicated their α-axial orientation and thus the β-orientation of the oxygen at C-3. Extensive 2D NMR analysis confirmed the structure of the aglycone to be oleanolic acid. The chemical shift values at δ_C 88.8 (C-3) and 176.7 (C-28), suggested that the saponin was a bisdesmosidic glycoside with saccharide units attached to these positions.

The presence of eight sugar moieties in **1** was evidenced by the ^{1}H NMR spectrum which displayed eight anomeric protons at δ_H 5.38 (d, *J* = 1.5 Hz), 5.37 (d, *J* = 8.3 Hz), 5.21 (d, *J* = 1.1 Hz), 5.16 (d, *J* = 1.7 Hz), 4.62 (d, *J* = 7.9 Hz), 4.47 (d, *J* = 7.7 Hz), 4.39 (d, *J* = 7.2 Hz), and 4.36 (d, *J* = 7.6 Hz) (Table 2) giving correlations with eight anomeric carbons at δ_C 99.9, 94.3, 100.7, 101.3, 104.1, 105.0, 105.0, and 103.2, respectively in the HSQC spectrum.

Complete assignments of each sugar were achieved by extensive ^{1}D- and ^{2}D-NMR analyses (^{1}H-^{1}H-COSY, TOCSY, HSQC, HSQC-TOCSY, and ROESY) and by optical rotation (see experimental), allowing the characterization of two β-D-xylopyranosyl (Xyl I, δ_{H-1} 4.39 and Xyl II, δ_{H-1} 4.47), three α-L-rhamnopyranosyl (Rha I δ_{H-1} 5.38; Rha II, δ_{H-1} 5.21; and Rha III δ_{H-1} 5.16), and three β-D-glucopyranosyl (Glc I, δ_{H-1} 5.37; Glc II, δ_{H-1} 4.62; and Glc III δ_{H-1} 4.36) (Table 2). The large coupling constant (> 7 Hz) for xylose and glucose coupled with the ROE effect between H-1/H-3/H-5 axial, indicated their β-configuration. The small *J*-value of the anomeric proton of rhamnose ant the chemical shift of Rha-C-5 at δ_C 67-68 indicated their α-configuration (Kasai et al, 1979). The deshielding signals of Xyl I-C-2 (δ_C 76.7), Rha I-C-3 (δ_C 80.7), Xyl I-C-3 (δ_C 81.7), Rha II-C-4 (δ_C 82.3), Glc I-C-3 (δ_C 83.3), and Glc II-C-6 (δ_C 68.0), suggested that Xyl I, Xyl II, Rha I, Rha II, Gc I and Glc II were monosubstituted and that the two sugars chains were linear. The linkages between these sugars were established using mainly HMBC and ROESY spectra. The HMBC correlation between Xyl I-H-1 and C-3 of the aglycone and the ROESY correlation between Xyl I-H-1 and H-3 of the aglycone indicated the glycosidic linkage of the Xyl I at the C-3 position of the aglycone (Fig. 2). Moreover, the HMBC spectrum of compound **1** displayed long-range correlations between Rha I-H-1 /Xyl I-C-2, Xyl II-H-1/Rha I-C-3, Rha II-H-1/Xyl I-C-3, Glc I-H-1/ Rha II-C-4, and Rha III-H-1/ Glc I-C-3 indicating that the saccharidic chain α-L-rhamnopyranosyl-(1→3)-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-β-D-xylopyranoside- was linked to C-3 of the aglycone. In a similar fashion the cross peak observed in the HMBC spectrum between Glc III-H-1/Glc II-C-6 and Glc II-H-1/aglycone-C-28 established that the disaccharide gentiobiose [Glc III-(1→6)-Glc II] was linked to the C-28 of the aglycone. The linkages were ensured by the ROESY cross-peaks at: Rha III H-1/Glc I-H-3, Glc I-H-1/Rha II-H-4, Rha II-H-1/Xyl II-H-3, Xyl II-H-1/Rha I-H-3, Rha I-H-1/Xyl I-H-2, Xyl I-H-1/aglycone-H-3 and Glc III-H-1/Glc II-H_2-6. Compared to scabiostellatoside D (**16**), the sugar part at C-3 in **1** is composed of a supplementary rhamnopyranosyl unit (Lehbili et al., 2018a), and the signals due to gentiobiosyl at C-28 were superimposable.

The partial sequence –Rha-(1→2)-Xyl- at the C-3 position of the aglycone was often encountered in the *Scabiosa* genus, (Lehbili et al., 2018a; Zheng et al., 2004) and was characterized for all saponins **1**, **3**, **4**, **6**, **11**-**16**.

According to the above-described results, the structure of compound **1** was elucidated as 3-*O*-[α-L-rhamnopyranosyl-(1→3)-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-

xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-β-D-xylopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-oleanolic acid (Fig. 1), named semipapposide A.

For semipapposide B (**2**), the same molecular formula as **1** ($C_{76}H_{124}O_{38}$) was obtained according to its HR-ESI-MS (positive-ion mode) spectrum which displayed a pseudo-molecular ion peak at *m/z* 1645.7859 $[M+H]^+$, suggesting that compounds **1** and **2** were isomeric. All NMR signals observed for **2** were comparable to those of **1** (Tables 1 and 2), excepted for the sugar linked at the C-3 position of the aglycone. Its ring protons were assigned starting from anomeric protons resonance at δ_H 4.50 (d, J = 5.9 Hz) by means of the ^1H–^1H COSY, TOCSY, HSQC, and HMBC experiments and it was thus identified as an α-L-arabinopyranosyl moiety, characterized by its axial hydroxyl in C-4 position ($J_{H\text{-}3\text{-}H\text{-}4}$ = 3.7 Hz) (Lehbili et al., 2018a). Its attachment to the aglycone was confirmed by the HMBC correlation between δ_H 4.50 (Ara-H-1) and δ_C 89.2 (aglycone-C-3), and by the ROESY correlation between Ara-H-1 and aglycone H-3 (3.14, dd, *J*=11.8, 4.5 Hz, H-3). Hence, the structure of semipapposide B (**2**) was established as 3-*O*-[α-L-rhamnopyranosyl-(1→3)-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-oleanolic acid (Fig. 1).

Semipapposide C (**3**) exhibited in the HR-ESI-MS spectrum a quasi-molecular ion peak at *m/z* 1675.7977 $[M+H]^+$ (calcd for 1675.7954), compatible with the molecular formula $C_{77}H_{126}O_{39}$. Extensive 2D NMR analysis (Tables 1 and 2) showed that compounds **1** and **3** differed only by the nature of the sugar linked at the C-3 of the Rha I. The analysis of mass spectroscopy and 1D- and 2D-NMR spectra allowed the identification of one β-D-glucopyranosyl in **3** instead of a β-D-xylopyranosyl in **1**. The HMBC correlations between Glc I-H-1 (δ_H 4.50, d, *J*=7.9 Hz) and Rha I-C-3 (δ_C 81.5) suggested that Glc I was linked to the Rha I-C-3, which was confirmed by the ROESY cross-peak between Glc I-H-1 and Rha I-H-3 (δ_H 3.92, dd, *J*=9.6, 3.1 Hz). These evidences led to the elucidation of semipapposide C (**3**) as 3-*O*-[α-L-rhamnopyranosyl-(1→3)-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-β-D-xylopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-oleanolic acid.

The molecular formula of Semipapposide D (**4**) was determined as $C_{75}H_{122}O_{38}$ by the HR-ESI-MS showing a molecular ion peak at *m/z* 1631.7701 $[M+H]^+$ (calcd for 1631.7692). As in compounds **1–3**, the aglycone of **4** was identified by 2D-NMR analysis as oleanolic acid. Extensive 1D- and 2D-NMR analysis (Tables 1 and 2) showed that compounds **4** and **1** differed only by the sugar part. Eight sugar units were identified in **4** as three β-D-xylopyranose (Xyl I, Xyl II, and Xyl III), two α-L-rhamnopyranose (Rha I and Rha II) and three β-D-glucopyranose

(Glc I, Glc II, and Glc III) (Table 2). All signals corresponding to the partial sequence Xyl II-(1→3)-Rha I-(1→2)-Xyl I-(1→3)-oleanolic acid were almost superimposable to those of compound **1**. The sequence of the three remaining sugar units was suggested to be Rha II-(1→3)-Xyl III-(1→4)-Glc I-(1→4)- linked to C-4 of Xyl II based on the HMBC correlations between Rha II-H-1 (δ_H 5.18) / Xyl III-C-3 (δ_C 82.1), Xyl III-H-1 (δ_H 4.35) / Glc I-C-4 (δ_C 79.0), and Glc I-H-1 (δ_H 4.41)/Xyl II-C-4 (δ_C 76.9). The linkage was confirmed by the rOe effects observed between Rha III-H-1/Xyl III-H-3, Xyl III-H-1/Glc I-H-4, and Glc I-H-1/Xyl II-H-4. All these data were consistent with the structure of semipapposide D (**4**) as 3-*O*-[α-L-rhamnopyranosyl-(1→3)-β-D-xylopyranosyl-(1→4)-β-D-glucopyranosyl-(1→4)-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-β-D-xylopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-oleanolic acid (Fig. 1).

