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Full Length Research Paper

Essential oil composition of *Phryna ortegioides* (Fisch. & Mey.) Pax & Hoffm. from Turkey

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The essential oil components of aerial parts of *Phryna ortegioides* (Fisch. & Mey.) Pax & Hoffm. was investigated by GC and GC-MS. The yield of oil was ca. 0.2 mL/100 g. Twenty components were identified representing 90.9% oil. Germacrene D (26.6%), borneol (19.1%), bicyclogermacrene (9.2%) were identified as major components of *Phryna ortegioides.* Caryophylaceae genera like *Gypsophila* L., *Minuartia* L. and *Cerastium* L. produced many similar major compounds in their essential oils that could be justified by the similar ecological conditions of their habitat, but also differences were found that could confirm their taxonomic separation. Also, in this study chemical distribution of the essential oil compounds in the genus pattern were discussed in means of chemotaxonomy and natural products.

Key words: *Phryna*, essential oil, germacrene D, borneol.

INTRODUCTİON

Plants have always been part of the daily life of man, since it is used for food, medicine and sometimes in religious rites. Turkey is situated at the junction of three important phytogeographic regions, namely Mediterranean, Irano-Turanian and Euro-Siberian with three different climates. Therefore its flora, which is highly used with medicinal purposes, is rich and diverse with over 10,000 vascular plant taxa and 32% of endemism (Baser, 2002).

Throughout history, humans have derived many uses and benefits from the plants found in their own region. Initially, wild plants were collected from their natural habitat, followed by the cultivation of those that were used most commonly (Akan et al., 2008). Today the value of the plants is acknowledged and a number of studies are conducted on the plants. There is a growing body of research particularly concentrating on taxonomy, ethnobotanics, plant morphology, anatomy and plant chemistry (Kıvcak et al., 2009; Cabi et al., 2010; Duran et al., 2010; Koyuncu et al., 2010; Bani et al., 2011; Kilic et al, 2011; Jabeen and Aslam, 2011; Korkmaz and Ozcelik, 2011; Kilic and Bagci, 2011).

The genus *Phryna,* which belongs to Caryophyllaceae family, is represented in Turkey by only *Phryna ortegioides. P. ortegioides* is a monotypic and endemic taxa for Turkey. Synonyms of *P. ortegioides* are, *Tunica ortegioides* Fisch & Mey. ; *Saponaria ortegioides* (Fisch & Mey.) Boiss and Bal. ; *Gypsophila ortegioides* (Fisch & Mey.) Boiss. The genus *Phryna*, perennial herb with woody caudex and several glandular-puberulent, dichotomously forked stems, linear leaves, 5-costate long campanular calyx provided with 2-3 pairs of bracteoles at the base, petals linear-cuneate, white with pink veins. The genus is closely related to *Gypsophila* L., from which it differs by the involucrate calyx (Davis, 1975). For centuries, indigenous plants have been used in herbal medicine for curing various diseases and there is a popularity and scientific interest to for screen essential oils and extracts of plants used medicinally all over the world (Cowan, 1999). Many infectious diseases are

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known to be treated with herbal remedies throughout the history of mankind. Even today, plant materials continue to play a major role in primary health care as therapeutic remedies in many developing countries (Zakaria, 1991).

Essential oils are mostly natural mixtures of terpenes/terpenoids, most of which are obtained from aromatic and pharmaceutical plants. The chemical composition of essential oil differs in each species or subspecies and is characteristic for the species in question. Identification of individual components of complex mixtures such as terpenes/terpenoids in essential oils requires the use of several techniques. One of the most popular methods of studying essential oil composition is gas chromatography–mass spectrometry (GC–MS), which allows the identification of the specific natural compounds found in an essential oil by comparing their relative retention times/indices and their mass spectra (Adams, 1995; Flamini et al., 2002; Skaltsa et al., 2003, 2000; Jovanovic et al., 2004; Warthen et al., 1997; Javidnia et al., 2005; Ertugrul et al., 2003).

