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Teucrium polium L. essential oil: phytochemiacl component and antioxidant properties

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Abstract

The essential oil (EO) obtained by hydrodistillation from *Teucrium polium* L. was analyzed by gas chromatography and gas chromatography mass spectrometry (GC/MS). Antioxidant activity was evaluated through DPPH assay and Beta-carotene/linoleic acid assay. Twenty one components were identified representing (90.4%) of the EO. Main constituents of the essential oil were found as Spathulenol (15.06%), Beta-Pinene (11.02), Beta-Myrcene (10.05), Germacrene B (10.11%), Germacrene D (8.15%), Bicyclogermacrene (8.25%) and Linalool (4.02). This essential oil was able to reduce the stable free radical DPPH with an IC_{50} of 9200 µg/ml. In Beta-carotene/linoleic acid assay, these samples were not effectively able to complete inhibit the linoleic acid oxidation, exhibiting only 61% inhibitions at 2 mg/ml. These results demonstrate that the *T. polium* EO possesses antioxidant activity *in vitro.* Further investigations are needed to verify whether this antioxidant effect occurs *in vivo*.

Introduction

Teucrium is a genus of perennial plants which belongs to the family *Lamiaceae* is represented by more than 340 species widespread all around the world and comprises about 12 species in Iran. They are most common in Mediterranean climates and Middle East. An unusual feature of this genus compared with other member of *Lamiaceae* is that flower completely lack of the upper lip of corolla. Its flower is small and range from pink to white and its leaves are used in cooking and for medicinal purposes (Rechinger, 1982; Zargari, 1992; Mozaffarian, 1996). *T. polium* L. is a plant that has been used for over 2000 years in traditional medicine due to its diuretic, diaphoretic, tonic, antipyretic, antispasmodic and cholagogic properties (Galati *et al*., 2000; Said *et al.*, 2002). In addition, the plant possesses hypoglycemic, insulinotropic, anti-inflammatory activities, hypolipidemic, antinociceptive and antioxidant properties (Couladis *et al.*, 2003; Esmaeili *et al*., 2004). The therapeutic benefit of medicinal plants is often attributed to their antioxidant properties (Rice, 2004; Dixon *et al.*, 2005). In this regard, a methanolic extract of *T. polium* protected red blood cells (RBCs) against lipid peroxidation induced by 10 mmol hydrogen peroxide (Soubh *et al*., 2004). In another study, Kadifkova-Panovska *et al*. (2005) demonstrated that extracts of *T. polium* prepared using different organic solvents (diethyl ether, ethyl acetate and n-butanol) were effective inhibitors of b-carotene oxidation. Ljubuncic *et al.* (2005) showed that the aqueous extracts prepared from the foliage of *T. polium* suppressed iron (Fe2)-induced lipid peroxidation in rat liver homogenates to the same extent as Trolox, the water soluble analog of vitamin E. Additionally, this extract was not cytotoxic because it did not adversely affect cell membrane integrity or suppress mitochondrial respiration of cultured Hep G2 and PC12 cells following 24 h exposure (Ljubuncic *et al*., 2005). In another study, Azaizeh *et al.* (2005) reported that adding fertilizer caused a significant concentration-dependent increase in antioxidant activity of the cultivated *T. polium* compared with the wild-type (Azaizeh *et al.*, 2005). In the present work we have studied the chemical composition and antioxidant properties of the *T. polium* EO that grow in Kerman province in Iran.

Material and Methods

Plant material

The *T. polium* L. was collected during flowering stage from West of Iran (Kerman province) and identified by the Herbarium of faculty of Pharmacy, University of Tabriz, Tabriz, Iran, Then shade dried and grinded into powder. The prepared powder was kept in tight containers protected completely from

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light.

EO preparation

Dry aerial parts (100 g) of *T. polium* L. were subjected to the hydrodistillation of 2.5 h, using a cleavenger-type apparatus, according to the method recommended by the European Pharmacopia to produce oils (Maissoneuve, 1983). The obtained essential oil was dried over anhydrous sodium sulphate and stored at +4°C until tested and analyzed.