Semipapposide E (**5**) has the same molecular formula $C_{75}H_{122}O_{38}$ as compound **4**, as suggested by the HR-ESI-MS spectrum (*m/z* 1631.7699 $[M + H]^+$). The ^1H and ^{13}C NMR spectra (Tables 1 and 2) indicated that **5** was an oleanolic acid bidesmoside but differed from **4** in the nature of the sugar part. The HSQC spectrum of the sugar part showed eight cross-peaks at δ_H/δ_C 4.50 (d, *J*=4.5 Hz)/103.9, 4.51 (d, *J*=7.3 Hz)/104.9, 4.35 (d, *J*=7.7 Hz)/103.9, 5.21 (br s)/100.0, 5.18 (d, *J*=1.2 Hz)/101.2, 4.41 (d, *J*=7.8 Hz)/101.7, 5.37 (d, *J*=8.2 Hz)/94.3 and 4.36 (d, *J*=7.8 Hz)/103.2, indicating the presence of eight sugar units: Ara, Xyl I, Xyl II, Rha I, Rha II, Glc I, Glc II, and Glc III, respectively (Table 2). All the NMR signals observed for **5** were similar to those of **4**, except for the pentosyl moiety linked to the C-3 of the aglycone which was Ara in **5** instead of Xyl I in **4**. This was confirmed by the HMBC correlation at δ_H/δ_C 4.50 (Ara-H-1)/89.2 (C-3), and by the ROESY correlation at δ_H/δ_H 4.50 (Ara-H-1)/3.13 (dd, *J*=11.7, 4.3 Hz, aglycone-H-3). Hence, the structure of semipapposide E (**5**) was established as 3-*O*-[α-L-rhamnopyranosyl-(1→3)-β-D-xylopyranosyl-(1→4)-β-D-glucopyranosyl-(1→4)-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-oleanolic acid (Fig. 1).

For semipapposide F (**6**), the molecular formula $C_{82}H_{134}O_{43}$, obtained according to its $[M+H]^+$ ion at *m/z* 1807.8400 (in HR-ESI-MS positive-ion mode), differs from **1** only by 162 amu, corresponding to a supplementary hexosyl group. The findings from the HR-ESI-MS analysis were confirmed by the 2D-NMR data (Tables 3 and 4), which showed that compounds **6** and **1** differed only by the presence of one additional hexose unit identified as β-D-galactopyranosyl (Gal) (Lehbili et al., 2017). The observation of a HMBC correlation at δ_H/δ_C 4.46/72.9 (Gal-H-1/Glc I-C-4) and a ROESY cross-peak at δ_H/δ_H 4.46 (Gal-H-1)/3.83 (Glc I-H-4) proved the location of this additional Gal at Glc I-C-4. In addition, the HMBC correlation at δ_H/δ_C 5.39

(Rha III-H-1)/ 76.1 (Glc I-C-3) confirmed the linkage of Rha III to Glc C-3 position. Thus, the structure of semipapposide F (**6**) was elucidated as 3-*O*-[β-D-galactopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-β-D-xylopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-oleanolic acid (Fig. 1).

Semipapposide G (**7**) exhibited in the HR-ESI-MS experiment a quasi-molecular ion peak at *m/z* 1807.8390 $[M+H]^+$ ($C_{82}H_{135}O_{43}$, calcd 1807.8377). Extensive 2D-NMR analysis (Tables 1-4) showed that compounds **7** and **2** differed by the presence of one additional sugar unit which was identified as β-D-galactopyranosyl (Gal) (Table 4) ($\delta_{H\text{-}1}$ 4.46, d, *J*= 7.7 Hz; $\delta_{C\text{-}1}$ 102.9). The HMBC correlations at δ_H/δ_C 5.39 (Rha III-H-1)/76.1 (Glc I-C-3) and δ_H/δ_C 4.46 (Gal-H-1)/72.9 (Glc I-C-4) suggested the linkage of Rha III and Gal at the Glc I-C-3 and Glc I-C-4, respectively. The location of the supplementary Gal unit was confirmed by the ROESY cross-peak at δ_H/δ_H 4.46 (Gal-H-1)/3.83 (Glc I-H-4). Accordingly, the structure of semipapposide G (**7**) was elucidated as 3-*O*-[β-D-galactopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-oleanolic acid (Fig. 1).

The HR-ESI-MS spectrum of semipapposide H (**8**) showed a pseudo-molecular ion peak $[M+H]^+$ at *m/z* 1837.8517, indicating a molecular formula of $C_{83}H_{136}O_{44}$. It is higher than that of compound **7** by 30 amu, suggesting the presence of a hexose unit instead of a pentose unit. Extensive 2D-NMR analysis (Tables 3 and 4) showed that compounds **7** and **8** differed only by the nature of the sugar linked at Rha I-C-3. Analysis of 1D- and 2D-NMR spectra allowed the identification of one β-D-glucopyranose moiety in **8** instead of the β-D-xylopyranose unit in **7**. The HMBC correlations observed between Glc I-H-1/Rha I-C-3 indicated the linkage of Glc I at Rha I-C-3, and this was confirmed by the ROESY cross-peak between Glc I-H-1 and Rha I-H-3. Thus the structure of semipapposide H (**8**) was elucidated as 3-*O*-[β-D-galactopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-oleanolic acid (Fig. 1).

The molecular formula of semipapposide I (**9**) was determined as $C_{83}H_{136}O_{45}$ by the HR-ESI-MS showing a protonated molecular ion peak at *m/z* 1853.8469 $[M+H]^+$. According to the ^1H and ^{13}C NMR data (Tables 3-6), the compounds **9** and **8** showed the same sugar constituents and sequence of sugar chains. The difference was in the aglycone part, the methyl group at C-23 of aglycone in **8** was replaced by a hydroxymethyl group. The NMR spectra of **9** showed

two proton signals at δ_H 3.35 (H-23a) and 3.60 (H-23b) with carbon signal at δ_C 63.1 (C-23) instead of the methyl signal in **8**. In addition, the HMBC correlations of H-23 with C-3 (δ_C 81.0), C-5 (δ_C 46.7), and C-24 (δ_C 12.4) also confirmed the presence of the hydroxymethyl group at C-23. Thus, the aglycone of **9** was identified as hederagenin (Alabdul Magid et al., 2006; Mahato and Kundu 1994). Finally, the structure of **9** was assigned as 3-*O*-[β-D-galactopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-hederagenin (Fig. 1).

Semipapposide J (**10**) had the molecular formula $C_{82}H_{134}O_{44}$ [HR-ESI-MS (positive ion mode: *m/z* 1823.8344 $[M+H]^+$, (calcd for 1823.8326)], suggesting that this compound possessed a supplementary hydroxyl in comparison with the previous described saponins **7**. Comparative analysis of ^1H and ^{13}C NMR signals of compounds **10** and **7** indicated that **10** possessed the same glycosidic chains (Tables 5 and 6). The NMR spectra showed signals for only six methyl groups and one hydroxylated methylene (δ_C 63.1; δ_H 3.35 and 3.56) characterizing hederagenin as the aglycone of saponin **10** as in **9**. An extensive analysis of 1D- and 2D-NMR spectra of **10** allowed to conclude that semipapposide J (**10**) was 3-*O*-[β-D-galactopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-hederagenin (Fig. 1).

Semipapposide K (**11**) has the same molecular formula $C_{82}H_{134}O_{44}$ as compound **10**, as suggested by the HR-ESI-MS spectrum (*m/z* 1823.8350 $[M+H]^+$). As for compounds **9** and **10**, the aglycone of semipapposide K (**11**) displayed classical signals in the ^1H and ^{13}C NMR spectra of hederagenin. All the NMR signals observed for **11** were similar to those of **10** (Tables 5 and 6), except for the pentose moiety linked to the C-3 of the aglycone which was Xyl I in **11** instead of Ara in **10**. The linkage was confirmed by the HMBC correlation at δ_H/δ_C 4.48 (Xyl I-H-1)/80.8 (aglycone-C-3), and by the ROESY correlation at δ_H/δ_H 4.48 (Xyl I-H-1)/3.62 (aglycone-H-3). Therefore, semipapposide K (**11**) was elucidated as 3-*O*-[β-D-galactopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-β-D-xylopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-hederagenin (Fig. 1).

Semipapposide L (**12**) gave the same molecular formula $C_{83}H_{136}O_{45}$ as **9** deduced from the HR-ESI-MS spectrum (*m/z* 1853.8445 $[M+H]^+$, calcd for 1853.8432). As for the pair of saponins **10** and **11**, the isomeric saponins **12** and **9** differed by the nature of pentose unit linked to the

aglycone (Tables 5 and 6). Consequently, semipapposide L (**12**) was deduced to be 3-*O*-[β-D-galactopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-β-D-xylopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-hederagenin (Fig. 1).

Semipapposide M (**13**) and semipapposide D (**4**) were obtained as an inseparable mixture with the ratio 4:6, based on the integral intensity of the corresponding signals in their ^{1}H NMR spectra and HR-ESI-MS. Exhaustive efforts to separate this mixture employing normal and reversed-phase flash chromatography and HPLC using various stationary and mobile phases were failed. Therefore, the structures of these two compounds were elucidated as a mixture. According to the HR-ESI-MS [$[M+H]^+$ at *m/z* 1631.7709 and NMR analysis, semipapposide D (**4**) was identified as the major compound of this mixture. The second protonated molecular ion $[M+H]^+$ at *m/z* 1837.8508 correspond to semipapposide M (**13**) that allowed establishment of the molecular formula $C_{83}H_{136}O_{44}$. Comparing the ^{1}H and ^{13}C NMR data (Tables 3-6) of **13** with those of **12** showed the same sugar components and sequence of sugar chains. The difference was in the aglycone part; the hydroxymethyl group at C-23 of the aglycone in **12** was replaced by a methyl group, suggesting oleanolic acid as aglycone. Thus the structure of semipapposide M (**13**) was elucidated as 3-*O*-[β-D-galactopyranosyl-(1→4)-[α-L-rhamnopyranosyl-(1→3)]-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-β-D-xylopyranosyl]-28-*O*-[β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl]-oleanolic acid (Fig. 1).