The species lacks detailed phytochemical investigation. Literature data on the chemical composition of the essential oil of *Phryna* taxa has not been reported. Therefore, the aim of this study is to provide chemical data of *P. ortegioides* prelimenary study, that might be helpful in potential usefulness and chemotaxonomical importance of this species.

MATERIALS AND METHODS

Plant material

P. ortegioides was collected from Aşağıköy village road side (altitude of 1400-1450 m), Bingöl/Turkey, in October 2011. A voucher specimen number (BIN-7415) kept at the Bingol University Herbarium (BIN).

Isolation of volatile oil

100 g dry weight aerial parts (flower-leaf-stem) of the plant materials were subjected to hydrodistillation using a Clevenger-type apparatus for 3 h. The essential oil was analyzed using HP 6890 GC equipped with FID detector and an HP- 5 MS column (30 m × 0.25 mm i.d., film tickness (0.25 µm) capillary column. The analysis conditions should be moved to here from the following paragraph. The percentage composition of the essential oils was computed from GC-FID peak areas without correction factors.

Gas chromatography/mass spectrometry (GC-MS)

The oils were analyzed by GC-MS, using a Hewlett Packard system. HP-Agilent 5973 N GC-MS system with 6890 GC in Plant Products and Biotechnology Research Laboratory (BUBAL) in Firat University. HP-5 MS column (30 m × 0.25 mm i.d., film tickness (0.25 µm) was used with helium as the carrier gas. Injector temperature was 250 ºC, split flow was 1 mL/min. The GC oven temperature was kept at 70 ºC for 2 min. and programmed to 150 ºC at a rate of 10 ºC/min and then kept constant at 150 ºC for 15 min to 240 ºC at a rate of 5 ºC / min. Alkanes were used as reference points in the calculation of relative retention indices (RRI). MS were taken at 70 eV and a mass range of 35-425. Component identification was carried out using spectrometric electronic libraries (WILEY, NIST).

RESULTS AND DISCUSSION

The essential oil components of aerial parts of *P. ortegioides* was investigated by GC and GC-MS. The yield of oil is ca. 0.2 mL/100 g. Twenty components were identified representing 90.9% oil. Germacrene D (20.6%), borneol (19.1%), bicyclogermacrene (9.2%) and *p*-cymene (7.3%) were identified as major components of *P. ortegioides*. The identified constituents of the essential oils are listed in Table 1 and the major components of some Caryophyllaceae taxa are listed in Table 2.

P. ortegioides is a flowering plant native to eastern part of Turkey. It is also cultivated and distributed for centuries in temperate regions of Western Asia and America. However, there is no report available on the chemical composition analysis of the essential oil of *P. ortegioides*, in general, and its other properties (antifungal, antioxidant etc.) in particular. Hence, efforts have been made to investigate the role of essential oil and to determine chemical composition flower, stem, leaf and root of this species. In the current study, we examined the chemical composition of the essential oil, isolated from the aerial parts of *Phryna ortegioides* for the first time, that might be helpful in potential usefulness and chemotaxonomical importance of this species.

GC-MS analyses of the *Silene armeria* L. oil led to the identification of 28 different components, representing 89.03% of the total oil. The oil contained a complex mixture consisting of olefinic hydrocarbons and mono and sesquiterpene hydrocarbon along with some other essential phytochemicals. The major components in the oil detected were 1-butene (39.2%), methylcyclopropane (21.48%), 2-butene (17.97%) and caryophyllene oxide (7.2%). Terpenoid hydrocarbons were the characteristic constituents of the oil of *S. armeria*. Coumarin (0.22%), eugenol (0.21%), α-humulene (0.07%), farnesol (0.05%), linalool (0.12%), pentylfuran (0.09%), benzene acetic acid (0.38%), isovaleric acid (0.05%), β-myrcene (0.08%), 2-butanone (0.07%) and acetophenone (0.08%) were also found to be the trace or minor components of *S. armeria* oil (Bajpai et al., 2008). In this study with *P. ortegioides,* twenty different components were identified representing 90.9% oil. Germacrene D (20.6%), borneol (19.1%), bicyclogermacrene (9.1%) and caryophyllene oxide (7.3%) were identified as major components, β-mrycene (0.3%), α-humulene (0.1%) and 2-pentylfuran (0.2%) were found to be minor components of *P. ortegioides* oil (Table 1).

