Research Article

New Compounds from *Bassia muricata* and *Fagonia indica*

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ABSTRACT

Six compounds were isolated and identified from *Bassia muricata* and *Fagonia indica*, two compounds are new natural products. Three metabolites, 3,4-dimethoxytoluene (1), 3′-methylquercetin (2) and a new flavonoid glycoside, 3-*O*-[α-L-arabinopyranosyl-(1→2)-L-α-arabinopyranosyl)]-3′-methylquercetin (3) were isolated from *Bassia muricata*. From *Fagonia indica* one flavonoid glycoside; 3-*O*-[α-L-rhamnopyranosyl-(1→6)-β-D-glucopyranosyl]-kaempferol (4) along with a new triterpenoid saponin; 28-*O*-[β-D-glucopyranosylester-(1→3)- β-D-glucopyranosyl] oleanolic acid (5) and known quinovic acid-3-*O*-(α-L-rahmnopyranosyl-)-28-*O*-β-D-glucopyranosly ester (6) have been isolated. The structure elucidation was performed by nuclear magnetic resonance (NMR) spectroscopic and mass spectrometric methods. Antioxidant activities of pure compounds were measured by DPPH (1,1-diphenyle-2-picryl-hydrazyl) and Xanthine – xanthine oxidase assays.

Keywords: Flavonoid glycosides, saponins, *Bassia muricata*, *Fagonia indica*., Antioxidant activity.

INTRODUCTION

Zygophyllaceae and Chenopodiaceae families are rich with medicinal plants.[1] The medicinal properties of the plant are due to its variety of the active phytochemical constituents. *Bassia muricata* (Chenopodiaceae) is used in folk medicine to treat renal and rheumatic diseases[2] and possess different degrees of antiinflammatory, analgesic and antipyretic effects as well as antispasmodic property.[3] *Fagonia indica Burm. f.* (Zygophyllaceae) is a small spiny shrub, mostly found in the deserts of Asia and Africa.[4] Plants belonging to the genus *Fagonia* are often used in folk medicine, mainly as a popular remedy for the treatment of various skin lesions.[5] It is used in folk medicine for cancer as well as most of the disorders considered to be due to poisons.[6] Species of *Fagonia* have been found to contain proteins and amino acids,[7] alkaloids,[8] flavonoids,[9] terpenoids,[10] saponins.[11] This study describes isolation and structure determination of two new compounds along with four known metabolites from *Bassia muricata* and *Fagonia indica* grown in Aseer region (Saudi Arabia) and evaluation of antioxidant activities of the pure compounds.

MATERIALS AND METHODS

General

Optical rotations were measured on a JASCO-1030 Polarimeter (JASCO, Germany). ^{1}H NMR (500.13 MHz), ^{13}C NMR (125.75 MHz) and 2D NMR spectra were measured on Bruker DRX 500 spectrometer (Bruker Biospin, Rheinstetten, Germany). Liquid chromatography electrospray ionization mass spectra (LCESIMS) (positive mode) were measured with a HP1100 HPLC chromatograph (*Hewlett Packard*, Avondale, PA, USA) coupled to an Orbitrap XL (*Thermo-Fisher*, San Jose, CA, USA) equipped with an electrospray (ESI) source. Column chromatography (CC): silica gel (40-0.063 ± 0.2 µm, Merck, Darmstadt, Germany). Thin layer chromatography (TLC): silica gel (0.25 and 1 mm precoated plates 60 F254, Merck. Polyamide 6 for CC (particle size 50- 160 µm - Fluka, Germany). Sephadex LH-20 (Sigma-Aldrich, Germany).

Plant Material

The aerial parts of *Bassia muricata* and *Fagonia indica* were collected from Abha city, Aseer region, Saudi Arabia, in April 2011. The plants were identified by Dr. Mahmoud Fawzy, Biology department, college of science, King Khalid University. A plant sample had been deposited in the Herbarium of Botany department, college of science, King Khalid University with the numbers of Bm042011 for *Bassia muricata* and Fi042011 for *Fagonia indica*.

