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BIOLOGICAL ACTIVITY OF THE EGYPTIAN MEDICINAL PLANTS: PART 3 ANTI-OXIDANT, CYTOTOXICITY, ANTI-DIABETIC ACTIVITIES AND CONSTITUENTS OF *BETA VULGARIS SUBSP. PERENNIS*

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Abstract:-Several columns chromatography of the ethyl acetate fraction of *Beta vulgaris* subsp. *perennis* (family Chenopodiacaea) resulted in isolation of quercetin, 4'-hydroxy-5methoxy-6,7-methylenedioxy flavanone, quercetrin and rutin. In addition to the phenolic acids: syrinigic, ferulic and the monoterpene dehydro-vomifoliol. The structure of these compounds was confirmed by spectral methods as well as comparison with reported data. The antioxidant activity indicated moderate activity SC_{50} 8.5µg/ml, compared with that of vitamin C (SC_{50} 1.24µg/ml). The total ethanolic extract of *Beta vulgaris* subsp. *perennis* exhibited a mild cytotoxic activity (IC_{50} 60.26µg/ml compared with Doxorubicin IC_{50} 21.4µg/ml) against Hep-G2 cells. The ethyl acetate extract (400mg/kg) has nearly the same potency as glibenclamide (5mg/kg), but the aqueous extract (400mg/kg) has higher potency than glibenclamide. The aqueous extract significantly decreased rat hind paw edema thickness compared to control group. Ethanolic extract has no anti-inflammatory effect.

Keywords:*Beta vulgaris*, Chenopodiacaea, syringic acid, ferulic acid, flavonoids, sterols, antioxidant, anti-diabetic, cytotoxicity, anti-inflamatory.

INTRODUCTION :

Beta vulgaris is an annual or perennial herb; grow mainly in halophytes and xeric habitats (Evans, 2002). It is widely distributed in the coastal areas of South and East to North Africa, Europe and Mediterranean coastal strip in Egypt (Boulos, 1999). The family Chenopodiaceae comprises wide variety of constituents represented by alkaloids (Muhtadi and Hassan, 1981), volatile oils, lipids, phenolic acids, carbohydrates (Darnley, 1974), flavonoids (Reznik, 1957), saponins (Darnley, 1974; Rastrelli, et al., 1996), proteins and amino acids (Silva and Pereira, 1976), cyanogenic glycosides (Darnley, 1974), pigments, amides and amines (Darnley, 1974). Concerning the current literature, there is no report concerning the chemistry and biology of the Egyptian plant *Beta vulgaris* subsp. *perennis* L. Therefore, it was interesting to carry out a pharmacognostical study on this plant. The present phytochemical study of *Beta vulgaris* subsp. *perennis* L. revealed the presence of thirteen compounds, isolated and subsequently identified by the spectral means (Rasha, 2013). Also, the biology of this edible plant was conducted to uncover the potential of the different extracts towards the current Egyptian diseases.

MATERIALS AND METHODS

General Experimental Procedures

Melting points were determined on SMP3 (Stuart Scientific, U.K.) and are uncorrected; IR spectra were carried out by Jasco FT/IR 6100 type spectrophotometer; ^{1}H and ^{13}C-NMR spectra were carried out by: Varian Mercury-VX-300 NMR spectrometer (300 MHz for ^{1}H- NMR and 75 MHz for ^{13}C-NMR) and Bruker, Avance II NMR spectrometer (600 MHz for 1H-NMR and 150 MHz for ^{13}C-NMR). Mass spectra were carried out by: Jeol JMS-AX 500, 70 ev. and Shimadzu GC/MS-QP 5050A, 70 ev. TLC was performed on precoated TLC, Kieselgel 60 GF254, (60-250 mesh), Fluka using the following solvent systems: Solvent 1(for identification of syringic acid and ferulic acid); CHCL-MeO_3H (9:1, v/v) and Solvent 2 (for

identification of flavonoids); EtOAc-HCOOH-H_2O (4:1:5 v/v). Detection was made by UV lamp and 50% sulphuric acid as visualizing agents.