Like present study, Germacrene D (23.4%) and

Table 1. Chemical profiles of *Phryna ortegioides*.

No	Compounds	RRI	Percentage (%)
1	Camphene	1034	0.8
2	β-mrycene	1061	0.3
3	2-pentyl furan	1064	0.2
4	*p*-cymene	1090	2.3
5	1,8-cineole	1097	0.5
6	α-terpinolene	1138	4.5
7	Linalool	1145	1.7
8	Nonanal	1152	3.7
9	Camphor	1182	0.9
10	Borneol	1200	**19.1**
11	3-cyclohexan-1-ol	1205	4.6
12	β-caryophyllene	1393	2.5
13	α-humulene	1412	0.1
14	Trans-β-ocimene	1418	2.4
16	Germacrene D	1432	**26.6**
17	Bicyclogermacrene	1450	**9.2**
18	Caryophyllene oxide	1510	1.3
19	2-pentadecanone	1630	5.8
20	Nonacosane	1941	4.4
	Total		**90.9**

Table 2. The major components of some Caryophyllaceae taxa.

	Germacrene D	nonanal	nonacosane	γ-dodeca dienolactone	Bicyclo germacrene	2-penta decanone	*p*-cymene	3-methyl tetradecane
1	26.6	3.7	4.4	-	9.2	5.8	2.3	-
2	-	10.5	-	-	-	-	-	17.3
3	-	-	-	-	-	37.6	-	-
4	-	4.6	6.2	-	-	5.1	-	-
5	21.2	-	-	13.7	17.6	-	20.6	-
6	23.4	-	-	6.8	7.5	-	6.7	-
7	12.6	-	-	28.5	-	-	12.5	-

1. *Phryna ortegioides* (Studied sample) **2**. *Cerastium candidissimum (*Couladis & Tzako, 2000)
3. *Herniaria incana* (Lazari *et al*., 2000) **4.** *Minuartia meyeri* (Yayli *et al*., 2006)
5-6-7. *Gysophila bicolor* flower – leaf - stem (Shafaghat & Shafaghatlonbar, 2011)

bicyclogermacrene (17.6%) were also detected among the major components of *Gypsophila bicolor* (Freyn & Sint.) Grossh. from Iran (Shafaghat and Shafaghatlonbar, 2011). According to Yayli et al., (2006), nonacosane (6.2%), 6,10,14-trimethyl-2-pentadecanone (5.1%), nonanal (4.6%) and β-caryophyllene (2.9%) were the main components of the essential oil of *Minuartia meyeri* (Boiss.) Bornm. (Caryophyllaceae). Similarly, in our study 2-pentadecanone (5.8%) and nonanal (3.7%) were determined among the main compounds. But β-caryophyllene (2.5%) was found in lower amounts in this study (Table 1). According to Lazari et al., (2000), *Herniaria incana* Lam. from Greece essential oil contained 6,10,14-trimethyl 2-pentadecanone (37.6%) and palmitic acid (4.0%) were the main components. In our study 6,10,14-trimethyl and palmitic acid were absent, 2-pentadecanone (5.86%) presented only in low percentages (Table 1). In another study, according to Couladis and Tzakou (2000), nonanal (10.5%), geranyl acetone (13.5%), 3-methyltetradecane (17.3%), (E)-beta-ionone (11.1%) and hexahydrofarnesyl acetone (21.7%) were determined as the major components aerial parts of *Cerastium candidissimum* Corr. from Grecee. Whereas only nonanal (3.7%) was detected in the essential oil of *Phryna ortegioides* (Table 1).