3. Conclusion

The present phytochemical study carried out for the first time on the Algerian *S. semipapposa* species, allowed the isolation and elucidation of twelve undescribed oleanolic acid (**1-8**) and hederagenin (**9-12**) bidesmosidic saponins, with one undescribed oleanolic acid bidesmosidic saponin (**13**) obtained as an inseparable mixture with compound **4**, together with three known oleanolic acid glycosides (**14-16**). All triterpene saponins (**1-16**) were characterized with a gentiobiose unit at C-28 and a common partial sequence as (-Rha-(1→2)-Xyl-) or (-Rha-(1→2)-Ara-) at the C-3 position of the aglycone with additional glycosylation at the C-3 of the Rha, frequently encountered in several *Scabiosa* species (Baykal et al., 1998; Zheng et al., 2004; Lehbili et al., 2018a) and in some plants of the Caprifoliaceae family such as *Cephalaria* (Sarikahya et al., 2014), and seems to represent a chemotaxonomic marker for this genus. Our investigation contributes to phytochemical database of *S. semipapposa* associated with

chemotaxonomic meaning of *Scabiosa* spp. Therefore, our results are in accordance with these findings and bring an additional contribution to the knowledge of the saponins of *Scabiosa* species. They confirmed the richness of pentacyclic triterpenoids in the *Scabiosa* species observed by Pinto et al. (2018) with the presence of oleanolic acid as main aglycone, and glucose, xylose, rhamnose and arabinose as sugars. It should be interesting to evaluate the biological activities of these saponins.

4. Experimental

4.1. General experimental procedures

Optical rotations values were recorded on a PerkinElmer 341 Polarimeter. The 1D and 2D NMR spectra (^{1}H and ^{13}C NMR, ^{1}H–^{1}H COSY, TOCSY, ROESY, *J*-modulated HSQC, HSQC-TOCSY and HMBC) were performed using a Bruker Avance III 600 spectrometer (^{1}H at 600 MHz and ^{13}C at 150 MHz) equipped with a 5 mm TCI cryoprobe. 2D-NMR experiments were performed using standard Bruker microprograms (TopSpin 3.5 software). HR-ESI-MS experiments were performed using a Micromass Q-TOF instrument. Flash chromatography was carried out on a Grace Reveleris system equipped with dual UV and ELSD detection using Grace® cartridges (Silica gel or RP-C_{18}). Preparative HPLC was performed on Armen Instrument apparatus equipped with an AP 250 pump and a Knauer (Merck) detector UV K-2501. A manually packed C_{18} column (LiChrospher, 20 x 5 cm, 12 µ) was used for preparative HPLC. The mobile phase consisted of H_2O with TFA (0.0025%) and CH_3CN with a flow rate of 75 mL/min and the chromatograms were monitored at 205, 254, 300 and 360 nm. Semi-preparative HPLC was realized on a Dionex apparatus equipped with an ASI-100 automated sample injector, a STH 585 column oven, a P580 pump, a diode array detector UVD 340S and the Chromeleon® software version 6.8. A prepacked RP-C_{18} column (Phenomenex 250 x 10 mm, Luna 5 µ) was used for semi-preparative HPLC. The eluting mobile phase consisted of H_2O with TFA (0.0025%) and CH_3CN with a flow rate of 5 mL/min and the chromatogram was monitored at 205 and 215 nm. Analytical HPLC experiments were performed using a Thermofisher Ultimate 3000 (Thermo Fischer Scientific, Villebon sur Yvette, France), equipped with a 4 ways pump LPG 3400 SD, an automatic injector WPS 3000 SL, a UV/visible diode array detector 3000 and the Chromeleon® software version 6.8. A prepacked C_{18} column Uptisphere Strategy C_{18} (Interchim, 4.6 × 250 mm, 5 µ) was used for analytical HPLC and the mobile phase consisted of H_2O with TFA (0.0025% v/v) and CH_3CN with a flow rate of 1 mL/min and the chromatograms were monitored at 205, 215, 254, and 360 nm. TLC was

performed on pre-coated silica gel 60 F_{254} Merck and compounds were visualized by spraying the dried plates with 50% H_2SO_4, followed by heating.

4.2. Plant material

The plant material (roots) of *Scabiosa semipapposa* Salzem ex D.C. was collected from Alguemas, in the region of Constantine North-Eastern Algeria (latitude 36.3479 and longitude 6.650773) in May 2017. The plant material was authenticated by Mr. Kamel Kabouche. A voucher specimen (LOST Ss.05/17) has been deposited at the herbarium of LOST Laboratory, University Frères Mentouri-Constantine, Algeria.

4.3. Extraction and isolation

Dried powdered roots of S. *semipapposa* (1 kg) were extracted with 80% MeOH (3 × 10 L, 24 h) at room temperature. The resulting extracts were combined and concentrated under vacuum to give the crude extract (100 g). A part of this (55 g) was subjected to RP-C_{18} vacuum liquid chromatography (VLC) eluted successively with MeOH-H_2O (3:7, 4:6, 6:4, 8:2 and 10:0), to give 5 main fractions (VLC-A to VLC-E, respectively). The saponins enriched fraction E (4.6 g) was fractionated by flash chromatography over silica gel, eluted by a gradient system of $CHCl_3$-MeOH-H_2O (10:0:0 to 4:6:0.7), in 68 min to afford 12 sub-fractions (E1-E12). Sub-fraction E_{10} (482 mg) was purified by preparative HPLC using a gradient (30-35% CH_3CN, in 45 min) to give 56 sub-fractions. Sub-fractions E10-$_{27\text{-}28}$ contained the pure compound **5** (9.4 mg). The purification by semi-prep HPLC of sub-fractions E10-$_{35\text{-}44}$ (98.7 mg) led to compounds **4** (3.0 mg, t_R 11.5 min), **14** (2.2 mg, t_R 12.4 min), **3** (3.2 mg, t_R 13.1 min), **2** (7.28 mg, t_R 14.4 min), **16** (3.7 mg, t_R 15.4 min), **1** (10.4 mg, t_R 15.5 min) and **15** (2.5 mg, t_R 17.1 min) using a gradient 32-34% CH_3CN, in 18 min. Sub-fraction E11 (329 mg) was subjected to High Performance Flash Chromatography, over RP-C_{18}, using a binary gradient of MeOH–H_2O (20-80% MeOH in 34 min) as eluting solvent to obtain 39 sub-fractions. Sub-fraction E11-$_{26\text{-}27}$ was obtained as pure compound **7** (9.3 mg). Further separations of the remaining sub-fractions E11-$_{28\text{-}39}$ were performed by flash Chromatography on silica gel ($CHCl_3$–MeOH–H_2O 68/32/3) and then, purification by semi-prep HPLC eluted by gradient system 32-34% CH_3CN, in 18 min to give compounds **4** (2 mg, t_R 11.3 min), **7** (10.8 mg, t_R 12.4 min), **6** (21.4 mg, t_R 13.7 min) and **1** (2.7 mg, t_R 14.8 min).

Sub-fraction E12 (1.4 g) was subjected to preparative HPLC (30-35% CH_3CN, in 45 min) to afford 48 sub-fractions and the pure compounds **6** (70 mg) and **10** (7.9 mg) in the sub-fraction E12-$_{41}$ and E12-$_{14}$, respectively. The combined sub-fractions E12-$_{12\text{-}13}$ (16.3 mg) were purified

by semi-preparative HPLC (30-32% CH_3CN, in 15 min) to furnish compounds **9** (2.8 mg, t_R 6.9 min) and **10** (3.3 mg, t_R 7.7 min). Sub-fraction E12-$_{15}$ (7.9 mg) was purified by semi-prep. HPLC using a gradient of 30-32% CH_3CN, in 15 min as eluant to give compounds **12** (2.6 mg, t_R 8.2 min) and **11** (1.9 mg, t_R 8.9 min) whereas the sub-fractions E12-$_{25\text{-}27}$ (29.3 mg) and E12-$_{31\text{-}34}$ (77 mg) were purified by semi preparative HPLC using an elution program: 32-34% of CH_3CN, for 18 min to afford compounds **8** (17.5 mg, t_R 10.6 min), **7** (8.4 mg, t_R 11.1 min), mixture of **13** and **4** (15.3 mg, t_R 11.3 min) and **6** (5.5 mg, t_R 11.8 min). Finally compounds **6** (4.4 mg, t_R 11.0 min), **1** (9.7 mg, t_R 12.1 min) and **16** (1 mg, t_R 12.4 min) were obtained from E12-$_{46\text{-}48}$ (60 mg) by semi-prep HPLC using a gradient (33-35% CH_3CN, in 15 min).

*4.3.1. Semipapposide A (**1**)*

Amorphous white powder; $[\alpha]_D^{20}$- 40.7 (*c* 0.83, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 1 and 2. HR-ESI-MS *m/z*: 1645.7848 $[M+H]^+$ (calcd for $C_{76}H_{125}O_{38}$, 1645.7849).

*4.3.2. Semipapposide B (**2**)*

Amorphous white powder; $[\alpha]_D^{20}$-35.4 (*c* 0.65, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 1 and 2. HR-ESI-MS *m/z*: 1667.7676 $[M+Na]^+$, 1645.7859 $[M+H]^+$ (calcd for $C_{76}H_{125}O_{38}$, 1645.7884).