Isolation

Bassia muricata

The air dried plant (1.5 kg) was exhaustively extracted with 85% ethanol (20.0 litters). The ethanol extract was concentrated under reduced pressure at temperature 40°C. The ethanol extract (77.08 g) was partitioned using petroleum ether, ethyl acetate and *n*-butanol/water. The ethyl acetate fraction (5.68 g) was subjected to column chromatography on silica gel and eluted with chloroform to afford sub-fraction (26.0 mg). This fraction was purified on silica gel column eluted with chloroform /methanol (9:1) to afford pure compound **1** (20.9 mg). The *n*-butanol fraction (8.37 g) was subjected to column chromatography on polyamide material, eluted with water, water/methanol. The fractions eluted with water/methanol (8.5:1.5) afforded two major impure

compounds (255.2 mg) which were purified on Sephadex LH-20 using methanol as eluent to give the pure compounds **2** (5.8 mg) and **3** (25.7 mg).

Fagonia indica

The air dried plant (2.5 kg) was exhaustively extracted with 85% ethanol (17.0 litters). The ethanol extract was concentrated under reduced pressure at temperature 40°C. The ethanol extract (602 g) was partitioned using petroleum ether, ethyl acetate and *n*-butanol/water. The *n*-butanol fraction (105 g) was subjected to column chromatography using 53 g only on silica gel and the fraction eluted with chloroform/methanol/H_2O (7:5.5:1) afforded two sub-fractions. The first fraction (838 mg) was subjected to column chromatography on polyamide material and eluted with water/methanol (2.5:7.5) afforded semi-pure compound **4**. The purification was performed on Sephadex LH-20 column using methanol, to yield pure compound **4** (5.5 mg). The fractions eluted with water/methanol (7.5:2.5) and (6.6:4.3) afforded pure compound **5** (21.2 mg). The second fraction (1.21 g) was purified on silica gel column, and eluted with chloroform/methanol/H_2O (7:5.5:1), to yield pure compound **6** (250 mg).

Spectroscopic Data

*3-O-[α-L-Arabinopyranosyl-(1→2)-α-L-arabinopyranosyl)]-3′-O-methyl quercetin (**3**):* Yellow amorphous, $[\alpha]^{25}_D$ -93.62° (MeOH, *c* 0.34), HRESIMS: *m/z* 581.1487 ($[M+H]^+$, $C_{26}H_{28}O_{15}$, calcd. 581.1484); UV (MeOH) λ_{max}: 358, 270, 255; (NaOMe): 415, 330, 270; ($AlCl_3$): 407, 303, 270; ($AlCl_3$/HCl): 407, 303, 270; (NaOAc): 371, 319, 275; (NaOAc/H_3BO_3): 359, 270, 255. ^{1}H and ^{13}C NMR (Table 1).

*28-O-[β-D-Glucopyranosylester-(1→3)-β-D-glucopyranosyl] oleanolic acid (**5**):* White amorphous solid, $[\alpha]^{25}_D$ +22.13° (MeOH, *c* 0.23), HRESIMS: *m/z* 781.4729 ($[M+H]^+$, $C_{42}H_{69}O_{13}$, calcd. 781.4723); ^{1}H and ^{13}C NMR (Table 1).

Acid Hydrolysis

Compound **3** (10 mg) in EtOH (15 ml) and 5% HCl (15 ml) was heated at 80°C for 3.5 h. The EtOH was evaporated and the mixture was neutralized with 5% aq. K_2CO_3. The mixture was extracted with ethyl acetate (3 x 15 ml) to give the aglycone (1.2 mg) which was identified as 3'-methyl quercetin **2**.[12] The water was removed by azeotropic distillation with *n*-BuOH under reduced pressure. The sugar was compared with standard sugars on a TLC plate, with $CHCl_3$-MeOH-AcOH-H_2O (8:3:5:2) as developing solvent system[13] and confirmed with *n*-BuOH–EtOAc–iso-PrOH– AcOH–H_2O (7:20:12:7:6) with the anisaldehyde reagent and the R_f value of arabinose was 0.38.[14] The repeated precipitation with MeOH yielded α-arabinose (1.41 mg) with $[\alpha]_D^{25}$ = + 91.3 (c, 0.135, H_2O). The literature reported of optical rotation was +103 where the positive optical rotation indicated that this sugar is L-arabinose.[15]