Plant Material

Beta vulgaris subsp. *perennis* L. was collected in December 2008 in the flowering stage from farms of Metghamr, Dakahlia Governorate, Egypt. The plant was identified by Dr. A. Abd El-Mogly, Prof. of Plant Taxonomy, Flora Department, Horticulture Reasearch Institute, Agriculture Research Centre, Ministry of Agriculture, Cairo, Egypt.

Extraction and Isolation

The air-dried powdered whole plant of *Beta vulgaris* subsp. *perennis* L. (1kg) was extracted by cold maceration (90 % ethanol) (2 L, four times) till exhaustion. The combined alcoholic extract was evaporated under reduced pressure at 50^0C to give 70 g viscous residue. The obtained residue was dissolved in 500 ml of MeOH : H_2O mixture (1: 4) then extracted successively with light petroleum, chloroform then with ethyl acetate to yield 32, 3.8 and 10 g, respectively.

Investigation of Ethyl acetate fraction

About 7 g of the ethyl acetate fraction was applied on top of a silica column (3 x 90 cm, 350 g) packed in methylene chloride. A gradient elution was started with methylene chloride and the polarity was gradually increased by methanol. Fractions (250 ml each) were collected, concentrated and examined (tlc, solvent 1&2). Similar fractions were pooled, concentrated and crystallized to yield compounds 1-7.

Compound 1

Fractions 20-26 on concentration afforded white crystals (300mg, methanol) with mp: 205-207C°, R_f 0.34 (system 1); IR (KBr): υ_{max} cm^{-1} 3477-3368, 2927, 1682, 1600, 1432, 1024 and 1108; MS: m/z (% relative abundance):198 (M^+,10), 180 (M^+,1), 170 (0.4), 167 (57), 153 (45), 150(11), 124 (11), 96 (16), 77 (100), 78 (16), 62 (79), 45 (15) and 44 (34). ^{13}C-NMR: (150 MHz, CD_3OD, δ ppm), δ: 125.35 (C-1), 113 (C-2), 152 (C-3), 148 (C-4), 152 (C-5), 115 (C- 6), 170 (C=O) and 56 (OCH_3).

Compound 2

Fractions 27-30, yellowish white crystals (methanol, 190 mg), mp: 168-171C, R_f= 0.3 (system 1); IR (KBr): υ_{max} cm^{-1} 3434-3369, 2925, 2858, 1730, 1631, 1457, 1378, 1024 and 1125. MS: m/z (% relative abundance): 196 (M^+ + 2, 0.6 %), 178 (0.5), 149 (1.2), 119 (1.94), 91(1.85), 78 (68), 63 (100), 45 (67) and 15 (84).

Compound 3

Fractions 57-70, yellow oily substance (methanol), R_f 0.82 (system 2); UV λ_{max}: band at 230 nm. IR (KBr): υmax cm^{-1} 3416, 2925, 2898, 1660, 1600, 1443, 1324 and 1039. MS: m/z (% relative abundance): 222 (M^+, 2.4%), 194 (M^+-28, 0.7), 181 (2.3), 153 (3.9), 152 (4.45), 137 (5.25), 136 (2.59), 124 (2.56), 108 (4.4), 107 (11.15), 92 (6.4), 70 (20) and 56 (100).

Compound 4

Fractions 71-80, yellow powder (chloroform-methanol) 100 mg, mp: 314-316°C, R_f=0.77 (system 2); MS: m/z (% relative abundance): 303 (M^++1, 8%), 302 (M+, 4%), 275 (0.5), 274 (0.44), 218 (2), 153 (90), 152 (19.85), 137 (0.5), 134 (14) ,133 (45), 124 (0.76), 118 (6), 76 (80) and 49 (100). UV λ_{max} (nm): MeOH: 258, 297(sh.), 380; MeOH + $NaOCH_3$: 276, 327(sh.), 415; MeOH + $AlCl_3$; 267, 300 (sh.), 390; MeOH + $AlCl_3$ + HCl: 269, 356; MeOH + NaOAc: 286, 337; MeOH + NaOAc + H_3BO_3; 274, 304 (sh.), 358.