*4.3.3. Semipapposide C (**3**)*

Amorphous white powder; $[\alpha]_D^{20}$-24.8 (*c* 0.27, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 1 and 2. HR-ESI-MS *m/z*: 1697.7797 $[M+Na]^+$, 1675.7977 $[M+H]^+$ (calcd for $C_{77}H_{127}O_{39}$, 1675.7954).

*4.3.4. Semipapposide D (**4**)*

Amorphous white powder; $[\alpha]_D^{20}$-16.8 (*c* 0.25, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 1 and 2. HR-ESI-MS *m/z*: 1653.7532 $[M+Na]^+$, 1631.7701 $[M+H]^+$ (calcd for $C_{75}H_{123}O_{38}$, 1631.7692).

*4.3.5. Semipapposide E (**5**)*

Amorphous white powder; $[\alpha]_D^{20}$-33.3 (*c* 0.42, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 1 and 2. HR-ESI-MS *m/z*: 1631.7699 $[M+H]^+$ (calcd for $C_{75}H_{123}O_{38}$, 1631.7692).

*4.3.6. Semipapposide F (**6**)*

Amorphous white powder; $[\alpha]_D^{20}$-37.1 (*c* 0.83, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 3 and 4. HR-ESI-MS *m/z*: 1829.8225 $[M+Na]^+$, 1807.8400 $[M+H]^+$ (calcd for $C_{82}H_{135}O_{43}$, 1807.8377).

*4.3.7. Semipapposide G (**7**)*

Amorphous white powder; $[\alpha]_D^{20}$-19.7 (*c* 0.37, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 3 and 4. HR-ESI-MS *m/z*: 1807.8390 $[M+H]^+$ (calcd for $C_{82}H_{135}O_{43}$, 1807.8377).

*4.3.8. Semipapposide H (**8**)*

Amorphous white powder; $[\alpha]_D^{20}$-27.2 (*c* 1.14, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 3 and 4. HR-ESI-MS *m/z*: 1859.8307 $[M+Na]^+$, 1837.8517 $[M+H]^+$ (calcd for $C_{83}H_{137}O_{44}$, 1837.8483).

*4.3.9. Semipapposide I (**9**)*

Amorphous white powder; $[\alpha]_D^{20}$-17.8 (*c* 0.23, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 5 and 6. HR-ESI-MS *m/z*: 1875.8267 $[M+Na]^+$, 1853.8469 $[M+H]^+$ (calcd for $C_{83}H_{137}O_{45}$, 1853.8432).

*4.3.10. SemipapposideJ (**10**)*

Amorphous white powder; $[\alpha]_D^{20}$-28.2 (*c* 0.66, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 5 and 6. HR-ESI-MS *m/z*: 1845.8151 $[M+Na]^+$, 1823.8344 $[M+H]^+$ (calcd for $C_{82}H_{135}O_{44}$, 1823.8326).

*4.3.11. Semipapposide K (**11**)*

Amorphous white powder; $[\alpha]_D^{20}$-28.6 (*c* 0.79, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 5 and 6. HR-ESI-MS *m/z*: 1845.8151 $[M+Na]^+$, 1823.8350 $[M+H]^+$ (calcd for $C_{82}H_{135}O_{44}$, 1823.8326).

*4.3.12. Semipapposide L (**12**)*

Amorphous white powder; $[\alpha]_D^{20}$-22.8(*c* 0.21, MeOH); ^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables5 and 6. HR-ESI-MS *m/z*: 1875.8276 $[M+Na]^+$, 1853.8445 $[M+H]^+$ (calcd for $C_{83}H_{137}O_{45}$, 1853.8432).

*4.3.13. Semipapposide M (**13**)*

^{1}H (600 MHz, CD_3OD) and ^{13}C NMR (150 MHz, CD_3OD) data; see Tables 3 and 4. HR-ESI-MS *m/z*: 1837.8508 $[M+H]^+$ (calcd for $C_{83}H_{137}O_{44}$, 1837.8483).

4.4. Acid hydrolysis.

A part of the saponins rich fraction E (100 mg) was hydrolyzed with 15 mL of 2 N TFA (trifluoroacetic acid, aqueous solution) at 90 °C for 4 h. After extraction with CH_2Cl_2 (3×7 mL), the aqueous layer was evaporated to furnish the monosaccharide residue (56.7 mg). Five sugars were identified and compared with authentic samples by TLC using BuOH-iso-PrOH-Me_2CO-H_2O (20:10:7:6) as glucose, galactose, xylose, arabinose and rhamnose. After purification of these sugars by preparative TLC in the same solvent, the optical rotation of each purified sugar was measured and the obtained values were as follow: D-glucose [10.7 mg, R_f = 0.42, $[\alpha]^{20}_D$ +33 (*c* 0.89, H_2O)], D-galactose [2.7 mg, R_f = 0.45, $[\alpha]^{20}_D$ +25 (*c* 0.22, H_2O)], D-xylose [3.3 mg, R_f = 0.61, $[\alpha]^{20}_D$ +18.8 (*c* 0.27, H_2O)], L-arabinose [3.6 mg, R_f = 0.36, $[\alpha]^{20}_D$ +43 (*c* 0.30, H_2O)], and L-rhamnose [10.4 mg, R_f = 0.53, $[\alpha]^{20}_D$ +5.6 (*c* 0.86, H_2O)].

Declaration of competing interest

All authors state no conflict of interest

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Supplementary data

^1H, ^{13}C NMR, and HSQC spectra of new compounds **1-13** are available online.

References

Alabdul Magid, A., Voutquenne-Nazabadioko, L., Renimel, I., Harakat, D., Moretti, C., Lavaud, C., 2006. Triterpenoid saponins from the stem bark of *Caryocar villosum*. Phytochemistry 67, 2096-2102.

Alimbaeva, P.K., Akimaliev, A., Mukhamedziev, M.M., 1977. Triterpene glycosides of some representatives of the Dipsacaceae family. Khim. Prir. Soedin. 5, 708-709.

Al-Qudah, M.A., Otoom, N.K., Al-Jaber H.I., Saleh, A.M., Abu Zarga, M.H., Afifi, F.U., Abu Orabi, S.T., 2017. New flavonol glycoside from *Scabiosa prolifera* L. aerial parts with in vitro antioxidant and cytotoxic activities, Nat. Prod. Res. 31, 2865-2874.

Bammi, J., Douira, A., 2002. Les plantes médicinales dans la forêt de L'Achach (Plateau Central, Maroc). Acta. Bot. Malacit. 27, 131–145.

Baykal, T., Panayir, T., Tasdemir, D., Sticher, O., Çalis, I., 1998. Triterpene saponins from *Scabiosa rotata*. Phytochemistry 48, 867-873.

Bonet, M.À., Parada, M., Selga, A., Vallès, J., 1999. Studies on pharmaceutical ethnobotany in the regions of L'Alt Empordà and Les Guilleries (Catalonia, Iberian Peninsula). J. Ethnopharmacol 68, 145–168.

Boutaghane, N., Voutquenne-Nazabadioko, L., Harkat, D., Simon, A., Kabouche, Z., 2013. Triterpenoid saponins of *Genista ulicina* Spach. Phytochemistry 93, 176–181.

Carlson, S.E., Linder, H.P., Donoghue, M.J., 2012. The historical biogeography of *Scabiosa* (Dipsacaceae): implications for old world plant disjunctions. J. Biogeogr. 39, 1086-1100.

Don, G., 1834. A General History of Dichlamydeous Plants Comprising Complete Descriptions of Different Orders. Volume III. London J G and F Rivington and Others, pp 692.

Garaev, E.A., Movsumov, I.S., Isaev, M.I., 2008. Flavonoids and oleanolic acid from *Scabiosa caucasia*. Chem. Nat. Compd. 44, 520-521

Girre, L., 1980. Connaitre et Reconnaitre les Plantes Médicinales. Ouest, France, Rennes.

Papalexandrou, A., Magiatis, P., Perdetzoglou, D., Skaltsounis, A.L., Chinou, I.B.,

Harvala, C., 2003. Iridoids from *Scabiosa variifolia* (Dipsacaceae) growing in Greece. Biochem. Syst. Ecol. 31, 91-93.

Kasai, R.; Okihara, M.; Asakawa, J.; Mizutani, K.; Tanaka, O. 1979, ^{13}C NMR study of α- and β-anomeric pairs of D-mannopyranosides and L-rhamnopyranosides. Tetrahedron 35, 1427-1432.

Kılınç, H., Masullo, M., D'Urso, G., Karayildirim, T., Alankus, O., Piacente, S., 2020. Phytochemical investigation of *Scabiosa sicula* guided by a preliminary HPLC-ESIMS^n profiling. Phytochemistry 174, 112350.

Kose, L.S., Moteetee, A., Vuuren, S.V., 2015. Ethnobotanical survey of medicinal plants used in the Maseru district of Lesotho. J. Ethnopharmacol.170, 184–200.

Lehbili, M., Alabdul Magid, A., Kabouche, A., Voutquenne-Nazabadioko, L., Morjani, H., Harkat, D., Kabouche, Z., 2018a. Triterpenoid saponins from *Scabiosa stelatta* collected in the North- eastern Algeria. Phytochemistry 150, 40-49.

Lehbili, M., Alabdul Magid, A., Hubert, J., Kabouche, A., Voutquenne-Nazabadioko, L., Renault, J-H., Nuzillard, J-M., Morjani, H., Abedinib, A., Gangloff, S-C., Kabouche,A., 2018b. Two new bis-iridoids isolated from *Scabiosa stellata* and their antibacterial, antioxidant, anti-tyrosinase and cytotoxic activities. Fitoterapia 125, 41–48.