3-methyltetradecane was detected as one of the major compounds in the essential oil of *Cerastium candidissimum* (17.3%) (Table 2). However the absence of this compound from the essential oils of *Phryna ortegioides*, *Herniaria incana*, *Minuartia meyeri* and flower-leaf-stem oil of *Gypsophila bicolor* samples are noteworthy (Table 2). According to Shafaghat and Shafaghatlonbar (2011), germacrene D (21.2%, 23.4%, 12.6%), gamma-dodecadienolactone (13.7%, 6.8%, 28.5%) and p-cymene (20.6 %, 6.7%, 12.5%) were the main components in the flower, leaf and stem of *Gypsophila bicolor*, respectively. According to our study germacrene D (20.6%) and *p*-cymene (7.3%) also were the main components of *P. ortegioides* from Turkey. Whereas gamma-dodecadienolactone was not determined in the essential oils of *Cerastium candidissimum*, *Herniaria incana* and *Minuartia meyeri* (Table 2). Bicyclogermacrene was among the major compound of *P. ortegioides* (9.2%) and in flower (17.6%) and leaf (7.5%) oil of *Gypsophila bicolor* (Table 2). On the other hand bicyclogermacrene was not detected as major compound in *Cerastium candidissimum, Herniaria incana, Minuartia meyeri* and stem oil of *Gypsophila bicolor* (Table 2).

Jovanovic et al., (2009) reported that nonanal (9.9%), (Z)-3-hexenol (8.5%), hexahydrofarnesyl acetone (5.3%) and methyl 3-hydroxyoctadecanoate (4.5%) were present in high percentages volatile oil of *Minuartia recurva* (All.) Schinz et Thell. subsp. *recurva* (Caryophyllaceae) from Serbia. Whereas it is noteworthy that, (Z)-3-hexenol (8.5%), hexahydrofarnesyl acetone (5.3%) and methyl 3-hydroxyoctadecanoate (4.5%) were not identified at all in the present study (Table 1). The volatile constituents from flower, leaf and stem of *Gypsophila bicolor* growing in Iran were obtained by hydrodistillation and analyzed by GC and GC/MS. The flower oil was characterized by high amounts of germacrene-D (21.2%), *p*-cymene (20.6%), bicyclogermacrene (17.6%), γ-dodecadienolactone (13.7%) and terpinolene (9.4%). Twenty-four constituents representing 97.4% of the leaf oil were identified of which gemacrene-D (23.4%), terpinolene (14.5%), bicyclogermacrene (7.5%), γ-dodecadienolactone (6.8%), *p*-cymene (6.7%) and cis-β-ocimene (6.3%) were major components. The main components of the stem oil were γ-dodecadienolactone (28.5%), bicyclogermacrene (14.8%), germacrene-D (12.6%), *p*-cymene (12.5%), terpinolene (11.6%) and trans-β-ocimene (4.2%) (Shafaghat and Shafaghatlonbar, 2011). In this study with twenty constituents representing 90.9% of the aireal part oil were identified of which germacrene-D (20.6%), borneol (19.1%), bicyclogermacrene (9.1%) were major, α-terpinolene (4.5%) and trans-β-ocimene (2.4%) minor components (Table 1).

Conclusion

This study demonstrates the occurrence of germacrene-D/borneol/bicyclogermacrene chemotype in *P. ortegioides*. Besides, some *Gypsophila* and *Minuartia* taxa have different types of essential oils, like germacrene-D, *p*-cymene, bicyclogermacrene chemotype in *G. bicolor* from Iran (Shafaghat and Shafaghatlonbar, 2011) and nonacosane, 6,10,14-trimethyl-2-pentadecanone, nonanal chemotype in *M. meyeri* (Yayli et al., 2006). According to the chemotype results, some variations can be seen in *Phryna, Silene, Cerastium*, *Gypsophila* and *Minuartia* taxa. So, these differences both in the oil content and composition may be due to different reasons such as climatic and genetic factors, agronomical practices, or plant chemotype or nutritional status.

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