Gas chromatography–mass spectrometry (GC/MS)

The EO was analyzed by gas chromatography. The chromatograph (Agilent 6890 UK) was equipped with an HP-5MS capillary column (30 × 0.25 mm ID × 0.25 mm film thickness) and the data were taken under the following conditions: initial temperature 50°C, temperature ramp 5°C/min, 240°C/min to 300°C (holding for 3 min), and injector temperature at 290°C. The carrier gas was helium and the split ratio was 0.8 mL^{-1}/min. For confirmation of analysis results, essential oil was also analyzed by GC/MS (Agilent 6890 gas chromatograph equipped with an Agilent 5973 mass-selective detector; Agilent UK) and the same capillary column and analytical conditions as above. The MS was run in electron-ionization mode with ionization energy of 70 eV (Mahmoudi *et al.*, 2012).

Antioxidant assay

DPPH assay

The hydrogen atom or electron donation ability of the corresponding extracts and some pure compounds was measured from the bleaching of purple colored methanol solution of DPPH. This spectrophotometer assay (Pharmacia, Uppsala, Sweden) uses stable radical DPPH (Sigma, Aldrich) as a reagent. Aliquots (50 µl) of various concentrations of the essential oil were added to 5 ml of a 0.004% methanol solution of DPPH. After a 30 min incubation period at room temperature the absorbance was read against a blank at 517 nm. Inhibition free radical DPPH in percent (I %) was calculated in following way:

$$I\,\% = (A_{blank} - A_{sample}/A_{blank}) \times 100$$

Where A_{blank} is the absorbance of the control reaction (containing all reagents except the test compound), and A_{sample} is the absorbance of the test compound. Essential oil concentration providing 50% inhibition (IC_{50}) was calculated from the graph plotted inhibition percentage against essential oil concentration (Cuendet *et al.*, 1997; Burits and Bucar, 2000). Synthetic antioxidant reagent butylated hyroxytoluene (BHT) was used as the positive control and all tests were carried out in triplicate.

Beta-Carotene–linoleic acid assay

In this assay antioxidant capacity is determined by measuring the inhibition of the volatile organic compounds and the conjugated diene hydroperoxides arising from linoleic acid oxidation (Dapkevicius *et al*., 1998). A stock solution of B-carotene/linoleic acid (Sigma–Aldrich) was prepared as follows. First, 0.5 mg of B-carotene was dissolved in 1 ml of chloroform (HPLC grade), then 25 µl of linoleic acid and 200 mg of Tween 40 (Merck) were added. The chloroform was subsequently evaporated using a vacuum evaporator (Buchi, Flawil, Switzerland). Then 100 ml of distilled water saturated with oxygen was added with vigorous shaking. Aliquots (2.5 ml) of this reaction mixture were transferred to test tubes, and a 350 µl portion of the essential oil was added before incubating for 48 h at room temperature. The same procedure was repeated with BHT at the same concentration and a blank containing only 350 µl of ethanol. After the incubation period, the absorbencies of the mixtures were measured at 490 nm. Antioxidant capacities of the samples were compared with those of BHT and the blank.

Assay for total phenolics

Total phenolic constituent in *T. polium* EO was performed employing the literature methods (Slinkard and Singleton, 1977; Chandler and Dodds, 1983). Involving Folin-Ciocalteu reagent and gallic acid (both Sigma–Aldrich) as standard. Briefly, an aliquot (0.1) ml of extract solution containing 1 mg of extract was transferred to a volumetric flask, 46 ml distilled water and 1 ml Folin-Ciocalteu reagent was added and the flask was shaken thoroughly. After 3 min, 3 ml of solution 2% Na_2CO_3 was added and the mixture was allowed to stand for 2 h with intermittent shaking. Absorbance was measured at 760 nm. The same procedure was repeated for all standard gallic acid solutions (0–1000 mg in 0.1 ml) and a standard curve was obtained according to the equation

$$\text{Absorbance} = 0.0012 \times \text{Gallic acid } (\mu g) + 0.0033$$

Results

The EO was extracted by the hydrodistillation of dried parts of *T. polium* were analyzed by GC and GC–MS. the oil yields were calculated on a dry weight basis as 0.50% (w/w). GC and GC–MS analysis enabled the identification of a total of 21