Lehbili, M., Alabdul Magid, A., Kabouche, A., Voutquenne-Nazabadioko, L., Abedini, A., Morjani, H., Sarazin, T.,Gangloff, S.C., Kabouche, Z., 2017. Oleanane-type triterpene saponins from *Calendula stellata*. Phytochemistry 144, 33-42.

Mahato, S.B., Kundu, A.P., 1994. ^{13}C NMR spectra of pentacyclic triterpenoids. A compilation and some salient features. Phytochemistry 37, 1517-1575.

Moteetee, A., Kose, L.S., 2016 .Medicinal plants used in Lesotho for treatment of reproductive and post reproductive problems. J. Ethnopharmacol. 194, 827–849.Pasi, S., Aligiannis, N., Skaltsounis, A.L., and Chinou, I.B., 2002. A New Lignan Glycoside and Other Constituents from *Cephalaria Ambrosioides*. Nat Prod Lett 16, 365-370.

Polat, E., Alankus-Caliskan,O., Karayildirim, T., Bedir, E., 2010. Iridoids from *Scabiosa atropurpurea L*. subsp. *maritima* Arc. (L.). Biochem. Syst. Ecol. 38, 253-255

Pinto, D. C. G. A., Rahmouni, N., Beghidja, N., Silva, A. M. S., 2018. *Scabiosa* Genus: A Rich Source of Bioactive Metabolites. Medicines 5, 110.

Quezel, P., Santa, S., 1963. New flora of Algeria and the desert regions MeridionalTome II. Paris. In: French National Center for Scientific Research, pp. 890-893.

Reveal, J. L., and Chase, M. W. 2011. APG III: Bibliographical information and synonymy of Magnoliidae. Phytotaxa 19: 71–134.

Rigat, M., Bonet, M.À.; Garcia, S.; Garnatje, T.; Vallès, J., 2007.Studies on pharmaceutical ethnobotany in the high river Ter valley (Pyrenees, Catalonia, Iberian Peninsula). J. Ethnopharmacol. 113, 267–277.

Sarikahya, N.B., 2014. Aristatosides A-C, hederagenin-type triterpene saponins from *Cephalaria aristata*. Phytochem Lett. 8, 149-155.

Zhang, B.B., He, B.Q., Sun, J.B., Zeng, B., Shi, X.J., Zhou, Y., Niu, Y., Nie, S.Q., Feng, F.,Liang, Y., Wu, F.H., 2015. Diterpenoids from *Saliva plebeia* R. Br. and their antioxidant and anti-inflammatory activities. Molecules 20, 14879-14888.

Zheng, Q., Koike, K., Han, L.K., Okuda, H., Nikaido, T., 2004. New biologically active triterpenoid saponins from *Scabiosa tschiliensis*. J. Nat. Prod. 67, 604-613.

Table 1 ^{13}C NMR and ^{1}H NMR spectroscopic data of the aglycone moieties of compounds **1**–**5** in

Position	**1**		**2**		**3**		**4**		**5**	
	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H
1	38.7	1.00, td (13.5, 3.1) 1.63, m	38.6	1.00, td (13.6, 3.3) 1.64, m	38.6	1.00, td (13.5, 4.4) 1.62, m	38.6	1.00, td (13.7, 3.9) 1.63, m	38.5	1.00, m 1.64, m
2	25.9	1.70, m 1.82, m	25.7	1.73, m 1.86, m	25.9	1.70, m 1.82, m	25.9	1.70, m 1.82, m	25.7	1.73, m 1.86, td (13.3, 3.4)
3	88.8	3.14, dd (11.8, 4.3)	89.2	3.14, dd (11.8, 4.5)	88.9	3.14, dd (11.5, 4.2)	88.9	3.14, dd (11.7, 4.3)	89.2	3.13, dd (11.7, 4.3)
4	38.9	-	38.9	-	38.9	-	38.9	-	38.9	-
5	55.9	0.80, d (12.3)	55.7	0.80, d (12.3)	55.8	0.80, d (11.9)	55.9	0.80, d (12.0)	55.7	0.80, m
6	18.0	1.42, m 1.55, m	18	1.43, m 1.56, m	18.0	1.42, td (13.2,3.2) 1.56, m	18.0	1.43, m 1.56, m	18.0	1.43, m 1.55, m
7	32.5	1.33, m 1.51, td (13.7, 4.3)	32.5	1.34, m 1.51, td (12.2, 3.3)	32.5	1.33, m 1.50, td (13.0, 3.5)	32.5	1.34, m 1.50, td (12.4, 3.5)	32.5	1.34, m 1.50, td (12.2, 3.5)
8	39.3	-	39.3	-	39.3	-	39.3	-	39.3	-
9	47.8	1.59, m	47.6	1.60, m	47.9	1.60, m	47.6	1.60, m	47.7	1.60, m
10	36.5	-	36.5	-	36.5	-	36.5	-	36.5	-
11	23.2	1.91, m	23.1	1.91, m	23.1	1.90, m	23.1	1.90, m	23.1	1.91, m
12	122.4	5.27, t (3.7)	122.4	5.27, t (3.7)	122.4	5.27, t (3.6)	122.4	5.27, t (3.6)	122.4	5.27, t (3.4)
13	143.5	-	143.5	-	143.5	-	143.5	-	143.5	-
14	41.5	-	41.5	-	41.5	-	41.5	-	41.5	-
15	27.5	1.10, dm (13.7) 1.80, m	27.5	1.10, dm (13.7) 1.80, td (13.7,3.0)	27.5	1.10, dm (13.9) 1.80, m	27.5	1.10, dm (13.6) 1.80, m	27.5	1.10, dm (13.9) 1.80, m
16	22.6	1.73, m 2.07, td (13.2, 3.6)	22.6	1.73, m 2.07, td (13.3, 3.7)	22.6	1.73, m 2.07, td (13.6, 3.6)	22.4	1.73, m 2.07, td (13.2, 3.4)	22.6	1.73, m 2.07, td (13.2, 3.3)
17	46.6	-	46.6	-	46.6	-	46.6	-	45.8	-
18	41.1	2.88, dd (13.7, 4.2)	41.1	2.88, dd (13.7, 4.0)	41.1	2.88, dd (13.6, 4.1)	41.1	2.88, dd (13.5, 4.0)	41.1	2.88, dd (13.4, 4.1)
19	45.8	1.17, m 1.73, t (13.7)	45.9	1.16, m 1.73, t (13.7)	45.3	1.16, m 1.73, t (13.6)	45.8	1.16, m 1.73, t (13.5)	46.6	1.16, m 1.73, t (13.4)
20	30.2	-	30.2	-	30.1	-	30.1	-	30.1	-
21	33.5	1.24, m 1.42, td (13.2, 2.8)	33.5	1.24, m 1.42, td (13.0, 2.6)	33.5	1.24, m 1.42, td (13.6, 4.1)	33.5	1.24, m 1.42, m	33.5	1.24, m 1.42, td (13.2, 3.5)
22	31.8	1.62, m 1.73, m	31.8	1.62, m 1.74, m	31.8	1.62, m 1.74, m	31.7	1.62, m 1.74, m	31.8	1.62, m 1.74, m
23	27.1	1.07, s	27.3	1.05, s	27.1	1.07, s	27.1	1.07, s	27.2	1.05, s
24	15.9	0.88, s	15.8	0.88, s	15.8	0.87, s	15.85	0.88, s	15.8	0.87, s
25	14.8	0.97, s	14.7	0.98, s	14.8	0.96, s	14.8	0.97, s	14.8	0.98, s
26	16.4	0.82, s	16.4	0.82, s	16.4	0.82, s	16.4	0.82, s	16.4	0.82, s
27	24.9	1.17, s	24.9	1.18, s	24.9	1.17, s	24.9	1.17, s	24.9	1.17, s
28	176.7	-	176.7	-	176.7	-	176.7	-	176.7	-
29	32.1	0.93, s	32.1	0.93, s	32.1	0.93, s	32.1	0.93, s	32.1	0.93, s
30	22.7	0.96, s	22.6	0.96, s	22.6	0.96, s	22.6	0.96, s	22.6	0.96, s

CD_3OD.[a]

[a] in ppm, *J* in parentheses in Hz.