Antioxidant Activities

DPPH radical scavenging activity

The DPPH (1,1-diphenyl-2-picrylhydrazyl) radical scavenging activity of pure compounds was evaluated. 1 ml of methanolic solution of varying concentration samples (25, 50 and 100 µg/ml) were added to 1 ml of methanol solution of DPPH (60µM). The prepared solutions were mixed and left for 30 min at room temperature. The optical density was measured at 520 nm using a spectrophotometer (UV-1650PC Shimadzu, Japan). The percent scavenging effect was determined by comparing the absorbance of solution containing the test sample to that of control solution without the test sample taking the corresponding blanks. Mean of three measurements for each compound was calculated according to the method of Matsushige *et al*.[16]

Superoxide anion scavenging activity

Superoxide anion scavenging activity was determined according to a modified method of Matsushige *et al*.[16] Reaction mixtures containing 1.4 mL of 50 mM Na_2CO_3 (pH 10.2), 100 µL of 3 mM xanthine, 100 µL of 3 mM EDTA, 100 µL of BSA (1.5 mg/mL), 100 µL of 75 mM nitro blue tetrazonium, and 50 µL of the samples were preincubated at 30 °C for 10 min, and 50 µL of xanthine oxidase (0.3 unit/mL) was added. After incubation at 30°C for 20 min, 200 µL of 6 mM $CuCl_2$ was added to stop the reactions and the absorbance was measured at 560 nm.

RESULTS AND DISCUSSION

Isolation

The air-dried parts of both plants were exhaustively extracted with ethanol then partitioned using petroleum ether, ethyl acetate and *n*-butanol/water. The ethyl acetate fraction of *B. muricata* was subjected to successive column chromatography on silica gel to afford compound **1**. The *n*-butanol fraction of *B. muricata* was subjected to polyamide column followed by Sephadex LH-20 to afford compounds **2** and **3**. Column chromatography for *n*-butanol fraction of *F. indica* using silica gel gave two sub-fractions. The first fraction was purified on polyamide followed by Sephadex LH-20 to yield compound **5** and semi pure of compound **4** which was purified on Sephadex LH-20. The second fraction was purified on silica gel column, and eluted with chloroform/methanol/H_2O (7:5.5:1), to yield pure compound **6**.

Structure Determination

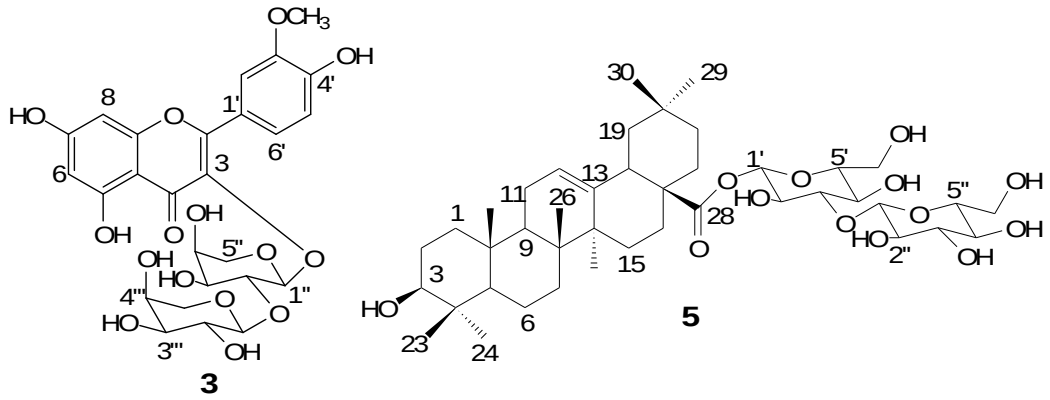
On the basis of mass spectrometric**,** 1D and 2D NMR spectroscopic data, compound **1** was identified as 3,4-dimethoxytoluene which previously isolated as a major component of *Phoenix dactylifera*.[17] It is the first time for compound **1** to be isolated from *Fagonia inidca*.