Table 1. Constituents of *T. polium* EO

Compound No.	Compound Name	RT (minute)	Percentage (%)
1	Verbenene	6.97	1.25
2	Beta -Pinene	7.59	11.02
3	Beta .-Myrcene	7.93	10.05
4	Benzene, 1-methyl	8.88	1.66
5	Cyclohexene, 1-methyl-4-(1-methylethenyl)	9.01	2.00
6	2,3,3-Trimethyl-3-cyclopentene acetaldehyde	11.80	1.71
7	Bicyclogermacrene	12.49	8.25
8	Benzene	13.87	1.46
9	Bicyclo[3.1.1]hept-3-en-2-one, 4,6,6-trimethyl	14.27	1.31
10	1,6,10-Dodecatriene, 7,11-dimethyl-3-methylene	20.71	1.23
11	Germacrene D	21.42	8.15
12	Germacrene B	23.28	10.11
13	sesquisabinene hydrate	23.46	5.26
14	Spathulenol	24.15	15.06
15	o-Menth-8-ene	24.63	1.23
16	3-Cyclohexene-1-methanol, .alpha .,4-dimethyl	24.75	1.23
17	1H-3a,7-Methanoazulene	25.12	1.84
18	Naphthalene	25.81	1.21
19	Linalool	25.97	4.02
20	Cyclolongifolene oxide, dehydro	26.22	1.05
21	Benzenemethanol, 4-(1-methylethyl)	26.98	1.30
	Total		90.40

RT:Retention Time (minute)

constituents, representing 90.40% of the oil. The relative concentrations of the identified volatile components (%) and retention time are presented in Table 1. The combinations of Spathulenol (15.06%), Beta-Pinene (11.02), Beta-Myrcene (10.05), GermacreneB (10.11%), GermacreneD (8.15%), bicyclogermacrene (8.25%) and Linalool (4.02) with 66.66 percent constituent the highest percentage of EO.

In our study, we have investigated the free radical scavenging activity and lipid oxidation inhibition of *T. polium* EO *in vitro*. Free radical scavenging activities of the EO were measured in DPPH assay and the reaction followed a concentration dependent pattern. Free radical scavenging increases with increasing EO concentration and essential concentrations providing 50% inhibition (IC_{50}) was 9200 ± 1.89 µg/ml. The free radical scavenging activity of essential oil in compared with BHT was very low (IC_{50} = 9200 ± 1.89 µg/ml and 19.8 ± 0.32 respectively). Total phenolic constituent of EO was determined as gallic acid equivalent. 62 ± 0.42 mg gallic acid equivalent total phenolic in 1 mg essential oil, was determined that might be responsible for the radical scavenging activity of EO. In the case of inhibition of linoleic acid assay, essential oil was not able to effectively complete inhibit the linoleic acid oxidation, and only 71.12 ± 0.72% inhibitions were achieved at 2 mg/ml concentrations, which were far below the positive control BHT at the same concentration (Fig.1). Essential oil possessed relatively appropriate inhibition effect on linoleic acid oxidation and this subject related to its total phenolic constituent. Activity might be improved at higher concentrations but this was not considered here.

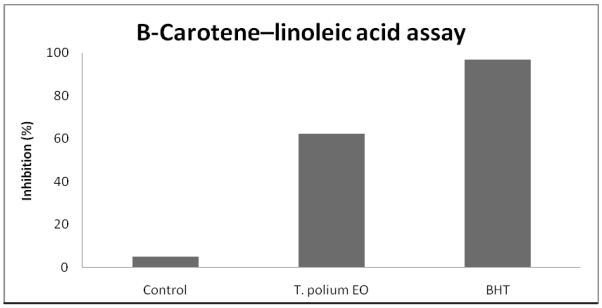

Figure1. Antioxidant activity of *T. polium* EO defined as inhibition percentage in Beta-carotene-linoleic acid assay

Discussion

The EO of *Teucrium* species has been studied in Iran and in the world. A scientific study of the oil of *T. stocksianum* subsp. Stocksianum in April 2003 found thirty-eight compounds with 97.7% of the oil were identified. The oil yield obtaind 0.7% on a dry weight basis. The major component of the oil as Camphene (20.6%), α pinene (19.7%), Myrcene (10.2%) and Carvacrol (9.90%) (Jaimand *et al*., 2006). The studies made have reported the volatile constituents of *T. flavum* leaves from Iran at the full flowering stage in May 2001, eighteen compounds were identified constituenting about 99% with 0.2% yield of the oil that major components were beta-caryophyllene (30.7%), Germacrene (21.3%) and alpha-humulene (8.4%) (Baher and Mirza, 2003).