Table 2 ^{13}C NMR and ^{1}H NMR spectroscopic data in CD_3OD of the sugar moieties of compounds **1-5**[a]

Position	1		2		3		4	
	δ_C	δ_C	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H
C-3	**Xyl I**		**Ara**		**Xyl I**		**Xyl I**	
1	105.0	4.39, d (7.2)	103.9	4.50, d (5.9)	105.2	4.38, d (7.3)	105.0	4.38, d (7.1)
2	**76.7**	3.41, dd (8.5, 7.2)	**74.9**	3.77, dd (8.6, 5.9)	**77.4**	3.33, m	**76.6**	3.41, dd (8.3, 7.1)
3	77.5	3.45, t (8.5)	72.4	3.72, dd (8.6, 3.7)	77.1	3.40, t (9.4,)	77.5	3.44, t (8.3)
4	70.1	3.47, m	67.8	3.78, d (3.7)	70.2	3.48, m	70.3	3.48, m
5	65.2	3.19, dd (11.6, 9.8)	63.4	3.52, m	65.2	3.18, t (10.6)	65.2	3.19, dd (12.5, 9.9)
		3.84, dd (11.6, 5.0)		3.86, dd (9.5, 2.2)		3.85, dd (10.6, 5.5)		3.85, dd (12.5, 2.4)
	Rha I		**Rha I**		**Rha I**		**Rha I**	
1	99.9	5.38, d (1.5)	100.1	5.22, brs	100.3	5.30, d (1.3)	99.9	5.39, d (1.3)
2	70.2	4.13, dd (3.1, 1.5)	70.4	4.08, dd (2.8-,1.7)	69.5	4.27, dd (3.1, 1.3)	70.2	4.13, m
3	**80.7**	3.90, dd (9.5, 3.1)	**80.6**	3.85, dd (9.7, 2.8)	**81.5**	3.92, dd (9.6, 3.1)	**80.8**	3.88, dd (7.1, 3.1)
4	71.4	3.57, t (9.5)	71.4	3.57, t (9.7)	71.2	3.57, t (9.6)	71.3	3.58, t (9.5)
5	68.6	4.00, m	68.6	3.91, m	68.7	3.98, m	68.6	4.02, m
6	16.6	1.25, d (6.2)	16.6	1.25, d (6.2)	16.8	1.25, d (6.2)	16.7	1.25, d (6.2)
	Xyl II		**Xyl I**		**Glc I**		**Xyl II**	
1	105.0	4.47, d (7.7)	105.1	4.48, d (7.8)	104.2	4.50, d (7.9)	104.9	4.49, d (7.6)
2	74.5	3.40, dd (8.8, 7.7)	74.5	3.40, dd (9.0, 7.8)	74.7	3.41, m	73.7	3.36, m
3	**81.7**	3.49, t (8.8)	**81.6**	3.48, t (9.0)	**82.1**	3.55, t (9.0)	74.5	3.51, t (8.7)
4	68.3	3.53, m	68.3	3.53, m	68.3	3.39, t (9.5)	**76.9**	3.71, m
5	65.7	3.23, dd (11.6, 9.3)	65.7	3.24, dd (11.9, 9.2)	76.3	3.32, m	63.1	3.31, t (11.8)
		3.87, dd (11.6, 5.5)		3.88, dd (11.9, 4.8)				4.04, dd (11.8, 5.4)
					60.9	3.72, dd (12.8, 5.7)		
						3.86, d (12.8)		
	Rha II		**Rha II**		**Rha II**		**Glc I**	
1	100.7	5.21, d (1.1)	100.8	5.21, brs	100.9	5.22, d (2.1)	101.7	4.41, d (7.8)
2	70.8	3.97, dd (3.3, 1.1)	70.9	3.97, m	70.8	3.98, dd (3.3, 2.1)	73.0	3.28, dd (9.1, 7.8)
3	70.9	3.96, dd (9.6, 3.3)	70.8	3.95, dd (9.2, 3.3)	70.9	3.96, dd (9.4, 3.3)	74.4	3.50, t (9.1)
4	**82.3**	3.63, t (9.6)	**82.3**	3.63, t (9.2)	**82.3**	3.64, t (9.4)	**79.0**	3.53, t (9.5)
5	67.1	4.12, m	67.1	4.13, m	67.2	4.11, m	75.1	3.46, m
6	16.5	1.34, d (6.2)	16.6	1.34, d (6.2)	16.6	1.35, d (6.2)	60.1	3.83, dd (12.4, 4.5)
								3.92, dd (12.4, 3.3)
	Glc I		**Glc I**		**Glc II**		**Xyl III**	
1	104.1	4.62, d (7.9)	104.1	4.62, d (7.9)	104.1	4.62, d (7.9)	103.8	4.35, d (7.8)
2	75.1	3.32, m	75.1	3.32, m	75.1	3.32, m	73.9	3.33, m
3	**83.3**	3.53, t (8.9)	**83.3**	3.53, m	**83.3**	3.53, t (9.0)	**82.1**	3.48, t (8.6)
4	68.5	3.39, t (8.9)	68.6	3.39, dd (9.2, 5.2)	68.6	3.39, m	68.3	3.57 m
5	76.5	3.31, m	76.6	3.31, m	76.6	3.31, m	65.7	3.30, t (11.1)
								3.94, dd (11.1, 3.8)
6	61.2	3.73, dd (11.9, 5.2)	61.3	3.73, dd (12.0, 4.7)	61.0	3.72, dd (12.8, 4.8)		
		3.86, m		3.88, dd (12.0, 2.4)		3.86, dd (12.8)		
	Rha III		**Rha III**		**Rha III**		**Rha II**	
1	101.3	5.16, d (1.7)	101.3	5.15, d (1.4)	101.3	5.15, d (1.4)	101.2	5.18, d (1.5)
2	70.9	3.97, dd (3.3-1.7)	70.9	3.97, dd (3.5, 1.4)	70.9	3.97, m	70.9	3.95, dd (3.3, 1.5)
3	70.8	3.71, dd (9.0-3.3)	70.8	3.72, dd (9.5, 3.5)	70.8	3.72, dd (9.6, 3.1)	70.8	3.72, dd (9.5-3.3)
4	72.5	3.42, t (9.0)	72.5	3.42, t (9.5)	72.5	3.42, t (9.6)	72.6	3.41, t (9.5)
5	68.7	4.01, m	68.7	4.00, m	68.7	4.00, m	68.5	4.04, m
6	16.5	1.27, d (6.2)	16.5	1.27, d (6.2)	16.5	1.27, d (6.3)	16.5	1.26, d (6.2)
C-28	**Glc II**		**Glc II**		**Glc III**		**Glc II**	
1	94.3	5.37, d (8.3)	94.3	5.37, d (8.2)	94.3	5.36, d (8.2)	94.3	5.37, d (8.2)
2	72.4	3.35, m	72.4	3.35, t (8.5)	72.4	3.35, m	72.4	3.35, m
3	76.7	3.42, t (8.7)	76.7	3.43, t (8.6)	76.7	3.43, m	76.7	3.43, m
4	69.5	3.44, t (8.7)	69.5	3.45, t (8.8)	69.5	3.46, t (8.6)	69.5	3.44, t (8.7)
5	76.4	3.52, m	76.4	3.52, m	76.4	3.52, m	76.4	3.52, m
6	**68.0**	3.78, dd (11.6, 4.9)	**68.0**	3.78, dd (11.5,5.1)	**68.0**	3.78, dd (11.5, 4.7)	**68.0**	3.77, dd (11.6, 4.9)
		4.14, dd (11.6, 1.4)		4.14, dd (11.5, 1.8)		4.13, m		4.14, dd (11.6, 1.7)
	Glc III		**Glc III**		**Glc IV**		**Glc III**	
1	103.2	4.36, d (7.6)	103.2	4.36, d (7.8)	103.2	4.36, d (7.8)	103.2	4.36, d (7.8)
2	73.7	3.23, t (7.8)	73.7	3.23, dd (9.2, 7.8)	73.7	3.23, dd (8.9, 7.8)	73.7	3.23, dd (9.1, 7.8)
3	76.6	3.36, m	76.6	3.37, t (9.2)	76.6	3.37, m	76.6	3.37, m
4	70.1	3.31, t (8.9)	70.1	3.30, t (9.2)	70.1	3.31, t (8.6)	70.1	3.31, m
5	76.6	3.27, m	76.6	3.26, m	76.6	3.26, m	76.6	3.26, m
6	61.3	3.66, dd (11.9, 5.6)	61.3	3.68, dd (12.0,5.6)	61.3	3.68, dd (11.9, 5.6)	61.3	3.68, dd (12.0, 5.5)

3.86, m	3.87, dm (12.0)	3.86, m	3.85, dd (12.0, 2.4)

[a] in ppm, *J* in parentheses in Hz

Table 3 ^{13}C NMR and ^{1}H NMR spectroscopic data of the aglycone moieties of compounds **6**–**8** and **13**in CD_3OD.[a]