The ESIMS of compound **2** exhibited to $[M-H]^-$ at *m/z* 315. The analysis of 1D and 2D-NMR leads to the structure of compound **2** as 3'-methyl quercetin which showed

complete agreement with published data of Freitas et al.[12]

The molecular formula of compound **3** ($C_{26}H_{28}O_{15}$) was deduced from HRESIMS *m/z* 581.1487 ([M+H]$^+$). The UV spectra in methanol showed two major absorption bands at 358 nm and at 255 nm for band I and band II respectively, typical characteristic absorption spectra for flavonol.[18] The addition of sodium methoxide to the methanol solution resulted in a bathochromic shift of 57 nm with an increase in intensity of absorption in band I, indicating the presence of free 4'-hydroxyl group. The bathochromic shift of 49 nm in band I in the presence of $AlCl_3$ without any change by addition of HCl indicates the presence of free 5- hydroxyl group. A bathochromic shift of 20 nm in band II upon addition of sodium acetate (NaOAc) indicates the presence of free 7-hydroxyl group, and no changes by addition of H_3BO_3 to NaOAc, suggested the absence of ortho-dihydroxyl group in the B-ring.

Comparison of ^{1}H and ^{13}C-NMR data of **3** with those of **2** showed the same flavonoid aglycone (3'-methyl quercetin). The presence of two anomeric protons at δ 5.24 (d, *J* = 8.0 Hz) and δ 4.04 (d, *J* = 7.2 Hz) indicates to the presence of two sugar moieties (Table 1).



Structure of the new compounds 3 and 5

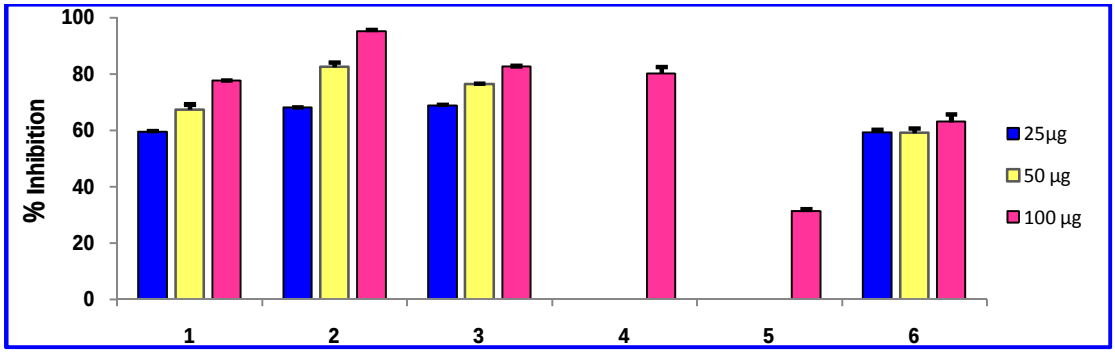


Figure 1: Free radical scavenging activity of pure compounds in the DPPH radical assay. Values are mean ±SD.

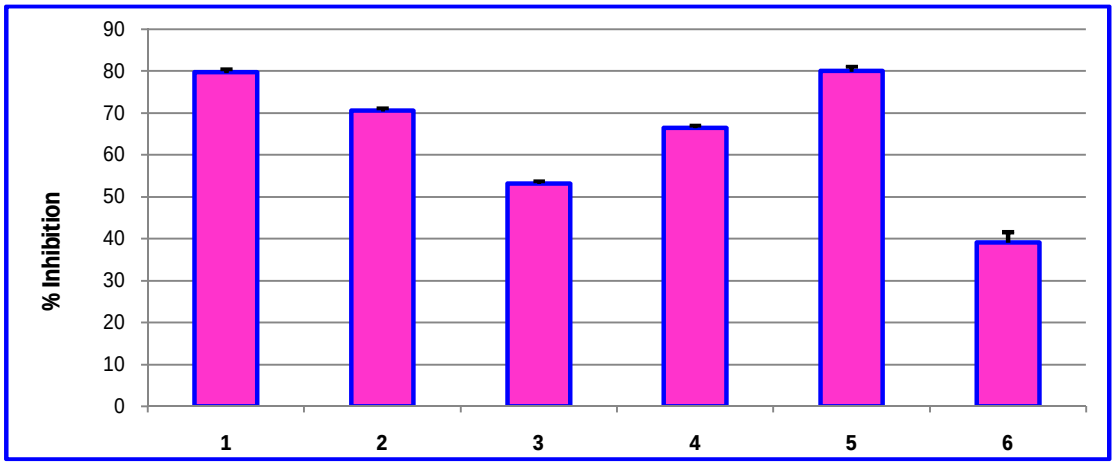


Figure 2: Free radical scavenging activity of pure compounds (50 µg/ml) in (Xanthine – xanthine oxidase assay). (Values are mean ±SD.)