A study on the oil obtained from *T. polium* grow in Iran (Eikani *et al.*, 1999) revealed the presence of sesquiterpenens as major components. Germacrene D (13.2%), beta-caryophyllene (18%), Spathulenol (10.4%) and bicyclogermacrene (9.0%%) being the major components identified. The main and important ingredient studied in different regions of the world in Italia on *T. fruticans* include p-Cymene (7.0%) (Cozzani et al., 2005). Chemical composition of the EO of the aerial parts of *T. polium* L. grow in Jordan was determined by GC/MS. The oil obtained by hydrodistillation was found to contain 39 components, of which 37 were identified. The major components determined were 8-cedren-13-ol(24.8%),beta-caryophyllene (8.7%), Germacrene D (6.8%) and Subinene (5.2%) (Aburja *et al.*, 2006). Thirty-seven components were detected in the EO obtained from the aerial part of Turkish *T. polium* (Cakir *et al*., 1998). The major components identified were beta-pinene (18%), beta-caryophyllene (17.8%), alpha-pinene (12%), Caryophyllene oxide (10%), Myrcene (6.8%), Germacrene D (5.3%), Limonene (3.5%) and Spathulenol (3.3%). It is the first time that a T.polium has been found to contain Spathulenol (14.65%) at such high percentage, Spathulenol was reported as an important constituents of *T. polium* (10.4%)

(Eikani *et al.*, 1999) and in the EO obtained from the aerial parts of Turkish *T. polium* (3.3%) (Cakir *et al*., 1998). Based on the results of GC/MS analysis of *T. polium* EO in our study, beta-Pinene, beta-Myrcene, Germacrene B, Germacrene D, bicyclogermacrene and Linalool were the major components and also these component has been reported as the major compounds of EO obtained from Teucrium species in other scientific researches (Cakir *et al.*, 1998; Eikani *et al.*, 1999; Baher and Mirza, 2003, Amiri, 2003; Aburjai *et al*., 2006).

The EOs of *Teucrium* species were characterized by a higher content of sesquiterpenenoids, in accordance with the results reported by previous studies (Cavalerio *et al*., 2002; Kucuk *et al.*, 2006; Hachicha *et al*., 2007; Saroglou *et al.*, 2007). So the Predominance of the sesquiterpenenoids (5.26%) in this *Teucrium* species is not surprising. It is possible to say that the difference in the quality or quantity of the composition of volatile oils may be due to genetic, differing chemotype, dring conditions, mode of distillation and or extraction and geographic or climatic factors.

The results of the Antioxidant activity *T. polium* EO are in agreement with those reported by other groups of investigators using a variety of other assays to assess the antioxidant activity of extracts of *T. polium* (Couladis *et al*., 2003, Suboh *et al*., 2004; Kadifkova-Panovska *et al*., 2005). In their study, Couladis *et al.* (2003) screened Greek aromatic plants from the *Lamiaceae* family for the antioxidant activity. Of the 21 plants tested, they found ethanol extracts prepared from *T. polium*, as well as 9 other plants, exhibited the same antioxidant activity as alpha-tocopherol in their ability to inhibit bleomycin-Fe(II) complex-induced arachidonic acid superoxidation to MDA. Regrettably, these authors did not include any data about the plant extract concentration(s) in their report. In another study, Suboh *et al.* (2004) investigated the antioxidant activity effects of various concentrations of methanol extracts of *T. polium* and the six other medicinal plants as high as 1 mg ml^{-1}, the identical concentration to the highest one used in our study. They reported that the extract significantly reduced 10 mmol hydrogen peroxide-induced lipid peroxidation in human erythrocytes. In contrast, the extract failed to protect erythrocytes against protein oxidation and loss of cell deformability of the oxidatively stressed erythrocytes. Kadifkova-Panovska *et al*. (2005) also investigated the antioxidant activity of extracts of *T. chamaedrys*, *T. montanum* and *T. polium* prepared using different organic solvents, namely diethyl ether, ethyl acetate and n-butanol. Using such extracts, they assessed their antioxidant abilities in a series of assays that included inhibition of 1,1-diphenyl-2-picrylhydrazyl radical (DPPH assay), inhibition of hydroxyl radical (d-ribose assay) and inhibition of B-carotene oxidation. They found that a 0.4 mg ml^{-1} extract of *T. polium* was very effective in inhibiting b-carotene oxidation. Based on our results and these reports, there can be little doubt that extracts of *T. polium*, irrespective of the mode of preparation, can inhibit oxidative processes leading us to conclude that the extract has substantial antioxidant activity *in vitro*. We did not attempt to identify the chemical constituents that could account for the antioxidant action of the extract. However, other investigators have reported that the aerial parts of *T. polium* are rich in flavonoids (Rizk *et al*., 1986; Harborne *et al*., 1986). Therefore, it would be reasonable to assume that the antioxidant properties of an aqueous extract of *T. polium* can be attributed to presence of these bioactive components.