Compound 5

Fractions 81-100, white crystals (methanol) 700 mg, mp: 180-183C, R_f = 0.61 (system 2). UV λ_{max} (nm): MeOH: 255, 287, 330 (sh); MeOH + $NaOCH_3$: 258, 299, 350 (sh); MeOH + $AlCl_3$: 268, 290; MeOH + $AlCl_3$ + HCl: 263, 291; MeOH + NaOAc: 250, 284; MeOH +NaOAc + H_3BO_3; 254, 284. IR (KBr) υmax cm^{-1}: 3440, 2924, 2856, 1700, 1621, 1450, 1378, 1141 and 936. MS: m/z (% relative abundance): 314 (M^+, 13 %), 194 (3), 195 (2), 167 (6), 153 (1), 152 (10), 124 (6), 123 (7), 120 (5), 94 (6) and 77 (13).

Compound 6

Fractions 111-146, yellow granules (chloroform-methanol) 1.25 g, mp: 180-184°C, R_f = 0.54 (system 2). UV λ_{max} (nm): MeOH: 264, 349; MeOH + $NaOCH_3$: 264, 320 (sh.), 392; MeOH + $AlCl_3$: 268, 430; MeOH + $AlCl_3$ + HCl: 258, 300 (sh.), 389; MeOH + NaOAc: 269, 382; MeOH + NaOAc + H_3BO_3: 260, 388. IR (KBr) υ_{max} cm^{-1}: 3450-3150, 1650, 1601, 1184, 1078, 1043 and 878. MS: m/z (% relative abundance): 449 (M^++1, 0.4 %), 303 (7), 302 (2), 274 (M+-28, 4), 153 (42), 152 (1), 137 (11), 134 (10), 124 (30) and 94 (100).

Compound 7

Fractions 154-175, yellow crystals (chloroform-methanol) 1.4 g, mp: 194-198°C, R_f = 0.34 (system 2). UV λ_{max} (nm): MeOH: 269, 284 (sh.), 343; MeOH + $NaOCH_3$: 279, 332 (sh.), 391; MeOH + $AlCl_3$: 274, 304 (sh.), 363; MeOH + $AlCl_3$ + HCl: 276, 303 (sh.), 348; MeOH + NaOAc: 272, 340; MeOH + NaOAc + H_3BO_3 269, 319 (sh.), 350. IR (KBr) υ_{max} cm^{-1}: 3426-3200, 1660, 1601, 1188, 1088, 1047 and 878. Ms: m/z (% relative abundance): 609 (M-1, 0.5 %), 303 (11), 302 (1), 274 (M+-28, 0.1), 153 (19), 152 (1), 137 (6), 134 (13), 124 (12) and 110 (100).

Biological Evaluation

Antioxidant activity of ethyl acetate extract

The colourimetric method using DPPH method, 20µl of different concentrations (0-25µg/ml final concentration) of tested sample were mixed with 180 µl of ethanolic DPPH gently shaken and incubated for 30 min at 37ºC. The absorbance of the remaining DPPH was measured at 520 nm using a micro titer plate reader. Its antioxidant activity (%) was calculated in comparison to the blank control (ascorbic acid) (Ratty *et al.*, 1988). For each sample, the radical scavenging activity was calculated from the equation:

DPPH Inhibition (%) = [$DPPH_{blank}$ – $DPPH_{test}$] x100 / [$DPPH_{blank}$]

A curve for sample concentration versus DPPH % inhibition was plotted and the half maximal scavenging capacity (SC_{50}) of each tested sample and ascorbic acid were calculated. The results are recorded in Fig. (1).

Cytotoxic activity of total ethanolic extract

Cytotoxic activity of ethanolic extract was carried on human hepatocarcinoma cell line (Hep-G2) using the MTT cell viability assay. The percentage viability was plotted against the extract concentrations and the 50% cell viability (IC_{50}) was calculated from the curve (Hansen *et al*., 1989). The results are presented in Fig. (2).