Table 1: ^{1}H- and ^{13}C-NMR spectroscopic data of compounds 3 and 5 (CD_3OD, 500 MHz for ^{1}H and 125 MHz for ^{13}C).

3			**5**		
Position	**δ_H (mult., *J* in Hz)**	**δ_C**	**Position**	**δ_H (mult., *J* in Hz)**	**δ_C**
2		158.5	**1**	1.01/1.68	38.5
3		135.6	**2**	1.48/1.59	22.9
4		179.6	**3**	3.16, dd, *J* = 4.5, 10.4	77.7
5		163.1	**4**		40.4
6	6.20 (d, *J* = 2.0)	100.0	**5**	1.41	52.0
7		166.1	**6**	1.32/1.45	18.7
8	6.42 (d, *J* = 2.0)	95.0	**7**	1.44/1.27	34.9
9		158.4	**8**		40.1
10		105.7	**9**	1.21	50.5
1′		123.0	**10**		37.9
2′	8.12 (d, *J* = 2.0)	114.5	**11**	1.61	26.9
3′		151.0	**12**	5.22 br.d, *J* = 10 Hz	125.5
4′		148.5	**13**		144.1
5′	6.91 (d, *J* = 8.5)	116.1	**14**		42.2
6′	7.64 (dd, *J* = 8.5, 2.0)	123.6	**15**	1.76/1.11	28.7
OCH_3	3.97, s	57.0	**16**	1.90	26.4
1″	5.24 (d, *J* = 8.0)	104.5	**17**		50.3
2″	3.70	76.7	**18**	3.12	40.4
3″	3.33	72.3	**19**	1.21	48.5
4″	3.61	69.6	**20**		30.8
5″	3.69/3.15	66.9	**21**	1.43/1.39	34.9
1‴	4.04 (d, *J* = 7.2)	104.6	**22**	2.10/1.42	33.1
2‴	3.84	73.2	**23**	1.22	30.9
3‴	3.12	74.1	**24**	0.91	15.4
4‴	3.82	70.4	**25**	0.88	16.6
5‴	3.80/3.71	68.7	**26**	0.86	17.5
			27	1.01	23.9
			28		176.0
			29	1.20	33.0
			30	1.00	23.0
			1′	5.42 (d, *J* = 8.1)	95.3
			2′	3.19	77.7
			3′	3.67	82.8
			4′	3.38	71.2
			5′	3.39	78.0
			6′	3.68/3.80	62.6
			1″	4.52 (d, *J* = 8.2)	105.3
			2″	3.10	76.0
			3″	3.40	78.4
			4″	3.30	71.7
			5″	3.32	78.6
			6″	3.70/3.81	62.8

Assignments were based on the ^{1}H NMR, COSY, HSQC, and HMBC experiments. Overlapped ^{1}H NMR signals are reported without designated multiplicity.

The coupling constants (*J* = 7.2-8.0 Hz) of the anomeric protons, indicated to the *trans*-diaxial correlation between H-1″ and H-2″ which reflects the β-configuration of the sugar units. The acid hydrolysis affords compound **2** and arabinose moiety which was identified by TLC with standard sugars.[14] The specific optical rotation of the arabinose was positive value (+91.3) which is similar to the previously reported (+103) for L-arabinose.[15] By ^{1}H-^{1}H COSY-45 and HSQC the two sugars were confirmed as arabinose moieties which showed agreement with the reported ^{13}C-data of the α-L-arabinose moiety.[19] The attachment of the sugar to the aglycon and the inter-

carbohydrate linkage were determined by an HMBC experiment. The HMBC correlations through three bonds between H-1''/C-3 and H-1'''/C-2'' indicated attachment of the di-arabinosyl moiety to C-3 of the aglycone and the terminal arabinose to C-2 of the first arabinose. Therefore, the assumed structure of compound **3** is 3-*O*-[α-L-arabinopyranosyl-(1→2)-α-L-arabinopyranosyl)]-3′-*O*-methyl quercetin which reported here for the first time. A similar compound with β-D-xylose moieties was previously isolated from *Astragalus galegiformis*.[20]