Conclusion

Based on the results of this study, we conclude that EO of *T. polium* possesses remarkable antioxidant activities. Accordingly, we propose that the therapeutic benefit of the essential oil as used in traditional medicine might be due to its antioxidant activity. Further investigations are needed to verify whether this antioxidant effect occurs *in vivo*.

References

Aburjai, T., Hudaib, M. and Cavrini, V. 2006. Composition of the essential oil from Jordanian germander (*Teucrium polium* L.). Journal Essential Oil Research 18: 97-99.

Amiri, H. 2008. Essential oil composition of *T. orientale* L. subsp. Taylori (Boiss). Journal of Medicinal Plants 7: 100-104.

Azaizeh, H., Ljubuncic, P., Portnaya, I., Said, O., Cogan, U. And Bomzon, A. 2005. Fertilization-induced changes in growth parameters and antioxidant activity of medicinal plants used in traditional Arab medicine. Evidence Based Complement Alternation Medicine 2: 549–556.

Bahar, Z.F. and Mirza, M. 2003. Volatile constituents of *Teucrium flavum* L. from Iran. Journal Essential Oil Research 15: 106-107.

Burits, M. and Bucar, F. 2000. Antioxidant activity of Nigella sativa essential oil. Phytotheraphy Research 14: 323–328.

Cakir, A., Duru, M. E., Harmandar, M., Ciriminna, R., Passannanti, S. and Kazlm, K. 1998. Volatile constituents of *Teucrium polium* L. from Turkey. Journal Essential Oil Research10: 113-115.

Cavalerio, C., Salguerio, L. R., Antunes, T., Sevinate-

Pinto, I. and Barroso, J. G. 2002. Composition of the essential oil and micromorphology of trichomes of *Teucrium salviastrum*, an endemic species from Portugal. Flavour Fragrance Journal 17: 287-291.

Chandler, S. F. and Dodds, J. H. 1983. The effect of phosphate, nitrogen and sucrose on the production of phenolics and solasidine in callus cultures of *Solanum lacinitum*. Plant Cell Reports 2: 105.

Couladis, M., Tzakou, O., Verykokidou, E. and Harvala, C. 2003. Screening of some Greek aromatic plants for antioxidant activity. Phytotherapy Research 17: 194–195.

Cozzani, S., Muselli, A., Desjobert, J. M., Bernardini, A. F., Tomi, F. and Casanova, J. 2005. Chemical composition of essential oil of *Teucrium polium* subsp. *Capitatum* L. from Corsica. Flavor Fragrance Journal 20: 436-441.

Cuendet, M., Hostettmann, K. and Potterat, O. 1997. Iridoid glucosides with free radical scavenging properties from Fagraea blumei. Helvetica Chemical Acta 80: 1144–1152.

Dapkevicius, A., Venskutonis, R., Van Beek, T. A. and Linssen, P. H. 1998. Antioxidan activity of extracts obtained by different isolation procedures from some aromatic herbs grown in Lithuania. Journal of the Science Food and Agriculture 77: 140–146.

Dixon, R. A., Xie, D.Y. and Sharma, S.B. 2005. Proanthocyanidins a final frontier in flavonoid research? New Philology 165:9–28.

Eikani, M. H., Goodarznia, L. and Mirza, M. 1999. Comparison between the essential oil of supercritical carbon dioxide extract of *Teucrium polium* L. Journal Essential Oil Research 11: 470-472.

Esmaeili, M. A. and Yazdanparast, R. 2004. Hypoglycaemic effect of *Teucrium polium*: studies with rat pancreatic islets. Journal Ethnopharmacology 95: 27–30.