Anti-inflammatory activity of alcoholic and aqueous extract

Using the hind paw edema method induced by carrageenan (Winter et al., 1962). Diclofenac sodium and dexamethasone were used as reference standards. Thirty five adult male rats were divided into seven groups (5 rats each). All samples were orally administered. The hind paw diameter was measured, using a micrometer, just before the injection of carrgeenan and 1, 2, 3, 4, 5 and 6h. after injection. The hind paw diameter was measured for each rat at each time interval and the mean thickness of edema was calculated. The results are recorded in Table (1) and Fig. (3).

Antidiabetic activity of aqueous and ethyl acetate extract

Thirty adult male rats were divided into five groups (n=6).The first group received gum acacia mucilage (10%) and served as a control. The second is diabetic group and received only the vehicle (10% gum acacia). The third, fourth and fifth diabetic groups received orally glibenclamide (5mg/kg), the total aqueous extract (400mg/kg) and ethyl acetate extract (400 mg/ kg), respectively for 5 days once a day (Sokeng et al., 2005). Diabetes was induced in rats by intraperitoneal injection of streptosotozin (STZ) in a single dose of 75 mg/kg. Rats became diabetic after 5 days of injecting STZ where their blood glucose levels range from 254 to 288 mg/dl. Blood glucose levels were determined using glucomen-glyco® blood glucose meter, 24 sensor strips. The results are presented in Table (2) and Fig. (4).

RESULTS AND DISCUSSION

The ethanolic extract of *Beta vulgaris* subsp. *perennis* L. was fractionated into light petroleum, chloroform and ethyl acetate fractions. Column chromatography of the ethyl acetate fraction afforded seven compounds.

Compound 1

Obtained as crystalline needles, mp: 205-207°C, IR showed a broad absorption bands at 3477-3368 cm^{-1} and 1024 cm^{-1} (OH) of acid, band at 1600cm^{-1} (C=C) and band at 1682cm^{-1} (C=O) (Albuquerque *et al*., 2003). MS with a parent ion at m/z 198 (M^{+}) calculated for $C_9H_{10}O_5$ and peaks at 180 (M-H_2O) indicated presence of hydroxyl group. The base peak at 77 indicated a phenyl moiety (Budzikiewicz *et al*., 1964). ^{13}C-NMR showed two methoxyls, two olefinic methines and five quaternary carbons including a carboxyl and olefinic carbon and three oxygenated olefinic carbons. The above data collectively confirm the presence of Syrinigic acid. To our knowledge, Syrinigic acid was previously isolated from *Beta vulgaris* subsp. cicla (Young *et al*., 2004). However, this is the first report for its isolation from Beta vulgaris subsp. perennis.

Compound 2

Obtained as yellowish white residue, mp: 168-171°C. IR at 3434-3369cm^{-1} and 1071cm^{-1} due to (OH) of acid, band at 1631 cm^{-1} (C=C) and band at 1730cm^{-1} (COOH) (Albuquerque *et al*., 2003). MS at m/z 196 for molecular formula $C_{10}H_{10}O_4$. Peaks at m/z 178 (M^{+}-H_2O) indicate presence of hydroxyl group, 149 (M-C_2OOH), 119 (M- O_2CH-C_2OOH), peak at 78 for phenyl moiety, peak at 91 for tropolium moiety (Budzikiewicz *et al*., 1964). The previous data confirm the presence of ferulic acid. Ferulic acid was previously isolated from Beta vulgaris subsp. conditiva (Kujala *et al*., 2001). However, this is the first report for its isolation from *Beta vulgaris* subsp. *Perennis* L.