The ESIMS spectrum of compound **4** recorded in the negative ionization mode exhibited a quasimolecular ion at *m/z* 593.09 $[M-H]^-$. This result together with NMR data suggested the molecular formula of $C_{27}H_{30}O_{15}$. The NMR data reflects a flavonoid glycoside nature of compound **4** by presence of AB and AA'XX' spin systems for ring A and B respectively. The sugar moieties were confirmed by two anomeric protons. The sugars linkages were determined by HMBC experiment. The spectroscopic data confirmed the structure of compound **4** as 3-*O*-[α-L-rhamnopyranosyl-(1→6)-β-D-glucopyranosyl]-kaempferol which previously was isolated from petals of *Catharanthus roseus*.[21]

The HRESIM mass spectrum of compound **5** exhibited an ion at *m/z* 781.4729 ($[M+H]^+$) corresponding to a molecular formula of $C_{42}H_{68}O_{13}$. The ^{1}H-NMR spectrum showed the existence of seven tertiary methyl groups (Table 1). The oxymethine signal at δ 3.16 was assigned to H-3 while the olefinic proton of H-12 appeared at δ 5.22. Analysis of 1D and 2D-NMR spectra led to the identification of the aglycone as oleanolic acid.[11] The presence of two protons at δ 5.42 (d, *J* = 8.1 Hz) and δ 4.52 (d, *J* = 8.2 Hz) with correlations in the HSQC spectrum for two anomeric carbons at δ 95.3 and 105.3 indicates the presence of two sugar moieties. The values of the coupling constants $^3J_{H\text{-}1'\text{-}H\text{-}2'}$ = 8.1 Hz and $^3J_{H\text{-}1''\text{-}H\text{-}2''}$ = 8.2 Hz reflect the β-configuration of the two sugar moieties. The ^{13}C-chemical shifts of the anomeric carbons indicate that one sugar bonded by acetal linkage and the other is bonded by ester glycoside linkage. The values of the coupling constants of $^3J_{H\text{-}1'\text{-}H\text{-}2'}$ = 8.1 Hz and $^3J_{H\text{-}1''\text{-}H\text{-}2''}$ = 8.2 as well as the coupling constants for H-1′- H-5′ and H-1′′- H-5′′ between 8 and 9 Hz supposed two glucose moieties. By using COSY, HSQC, HMBC experiments and comparison the sugar carbons with the reported ^{13}C-data of Atta-ur-Rahman,[19] the sugars moieties were identified as β-D-glucose. The linkage between aglycon and sugar was determined by means of HMBC experiment. The HMBC cross peaks between H-1′/C-28 and H-1′′/C-3′ indicates the linkage of glucose to the carboxylic group (C-28) of the aglycone by ester linkage and the second glucose unit at C-3′ of the first glucose by acetal linkage. From the above data, the assumed structure of compound **5** is: 28-*O*-[β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl] oleanolic acid which is a new natural product.

The molecular formula of compound **6** ($C_{42}H_{66}O_{14}$) was deduced from positive-ion mode HRESIMS which showed a pseudo-molecular ion peak at *m/z* 833.4105 $[M+K]^+$ (calc. for $C_{42}H_{66}O_{14}K$, 833.4102). The extensive analysis of 2D-NMR confirmed the presence of quinovic acid as aglycone.[22] 1D and 2D-NMR revealed the presence of two sugar moieties leading to the structure of compound **6** as quinovic acid-3-*O*-(α-L-rhamnopyranosyl-)-28-*O*-β-D-glucopyranosyl ester which was previously isolated from the bark of *Anthocephalus cadamba*.[23]

The DPPH free radical scavenging activity

The antioxidant activities by DPPH radical scavenging assay of the pure compounds at three different concentrations revealed that the flavonoid **2** has the highest antioxidant activity at different three concentrations followed by the new flavonoid **3** (Fig. 1).