position	6		7		8		13	
	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H
1	38.7	1.00, td (13.9, 3.3) 1.63, m	38.6	1.00, m 1.63, m	38.5	1.00, m 1.64, m	38.6	1.00, td (13.9, 3.3) 1.63, m
2	25.9	1.70, m 1.83, m	25.7	1.86, m 1.76, m	25.7	1.73, m 1.86, m	25.9	1.70, m 1.83, m
3	88.8	3.14, dd (11.5, 4.3)	89.2	3.14, dd (11.5, 4.2)	89.2	3.14, dd (11.6, 4.2)	88.9	3.14, dd (11.5, 4.1)
4	38.9	-	38.9	-	38.9	-	38.9	-
5	55.9	0.80, d (12.1)	55.7	0.80, m	55.7	0.80, m	55.8	0.80, m
6	18.0	1.43, m 1.56, m	18.0	1.42, m 1.56, m	18.0	1.42, td (13.8, 3.0) 1.55, m	18.0	1.43, m 1.56, m
7	32.5	1.34, m 1.50, td (12.6, 3.3)	32.5	1.32, m 1.50, td (11.6, 4.5)	32.5	1.34, m 1.50, td (12.0, 3.1)	32.5	1.34, m 1.50, m
8	39.3	-	39.3	-	39.3	-	39.3	-
9	47.8	1.60, m	47.8	1.60, m	47.9	1.60, m	47.8	1.60, m
10	36.5	-	36.5	-	36.5	-	36.5	-
11	23.1	1.90, m	23.2	1.92, m	23.2	1.92, m	23.2	1.90, m
12	122.4	5.27, t (3.6)	122.4	5.27, t (3.5)	122.4	5.27, t (3.6)	122.4	5.27, t (3.5)
13	143.5	-	143.5	-	143.5	-	143.5	-
14	41.5	-	41.5	-	41.5	-	41.5	-
15	27.3	1.10, dm (14.2) 1.80, m	27.5	1.11, dm (13.9) 1.80, td (13.9, 3.2)	27.5	1.10, dm (14.1) 1.80, td (14.1, 3.3)	27.5	1.10, dm (14.0) 1.80, m
16	22.6	1.73, m 2.07, td (13.4, 3.7)	22.6	1.73, m 2.07, td (13.2, 3.4)	22.6	1.73, m 2.07, td (13.2, 3.3)	22.6	1.73, m 2.07, td (13.4, 3.7)
17	46.6	-	46.6	-	46.6	-	46.6	-
18	41.1	2.88, dd (13.6, 4.1)	41.2	2.88, dd (13.5, 3.7)	41.1	2.88, dd (13.6, 4.3)	41.1	2.88, dd (13.7, 4.1)
19	45.9	1.17, m 1.73, t (13.6)	45.8	1.17, m 1.73, t (13.5)	46.6	1.16, m 1.73, t (13.6)	45.8	1.17, m 1.73, t (13.7)
20	30.2	-	30.2	-	30.1	-	30.2	-
21	33.5	1.24, m 1.42, td (12.9, 3.3)	33.5	1.24, m 1.42, td (13.4, 3.4)	33.5	1.24, m 1.42, td (13.4, 3.0)	33.5	1.24, m 1.42, td (13.2, 4.2)
22	31.8	1.62, m 1.74, td (13.6, 3.5)	31.8	1.62, m 1.74, m	31.8	1.62, m 1.74, td (13.7, 4.6)	31.8	1.62, m 1.74, td (13.6, 3.5)
23	27.2	1.07, s	27.2	1.05, s	27.2	1.06, s	27.1	1.07, s
24	15.9	0.88, s	15.8	0.88, s	15.7	0.87, s	15.8	0.87, s
25	14.8	0.97, s	14.8	0.98, s	14.8	0.98, s	14.8	0.97, s
26	16.4	0.82, s	16.4	0.82, s	16.4	0.82, s	16.4	0.82, s
27	25.0	1.17, s	24.9	1.19, s	24.9	1.17, s	24.9	1.17, s
28	176.7	-	176.7	-	176.7	-	176.7	-
29	32.1	0.93, s	32.1	0.93, s	32.1	0.93, s	32.1	0.93, s
30	22.7	0.97, s	22.6	0.96, s	22.6	0.96, s	22.6	0.96, s

[a] in ppm, *J* in parentheses in Hz

Table 4 ^{13}C NMR and ^{1}H NMR spectroscopic data in CD_3OD of the sugar moieties of compounds **6-8** and **13**.[a,]

Position	**6**		**7**		**8**		**13**	
	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H
C-3	**Xyl I**		**Ara**		**Ara**		**Xyl I**	
1	105.1	4.39, d (7.1)	103.9	4.50, d (6.2)	104.2	4.47, d (6.0)	105.2	4.39, d (7.1)
2	**76.7**	3.41, t (7.7)	**74.9**	3.78, m	**75.5**	3.71, m	**77.0**	3.41, m
3	77.5	3.45, t (8.7)	72.4	3.73, dd (7.7, 3.6)	72.2	3.72, dd (6.3, 4.6)	77.5	3.45, m
4	70.2	3.48, m	67.8	3.79, m	67.9	3.79, m	70.3	3.48, m
5	65.2	3.18, dd (11.4, 9.9)	63.4	3.52, dd (10.8, 4.7)	63.7	3.52, dd (12.0, 1.3)	65.2	3.19, t (10.7)
		3.85, m		3.85, d (10.8)		3.85, m		3.84, m
	Rha I		**Rha I**		**Rha I**		**Rha I**	
1	99.9	5.38, d (1.7)	100.1	5.22, d (1.6)	100.4	5.17, d (1.3)	100.3	5.30, d (1.3)
2	70.2	4.13, dd (3.2, 1.7)	70.4	4.08, dd (2.9,1.6)	69.8	4.23, dd (3.2, 1.3)	69.5	4.27, dd (2.4, 1.3)
3	80.7	3.90, dd (9.4-3.2)	**80.6**	3.85, dd (9.6, 2.9)	**81.4**	3.88, dd (9.5, 3.2)	**81.5**	3.93, dd (9.6, 2.4)
4	71.4	3.58, t (9.6)	71.4	3.57, t (9.6)	71.2	3.58, t (9.5)	71.2	3.58, t (9.6)
5	68.6	3.99, m	68.6	3.91, m	68.7	3.91, m	68.5	4.01, m
6	16.7	1.25, d (6.2)	16.7	1.25, d (6.2)	16.5	1.25, d (6.2)	16.7	1.25, d (6.2)
	Xyl II		**Xyl I**		**Glc I**		**Glc I**	
1	105.1	4.47, d (7.7)	105.1	4.48, d (7.9)	104.3	4.51, d (7.7)	104.2	4.51, d (7.7)
2	74.5	3.40, dd (8.7, 7.7)	74.6	3.40, m	74.7	3.42, m	74.7	3.40, dd (9.0, 7.7)
3	**81.6**	3.49, t (8.7)	**81.6**	3.48, t (8.9)	**82.0**	3.55, t (9.0)	**82.0**	3.53, t (9.0)
4	68.3	3.53, m	68.3	3.53, m	68.3	3.39, m	68.3	3.39, t (8.8)
5	65.7	3.22, dd (11.1, 9.2)	65.7	3.23, t (10.6)	76.3	3.33, m	76.3	3.33, m
		3.87, m		3.89, dd (10.6, 2.9)				
6					60.9	3.72, dd (11.6, 4.4)	60.9	3.72, dd (11.6, 3.3)
						3.86, m		3.85, dd (11.6, 2.0)
	Rha II		**Rha II**		**Rha II**		**Rha II**	
1	100.7	5.21, brs	100.7	5.21, brs	100.9	5.22, d (1.6)	100.9	5.22, d (1.2)
2	70.8	3.97, m	70.8	3.97, dd (2.8, 1.2)	70.8	3.98, dd (3.1, 1.6)	70.8	3.98, m
3	70.8	3.95, dd (9.4, 3.3)	70.9	3.95, dd (9.3, 2.8)	70.8	3.96, m	70.9	3.96, dd (9.5, 3.0)
4	**82.6**	3.61, t (9.4)	**82.6**	3.61, t (9.3)	**82.6**	3.61, t (9.5)	**82.6**	3.61, t (9.5)
5	67.1	4.13, m	67.1	4.13, m	67.2	4.12, m	67.2	4.12, m
6	16.7	1.34, d (6.1)	16.6	1.34, d (6.2)	16.7	1.35, d (6.1)	16.7	1.35, d (6.1)
	Glc I		**Glc I**		**Glc II**		**Glc II**	
1	104.4	4.62, d (7.8)	104.4	4.62, d (7.9)	104.4	4.62, d (7.8)	104.4	4.62, d (7.8)
2	76.3	3.41, m	76.3	3.40, m	75.8	3.41, m	76.2	3.41, m
3	**76.1**	3.80, t (9.2)	**76.1**	3.80, t (9.2)	**76.1**	3.80, t (9.1)	**76.1**	3.80, t (9.2)
4	**72.9**	3.83, m	**72.9**	3.83, t (9.3)	**72.9**	3.81, t (9.1)	**73.0**	3.83, t (9.2)
5	75.8	3.41, m	75.8	3.40, m	76.2	3.41, m	75.8	3.41, m
6	59.7	3.87, m	59.6	3.87, dd (12.2, 2.9)	59.7	3.88, m	59.6	3.88, dd (12.9, 4.1)
		3.95, dd (12.9, 2.2)		3.95, m		3.96, dd (12.8, 2.9)		3.95, m
	Rha III		**Rha III**		**Rha III**		**Rha III**	
1	99.5	5.39, d (2.1)	99.5	5.39, d (1.7)	99.5	5.39, d (1.5)	99.5	5.40, brs
2	71.0	3.91, dd (3.4, 2.1)	71.0	3.90, dd (3.1, 1.7)	71.0	3.91, dd (3.2, 1.5)	71.1	3.91, dd (3.2, 1.3)
3	70.3	3.95, dd (9.3, 3.4)	70.3	3.94, dd (9.2, 3.1)	70.3	3.95, dd (9.1, 3.2)	70.3	3.95, dd (9.3, 3.2)
4	73.3	3.32, m	73.4	3.33, m	73.4	3.32, m	73.9	3.33, m
5	67.7	4.50, m	67.7	4.46, m	67.7	4.51, m	67.6	4.50, m
6	16.6	1.26, d (6.3)	16.5	1.26, d (6.2)	16.5	1.27, d (6.0)	16.5	1.27, d (6.2)
	Gal		**Gal**		**Gal**		**Gal**	
1	102.9	4.46, d (7.6)	102.9	4.46, d (7.7)	102.9	4.46, d (8.2)	102.9	4.46 d (7.6)
2	71.8	3.45, m	71.7	3.45, m	71.7	3.46, m	71.7	3.46, m
3	73.6	3.45, m	73.6	3.46, m	73.6	3.46, m	73.6	3.45, m
4	68.8	3.85, m	68.8	3.84, m	68.9	3.85, d (2.3)	68.8	3.84, m
5	75.3	3.51, m	75.3	3.51, m	75.3	3.52, m	75.3	3.51, m
6	60.7	3.79, m	60.7	3.80, m	60.7	3.80, m	60.7	3.80, m
		3,81, m						
C-28	**Glc II**		**Glc II**		**Glc III**		**Glc III**	
1	94.3	5.37, d (8.2)	94.3	5.37, d (8.2)	94.3	5.36, d (8.2)	94.3	5.36, d (8.2)
2	72.4	3.35, m	72.4	3.35, m	72.4	3.35, m	72.4	3.35, m
3	76.7	3.43, t (8.6)	76.7	3.43, t (9.0)	76.7	3.43, t (9.1)	76.7	3.41, t (8.7)
4	69.5	3.45, t(8.6)	69.5	3.46, t (9.0)	69.5	3.46, t (9.0)	69.5	3.43, t (8.7)
5	76.4	3.52, m	76.4	3.53, m	76.4	3.52, m	76.4	3.52, m
6	**68.0**	3.77, dd (11.2, 4.5)	**68.0**	3.78, m	**68.0**	3.78, dd (11.6, 4.4)	**68.0**	3.78, dd (11.7, 4.7)
		4.13, dd (11.2 ,1.8)		4.13, dd 11.7, 1.8)		4.14, dd (11.6, 1.7)		4.14, dd (11.7, 1.9)