Compound 3

Obtained as yellow oily material, UV maximal absorption at 230nm. IR at 3416cm^{-1} and 1039cm^{-1} due to hydroxyl group, band at 1600cm^{-1} for olefinic carbon and peak at 1660cm^{-1} for conjugated carbonyl group. MS with m/z 222 for $C_{13}H_{18}O_3$, peaks at m/z 205 (M^{+}-OH) for hydroxyl group, 194 (M^{+}-CO) for carbonyl group and base peak at 56 indicates the presence of =CH-C=O-CH_3 moiety. Its physical and spectral data were in agreement with those reported data for dehydro-vomifoliol (Kim *et al.*, 2004). Dehydro-vomifoliol was previously isolated from Beta vulgaris subsp. cicla (Kim *et al*., 2004). However, this is the first report for its isolation from *Beta vulgaris* subsp. *perennis.*

Compound 4

Obtained as yellow powder (chloroform-methanol), mp: 314-316°C, intense yellow colour on treatment with alkali and aluminum chloride indicating the flavonoidal skeleton, also -ve Molisch's test suggesting the aglycone nature (Stahl, 1969). UV (Harborne *et al*., 1975) analysis suggests 5, 7, 3', 4'- tetra hydroxy flavonol. Ms m/z 302 (M^{+}) for formula $C_{15}H_{10}O_7$ also peaks at m/z 134 and 137 for ring "B" with two hydroxyl groups. The above mentioned data suggested the presence of quercetin. This was confirmed by direct comparison with authentic samples (MS, Co-tlc, mp. and m.mp.) as well as with published data of quercetin (Nayeem *et al*., 2010). To our knowledge, this compound was previously isolated from Beta vulgaris subsp. vulgaris (Chiji *et al*., 1986). However, this is the first report for isolation of quercetin from Beta vulgaris subsp. perennis.

Compound 5

As white crystals (methanol), mp: 180-183°C., give intense yellow colour with NaOH indicating flavonoid skeleton. IR 3440 cm^{-1} and 1141 cm^{-1} (OH) and band at 1700 cm^{-1} (C=O). Ms showed m/z at 314 (M^{+}) and the appearance of the fragment at m/z 120 (C_8H_8O) corresponding to ring "B" with one hydroxyl group and a fragment at m/z 194 ($C_9H_6O_3$) corresponding to ring "A" with methylenedioxy group at C$_6$ & C$_7$ and methoxy at C$_5$. UV analysis showed an absorbance bands at 255, 287, 330nm (sh) suggested presence of dihydroflavone skeleton ($C_{17}H_{14}O_6$) (Harborne *et al*., 1975). This was confirmed by direct comparison of (UV, IR, MS and mp.) with published data (Gelgert *et al*., 1973 and Elliger *et al*., 1994) which indicates presence of 4' hydroxy-5 methoxy -6,7-methylenedioxy flavanone. This report represents the first time for isolation of 4' hydroxy-5 methoxy -6,7-methylenedioxy flavanone from Beta vulgaris subsp. perennis L.

Compound 6

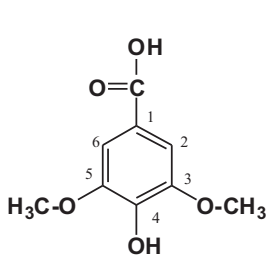
Obtained as yellow granules (chloroform-methanol) having mp: 180-184°C. IR at 3450 – 3150cm^{-1} several (OH) and peak at 1650cm^{-1} (C=O). The UV analysis suggests 5, 7, 3', 4'- tetra hydroxyl substituted flavonol (Harborne *et al*. 1975). MS with m/z 449 (M^{+}+ 1) then m/z 303 indicates flavonoidal glycosides, m/z 302 specific for quercetin aglycone and (M-146) corresponding to rhamnose as sugar part, m/z 153 for ring A with two hydroxy and m/z 134, 137 for ring B with orthodihydroxy group. This was confirmed by direct comparison with reference quercitrin (cotlc, mp) and (UV, IR, MS) with those published data (Bose *et al*., 2013) which confirm presence of quercitrin. Quercitrin was previously isolated from *Beta*

vulgaris subsp. *vulgaris* (Chiji *et al.*, 1986). However, this is the first report for its isolation from Beta vulgaris subsp. perennis L.