Scavenging ability for superoxide anion radical

The free radical scavenging activity on superoxide anion radical generated by an enzymatic method (X-XOD system) showed that the new saponin **5** and compound **1** were the most active compounds (≈80 % scavenging activity at a concentration of 50 µg/ml, while the other isolated compounds revealed moderate antioxidant activity of 53 -73 % (Fig. 2).

Superoxide anion is a precursor to active free radicals and plays an important role in the formation of other ROS (reactive oxygen species), such as hydrogen peroxide, hydroxyl radical and singlet oxygen, which induce oxidative damage in lipids, proteins and DNA.[24] Superoxide radical is normally formed first, and its effects can be magnified, because it produces other kinds of free radicals and oxidizing agents. Superoxide anions have the potential of reacting with biological macromolecules and have been implicated in several pathophysiological processes due to its transformation into more reactive species, such as hydroxyl radical that initiate lipid peroxidation.[25] In addition, it has been reported that antioxidant properties of some flavonoids are effective mainly via scavenging of superoxide anion radical.

It was suggested that flavonoids which possess adjacent di- and/or tri-hydroxyl groups in their structure show strong H_2O_2 generating activity via a superoxide anion radical. Flavonoids which generate H_2O_2 can scavenge free-radicals. Flavonoids generate H_2O_2 by donating a hydrogen atom from their pyrogallol or chatechol structure to oxygen, through a superoxide anion radical. The higher the H_2O_2 generation, the more potent is the radical trapping.[26]

Superoxide anion was of great interest because of the increased dominance in vivo in various disease conditions (inflammation, atherosclerosis and cancer.[26]

Xanthine Oxidase (XOD) enzyme is an important source of superoxide radical. The compound showing both superoxide anion scavenging as well as XOD inhibitory activity may have better therapeutic potential. Flavonoids with both these properties may in common have hydroxyl groups either at C-5, C-3 or C-3′ and C-4′.[26]

CONCLUSION

Two new metabolites, 3-*O*-[α-L-arabinopyranosyl-(1→2)-L-α-arabinopyranosyl)]-3′-methylquercetin (**3**) and 28-*O*-[β-D-glucopyranosylester-(1→3)- β-D-glucopyranosyl] oleanolic acid (**5**) along with four known compounds were isolated for the first time from *Fagonia indica* and *Bassia muricta* from Aseer region, KSA. This finding indicated that the plant in Aseer region is nearly unexplored which may provide a new compounds with a new activities. The antioxidant of two new compounds **3** and **5** could be important for evaluating the mode of action of these compounds and their derivatives in *vivo*.