	Glc III		GlcIII		GlcIV		Glc IV	
1	103.2	4.36, d (7.8)	103.2	4.36, d (7.8)	103.2	4.36, d (7.8)	103.2	4.36, d (7.8)
2	73.7	3.23, t (7.8)	73.7	3.23, dd (9.2, 7.8)	73.7	3.23, dd (9.1, 7.8)	73.7	3.23, dd (9.0, 7.8)
3	76.6	3.35, m	76.6	3.37, m	76.6	3.37, m	76.5	3.37, m
4	70.1	3.31, t (8.7)	70.1	3.31, t (8.7)	70.1	3.31, t (8.6)	70.1	3.31, t (8.7)
5	76.5	3.25, m	76.6	3.26, m	76.6	3.27, m	76.6	3.25, m
6	61.3	3.68, dd (12.0, 5.6)	61.3	3.68, dd (11.8, 5.5)	61.3	3.68, dd (11.8, 5.5)	61.3	3.67, dd (11.9, 5.5)
		3.86, dm (12.0)		3.87, m		3.87, m		3.86, dd (11.9, 2.0)

[a] in ppm, *J* in parentheses in Hz

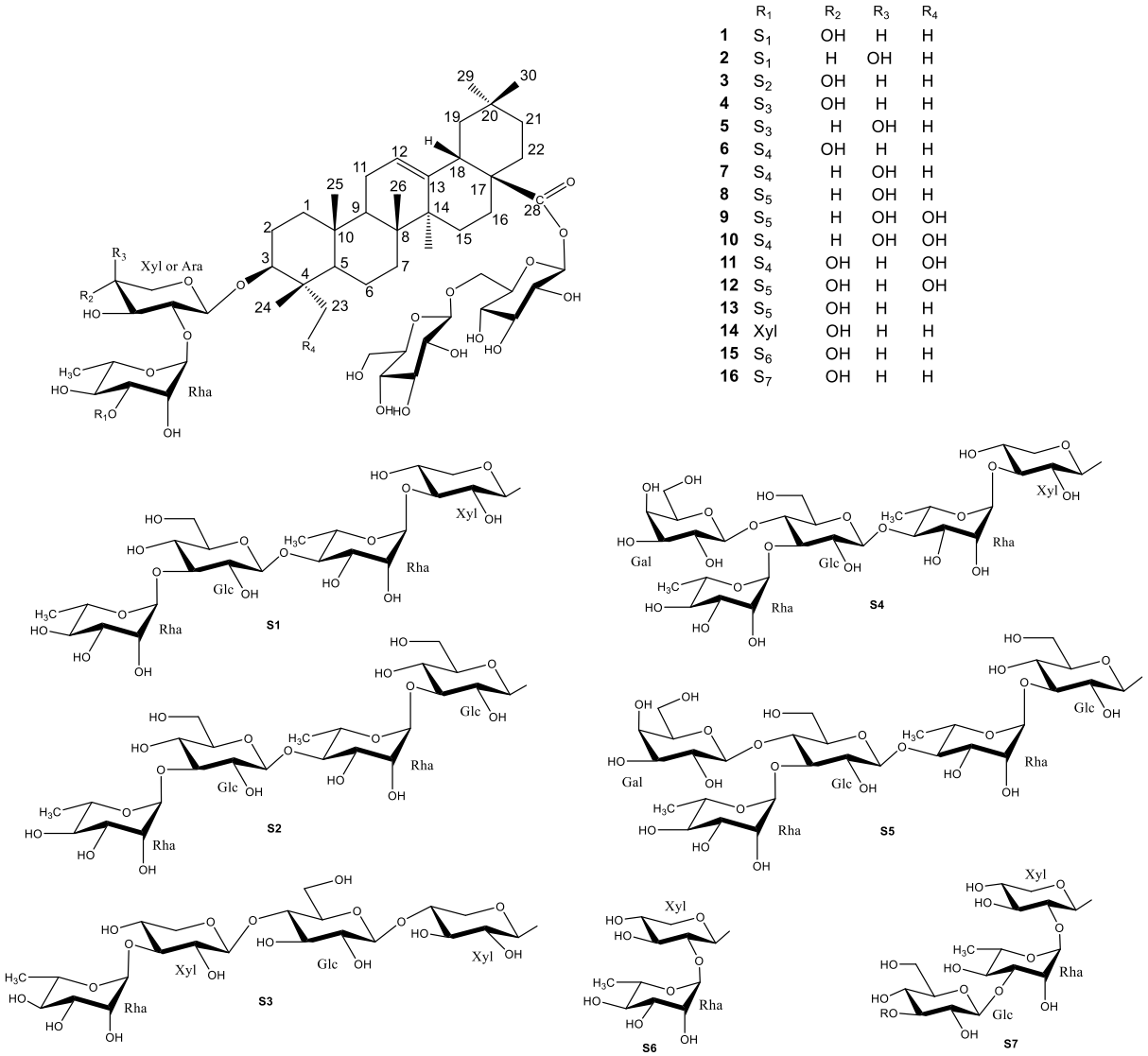

Fig. 1. The structures of compounds **1-16** isolated from *Scabiosa semipapposa*.

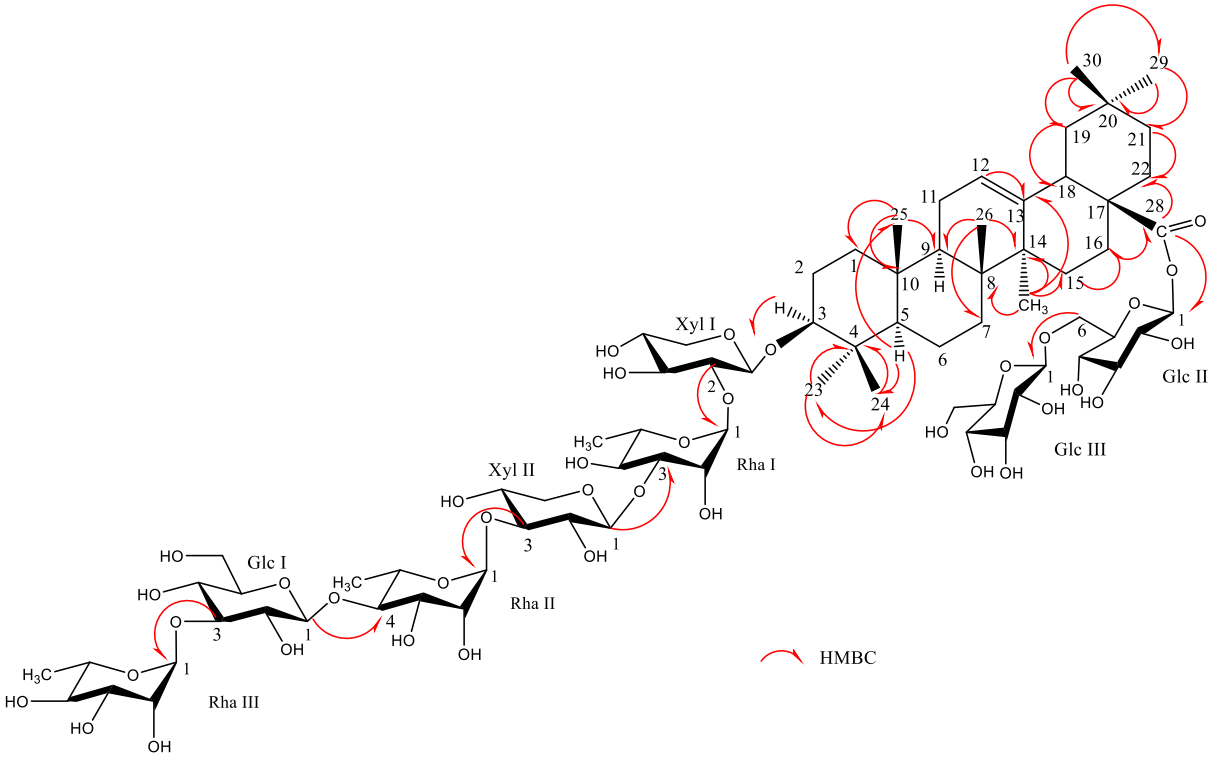

Fig. 2. Key HMBC correlations for compound 1.

Figure captions

Fig. 1. The structures of compounds **1-16** isolated from *Scabiosa semipapposa*.

Fig. 2. Key HMBC correlations for compound 1.