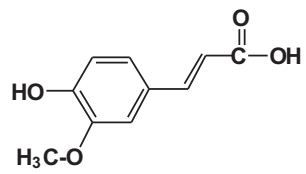
Compound 7

Isolated as yellow crystals (chloroform-methanol), mp: 194-198°C. IR and UV (Harborne *et al*., 1975) suggests the presence of 5, 7, 3', 4'- tetra hydroxyl substituted flavonol. MS m/z 609 (M^{-}-1) followed by significant fragment at m/z 303 indicates flavonoidal glycosides where fragment at m/z 303 for quercetin aglycone and sugar part is rhamnosyl-glucose ($\breve{M}$-308), m/z 153 for ring A with two (OH) and m/z 134, 137 for ring B with two hydroxyl groups. The above mentioned data suggested the presence of quercetin-3-O- rhamnosyl- glucose (Rutin). This was confirmed by direct comparison with reference rutin (cotlc, mp) and (UV, IR and MS) of the compound with published data of rutin (Nayeem *et al*., 2010).

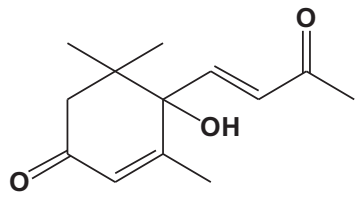
To our knowledge, rutin was previously isolated from Beta vulgaris subsp. cicla (Ninfali et al., 2007). However, this is the first report for its isolation from Beta vulgaris subsp. perennis L.



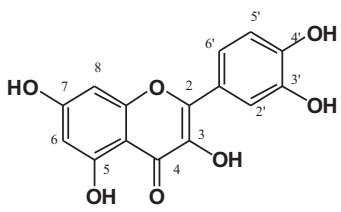
(1) Syringic acid



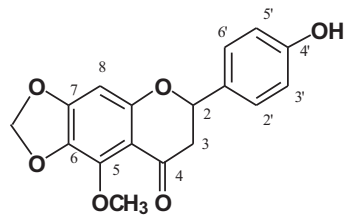
(2) Ferulic acid



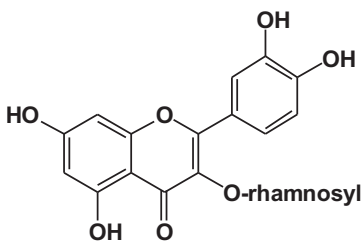
(3) Dehydro-vomifoliol



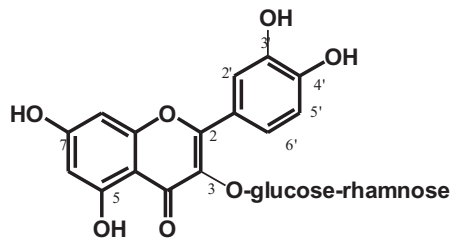
(4) Quercetin



(5) 4' -Hydroxy-5 methoxy-6, 7-methylenedioxy flavanone



(6) Quercetrin



(7) Rutin

Biological evaluation

Antioxidant activity

Considering the antioxidant activity Fig. (1), the ethyl acetate extract possessed a relatively good antioxidant activity with SC_{50} 8.5 µg/ml compared to ascorbic acid solution (SC_{50} 1.24µg/ml). This activity is attributed to the high content of flavonoids in the plant.

Cytotoxic activity

Regarding the anti-tumer activity using cell line (Fig. 2), the total ethanolic extract exhibited a moderate anti-

proliferative activity against hepatoma cells IC_{50} value 60.26µg/ml compared with Doxorubicin (IC_{50} 21.4µg/ml).

Anti-inflammatory activity

As shown in Table (1) and Fig. (3), oral pretreatment with aqueous extract significantly decreased rats hind paw edema thickness compared to control group. Ethanolic extract has no anti-inflammatory effect.

Antidiabetic activity

As shown in Table (2) and Fig. (4), the results indicated that the total aqueous and ethyl acetate extracts and glibenclamide reduced blood glucose levels of the diabetic rats significantly as compared to the diabetic group. There is no significant difference between the effect of glibenclamide and ethyl acetate extract on blood glucose levels showing that the ethyl acetate extract (400mg/kg) has nearly the same potency as glibenclamide (5mg/kg), but the aqueous extract (400mg/kg) has higher potency than glibenclamide.