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REFERENCES

1. El-Ghazali GE, Al-Khalifa KS, Saleem GA, Abdallah EM, Traditional medicinal plants indigenous to Al-Rass province, Saudi Arabia. Journal of Medicinal Plants Research, Vol. 4(24), 2010, 2680-2683.
2. Al-Yahya MA, Al-Meshal IA, Mosa JS, Al-Badr A, Tariq M, Saudi plants, a phytochemical and biological approach. King Saud University Press, KSA, Vol. 64, 1990.
3. El-Khatib, AS, Khaleel, AE, Evaluation of some pharmacological properties of different extracts of *Bauhinia racemosa* Lam. Leaf and *Bassia muricata* L., Whole plant. Bull. Fac. Pharm. 33(2), 1995, 59-65.
4. Beier BA, Nylander JAA , Chase MW, Thulin M, M. Phylogenetic relationships and biogeography of the desert plant genus *Fagonia* (Zygophyllaceae), inferred by parsimony and Bayesian model averaging. Mol. Phylogent. Evol., 33, 2004, 91-108.
5. Akhtar N, Begum S, Ethnopharmacological important plants of Jalala, District Mardan, Pakistan. Pak. J. Plant Sci. 15(2), 2009, 95-100.
6. Shehab NG, Mahdy A, Khan SA, Noureddin SM, Chemical constituents and biological activities of *Fagonia indica* burm F. Res. J. Med. Plant., **5**, 2011, 531-546.
7. Sharrma, S, Gupta V, Sharma G, Phytopharmacology of *Fagonia Indica* (L). A Review. Journal of Natura Conscientia, 1(1), 2010, 143-147 Sharrma S, Gupta V, Sharma G, Phytopharmacology of *Fagonia Indica* (L): A Review. Journal of Natura Conscientia, 1(1), 2010, 143-147.
8. Sharawy SM, Alshammari AM, Checklist of Poisonous Plants and Animals in Aja Mountain, Ha'il Region, Saudi Arabia. Australian Journal of Basic and Applied Sciences, 3(3), 2009, 2217-2225.
9. Ibrahim LF, Kawashty SA, El-Hagrassy, AM, Nassar ML, Mabry Tj, Anew kaempferol triglycoside from *Fagonia taeckholmiana*.: cytotoxic activity of its extracts. Carbohydrate research, 343(1), 2008, 155-158.
10. Perroni, A, Masullo MA, Basarello C, Hamed AI, Belisario MA, Pizza C , Piacente S, Journal of natural products. 70(4), 2007, 584-588.
11. Shaker KH, Bernhardt M, Elgamal MH, Seifert K, Triterpenoid saponins from *Fagonia indica*. Phytochemistry, 51(8), 1999, 1049-1053.
12. Freitas MO, Ponte FAF, Lima MAS, Silveira ER, Production of 7-*O*-Methyl Aromadendrin, a Medicinally Valuable Flavonoid, in *Escherichia coli*. J. Braz. Chem. Soc., 19 (3), 2008, 532-535.
13. Ouf SA, Abd El-Hady FK, ElGamal MH, Shaker KH, Isolation of antifungal compounds from some *Zygophyllum* species and their bioassay against two soil-borne plant pathogens. Folia Microbiol, 39 (3), 1994, 215-221.
14. Ngounou FN, Meli AL, Lontsi D, Sondengam BL, Rahman AU, Choudhary MI, Malik S, Akhtar F, New isoflavonones from Ceiba pentandra. Phytochemistry 54, 2000, 107–110.
15. Arima H, Danno G, Isolation of antimicrobial compounds from Guava (Psidium guajava L.) and their structural elucidation. Biosci. Biotechnol. Biochem. 66, 2002, 1727–1730.
16. Matsushige K, Basnet P, Kadota S and Namba T "Potent free radical scavenging activity of dicaffeoylquinic acid derivatives from propolis". *J.Trad. Med.,* 13, 1996, 217- 228.
17. Al-Taher AY, Anticonvulsant effects of 3, 4-Dimethoxy toluene, the major constituent of *Phoenix dactylifera* L Spathe in mice. Scientific Journal of King Faisal University, 9(2), 2008, 1429 (115-123).
18. Mabry, TJ, Markham KR, Thomas MB, *The Systematic Identification of Flavonoids*, Springer-Verlag, New York, 1970.
19. Rahman AU, Structural Elucidation of Saponins, Georges Massiot and Catherine Lavaud. Studies in Natural Products Chemistry, 15, 1995, 187.
20. Alaniya MD, Kavtaradze NShN Sh, Bassarello C, Skhirtladze AV, Pizza C, Kutateladze I, Flavonoid glycosides from *Astragalus galegiformis* leaves. Chemistry of Natural Compounds, Vol. 42, No. 6, 2006, 681- 685.
21. Pereira DM, Ferreres F, Oliveira J, Valentao P, Andrade PB, Sottomayor M, Targeted metabolite analysis of *Catharanthus roseus* and its biological potential. Food Chem. Toxicol. 47, 2009, 1349-1354.
22. Pollmann K, Elgamal MHA, Shaker KH, Seifert K, Triterpenoid saponuins from The roots of Zygophyllum species. Phytochemistry, 44, 1997, 485489.
23. Sahu Niranjan P, Koike K Jia, Zhonghua B, Sukdeb M, NB., Nikaido T. Triterpene glycosides from the bark of *Anthocephalus cadamba*. Journal of Chemical Research, Synopses, 1, 2000, 22-23.
24. Pietta PG. Flavonoids as antioxidants. *J. Nat. Prod*., 63, 2000, 1035.
25. Wickens AP. Aging and the free radical theory. *Respiratory Physiol.,* 128, 2001, 379-391.
26. Tiwari AK. Imbalance in antioxidant defense and human diseases: Multiple approach of natural antioxidants therapy. *Current Science* 81, 2001, 1179-1187.

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