Galati, E. M., Mondello, M. R. D., Aquino, A., Miceli N., Sanogo R. and Tzakou O, 2000. Effects of *Teucrium divaricatum* Heldr. ssp. divaricatum decoction on experimental ulcer in rats. Journal Ethnopharmacology 72: 337–342.

Hachicha, S. F., Skanji, T., Barrek, S., Ghrabi, Z. G. and Zarrok, H., 2007. Chemical Composition *Teucrium alopecurus* essential oil from Tunusia. Flavour Fragrance Journal 22: 101-104.

Harborne, J. B., Tomas, B., Williams, C. A. and Gil, M. I. 1986. A chemotaxonomic study of flavonoids from European Teucrium species. Phytochemistry 25: 2811–2816.

Kadifkova-Panovska, T., Kulevanova, S. And Stefova, M. 2005. *In vitro* antioxidant activity of some *Teucrium* species (*Lamiaceae*). Acta Pharmacology 55: 207–214.

Kucuk, M., Gulec, C., Yasar, A., Ucuncu, O., Yayli, N., Coskuncelebi, K., Terzioglu, S. and Yayli, N. 2006. Chemical composition and antimicrobial activities of the essential oils of *Teucrium chamaedrys* subsp. chamaedrys, *T. orientale* var. puberulens and *Teucrium chamaedrys* subsp. lydium. Pharmaceutical Biology 44: 592-598.

Ljubuncic, P., Azaizeh, H., Portnaya, I., Cogan, U., Said, O. and Saleh, K.A. 2005. Antioxidant activity and cytotoxicity of eight plants used in traditional Arab medicine in Israel. Journal Ethnopharmacology 99: 43–47.

Mahmoudi, R., Tajik, H., Ehsani, A. Farshid A. A. and Zare P. 2012. Effects of *Mentha longifolia* L. essential oil on viability and cellular ultrastructure of *Lactobacillus casei* during ripening of probiotic Feta cheese. International Journal of Dairy Technology, In press.

Maisonneuve, S. A. and Ruffine, R. 1983. European Pharmacopoeia, Part 1, p. 4-8.

Mimica-Dukic, N., Popovic, M., Jakovljevic, V., Szabo, A. and Gasic, O. 1999. Pharmacological studies of *Mentha longifolia* phenolic extracts II. Hepatoprotective activity. Pharmaceutical Biology 37: 221–224.

Mozaffarian, V. 1996. A Dictionary of Iranian Plant Names, Farhang Moaser, p. 542-544. Tehran: Iran,

Rechinger, K .H. 1982. Labiatae. P.150. Akademische Druck-u Verlagsanstalt, Graz.

Rice-Evans, C. 2004. Flavonoids and isoflavones: absorption, metabolism and bioactivity. Free Radical Biology Medicine 36: 827–828.

Rizk, A. M., Hammouda, F. M., Rimpler, H. and Kamel, A. 1986. Iridoids and flavonoids of *Teucrium polium* herb. Planta Medicine 2:87–88.

Said, O., Khalil, K., Fulder, S. and Azaizeh, H. 2002. Ethnopharmacological survey of medicinal herbs in Israel, the Golan Heights and the West Bank region. Journal Ethnopharmacological 83:251–265.

Saroglou, V., Arfan, M., Shabir, A., Hadjipavlou-Litina, D. and Skaltsa, H. 2007. Composition and antioxidant activity of the essential oil of *Teucrium royleanum* Wall. ex Benth growing in Pakistan. Flavour Fragrance Journal 22: 154-157.

Slinkard, K. and Singleton, V. L. 1977. Total phenol analyses: automation and comparison with manual methods. American Journal of Ecology and Viticulture 28: 49–55.

Suboh, S. M., Bilto, Y. Y. and Aburjai, T. A. 2004. Protective effects of selected medicinal plants against protein degradation, lipid peroxidation and deformability loss of oxidatively stressed human erythrocytes. Phytotherapy Research 18: 280–284.

Suleiman, M. S., Abdelghanl, A. S., EI-Khalil, S. and Amin, R. 1998. Effect of *Teucrium polium* boiled extract on Intestinal motility and blood pressure. Journal Ethnopharmacological 72: 111-116.

Zargari, A. 1992. Medicinal plants. P.130-131, University of Tehran Press: Tehran, Iran.