Table 1: The anti-inflammatory effect of total ethanolic and aqueous extracts (500 and 1000 mg/kg each) of *Beta vulgaris* subsp. *perennis* L.

Group	Percentage increase in edema thickness (%) (Mean±SEM) Time (h.)					
	1	2	3	4	5	6
Control	.5000± .1377	.7520± .2153	1.0100± .2524	1.2820± .2844	1.2820± .2844	1.2380± .3234
Diclofenac sodium (4 mg/kg)	.5520± .1621	.8740± .2196	.9880± .1940	1.2280± .1852	1.0980*± .1667	.7280± .1780
Dexamethasone (0.5 mg/kg)	.6580± .2557	1.084± .2740	1.162± .3061	1.6040± .2515	1.0460*± .4079	.7700 ± .3928
Ethanolic ext. (500 mg/kg)	.7960± .2336	.4980± .1303	1.0560± .1603	1.3640± .1062	1.3480± 7.774E-02	1.3380± .2472
Ethanoilc ext. (1000 mg/kg)	.8820± .1007	.9640± .1280	1.0500± .1011	1.1820 ± .1276	1.3420± 9.041E-02	1.1040± 8.406E-02
Aqueous ext. (500 mg/kg)	.8820± .1007	.9640± .1280	1.0500± .1011	1.1820± .1276	1.2560 * ± 5.483E-02	1.0900± 7.106E-02
Aqueous ext. (1000 mg/kg)	.6820± .1351	1.0400 ± .2660	1.4160 ± .2501	1.8040 ± .2035	2.4120± .2320	2.3500 * ± .3350

* Significantly different from the control group

Table 2: The effect of the aqueous and ethyl acetate extracts (400mg/kg each) of *Beta vulgaris* subsp. *perennis* L. and glibenclamide (5mg/kg) on blood glucose levels of STZ-induced diabetic rats.

	Control	Diabetic	Diabetic + Glibenclamide	Diabetic + B.V. Aqueous ext.	Diabetic + B.V. Ethyl acetate ext.
Blood glucose level (mg/dl)	76.3	248	182.4	192.4	176.8
	79.2	204.7	163.5	182	179
	82.5	253.8	167.8	186.5	190.3
	79.5	219.5	146.2	190.5	168.4
	83.7	198.4	183	184.9	160.5
	76.2	284.9	165.7	193	176.3
Mean	**79.6**	**234.9**	**168.1**	**188.2**	**175.2**
SD	3.1	33.2	13.7	4.4	10.1
SEM	1.3	13.6	5.6	1.8	4.1

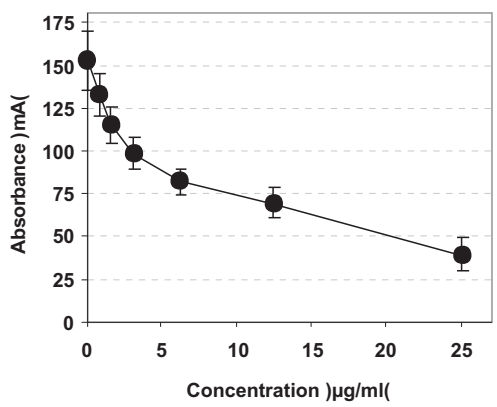


Fig. 1: Antioxidant activity of the ethyl acetate extract.

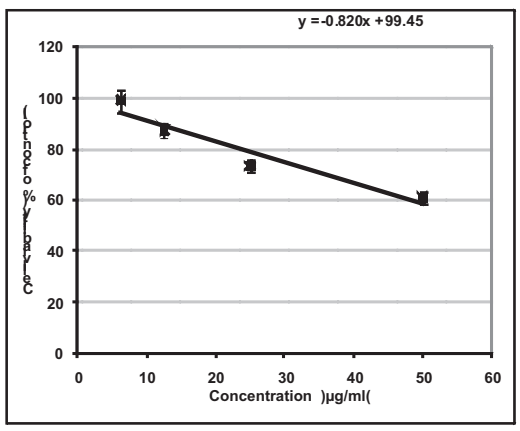


Fig. 2: Anti-tumor activity of the total alcoholic extract of *Beta vulgaris* subsp. *perennis* L. against Hep-G2 cells, IC_{50} value 60.26µg/ml.

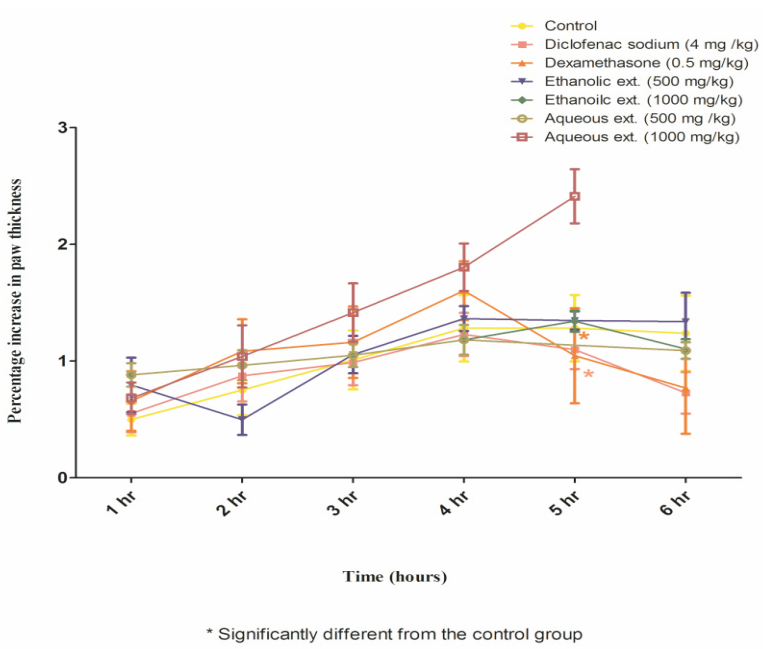


Fig. 3: The anti-inflammatory effect of total ethanolic and aqueous extracts (500 and 1000mg/kg each) of *Beta vulgaris* subsp. *perennis* L.

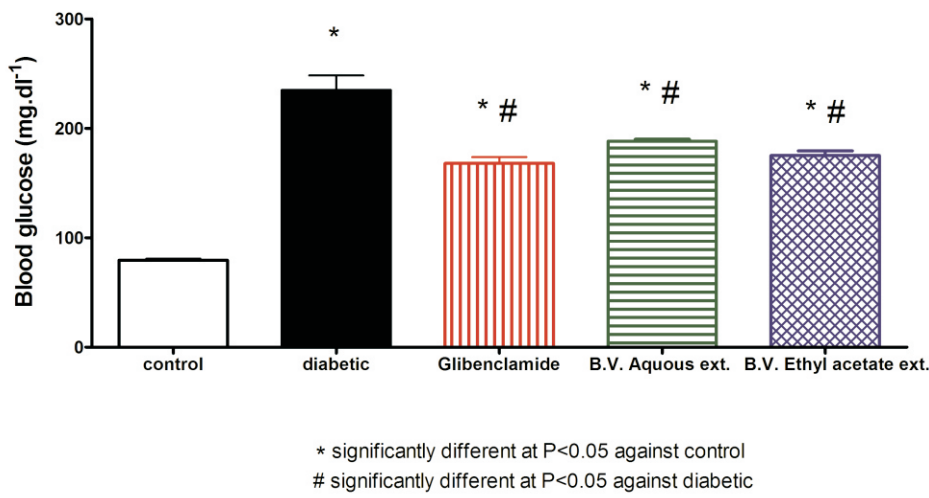


Fig. 4: Effect of the aqueous and ethyl acetate extracts (400mg/kg each) of *Beta vulgaris* subsp. *perennis* L. and glibenclamide (5mg/kg) on blood glucose levels of STZ-induced diabetic rats.

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