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Research article

An investigation on the determination of diurnal and ontogenetic variabilities of essential oil content and composition in *Hypericum triquetrifolium* Turra (Hypericaceae)

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Abstract: *Hypericum* genus includes especially species used as depression and wound healing in alternative and modern medicine. In this study, *Hypericum triquetrifolium* which grows naturally in western of Turkey (Kazdağ Mount / Edremit Balıkesir) were investigated essential oil content, essential oil composition, diurnal and ontogenetic variabilities. The diurnal and ontogenetic variabilities of the species were first studied. Essential oils in part of the aerial plant raised during flower ontogenesis and achieved the highest level at full flowering and decreased at the fresh fruiting phase. The highest level at full flowering is 0.30% while the lowest level of fresh fruiting is 0.09%. As a result of the study, 27 components were detected from the aboveground parts of *H. triquetrifolium* at the before flowering, beginning of flowering and full flowering stages. The major components were obtained caryophyllene (32.9%) and caryophyllene oxide (10.8%) at the before the flowering stage, 3-methyl nonane (17.1%) and caryophyllene (14.9%) at the flowering stage and 3-methyl nonane (43.5%) and α-pinene (17.6%) at the fresh fruiting stage.
Keywords: Diurnal variability - Essential oil components - *Hypericum triquetrifolium* - Ontogenetic variability.

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INTRODUCTION

Hypericum L. is an important member of Hypericaceae family and is a large genus of herbs or shrubs included medicinal species as *Hypericum perforatum* Turra which distributed in temperate areas of the world (Ciccarelli *et al.* 2001). The *Hypericum* contains about 500 species by the most recent count that has been classified into 36 taxonomic sections (Crockett & Robson 2010). The *Hypericum* is represented in Turkey by 96 species (106 taxa) of which 48 are endemic (Aslan *et al.* 2012). *Hypericum* species are morphologically characterized by the presence of secretory tissues including glands which to be dark or light-coloured on the stem, leaves and flowers (Campbell & Delfosse 1984, Ciccarelli *et al.* 2001, Cirak *et al.* 2006, Ayan *et al.* 2007).

Hypericum species mostly are showing the distribution South Europe, Africa, Cyprus, Syria, Syrian Iraq, Iran, Njery and Turkey. It is a perennial plant that grows in open dry, sandy ground and mesophytic areas as habitat in Turkey (Robson 1967).

Hypericum taxa have traditionally been used in the treatment of wounds, burns and stomachic diseases by local peoples in Turkey (Baytop 1999, Selvi & Pasa 2011). In addition it has been ethnomedical used in the world as antiseptic, antinociceptive, sedative, antimicrobial, antioxidant, and cytotoxic (Conforti *et al.* 2002, Couladis *et al.* 2002, Bertolli *et al.* 2003, Kizil *et al.* 2004, Pistelli *et al.* 2005).

In this study, the determination of diurnal and ontogenetic variabilities of essential oil content and composition in *H. triquetrifolium* distributed in naturally Turkey were investigated. The diurnal and ontogenetic variabilities of this species were first studied. With study, the amount of essential oil contained the plant; in

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which part of the plant, during which development period and at what times of the day and the essential character of essential oils will be determined. In addition, this study is important for the determination of harvest time in Western Anatolia conditions for *H. triquetrifolium*.

MATERIALS AND METHODS

Hypericum triquetrifolium was gathered at different stages of plant development from Kazdağı (Balıkesir) in between April and August. The collected locality is Turkey, B1 Balıkesir: Edremit, Kazdağ Mount (Ida Mount), Şahindere canyon, Olive grove fields, 39° 35' 23.67" N, 26° 50' 39.49" E, 125 m, 12.04.2016, (SV 1655). General habitus of *H. triquetrifolium* is figure 1.

Figure 1. General habitus of *Hypericum triquetrifolium* Turra.

For ontogenetic variability, herb samples were taken at different growth stages (before flowering, beginning of flowering and full flowering). At the same time, herb samples were taken at different times of day (09.00 am, 12.00 pm, 4.00 pm) for diurnal variability. Flower samples were taken for essential oil rate and its components were investigated. At the beginning of the flowering process, the shoots which had green bud were harvested. Again, at the full flowering process, only shoots with fully opened flowers were harvested. The plant drugs were dried at room temperature (21ºC) and later oil content of plant parts (50 g each sample*)* were determined.

The oil composition was identified by GC-MS and GC-MS analyzes were carried out in TUBITAK (MAM). Helium was used as carrier gas at a constant flow rate of 1 ml/min and 1 µl of the sample was injected.

The GC temperature program was set as follows; 50ºC, stand for 5 minutes, increase to 250ºC at 5ºC / min and stand for 10 minutes. The temperature of the MS transfer line was set at 220ºC. This study was used by the Thermo Scientific TSQ GC-MS/MS.

The soil structure of the area gathering of the samples was ecologically sandy, [sand (69%), silt (23%) and clay (7%)]. pH value (6.8) and organic matter (6.9%), The average temperature of the place where the trial is 22.2ºC, mean rainfall 26.9 mm and relative humidity 61.1% in 2016.

The differences between these tools were compared with Duncan's multi-class test (Duncan's test) and presented in table 1. The differences between that means were compared by Duncan’s test (Duncan’s multiple range test) and given in table 1.

RESULTS AND DISCUSSION

Total essential oils content in *H. triquetrifolium* during ontogenetic development was higher in full flowering stage (0.20–0.30 %), followed by before flowering stage (0.25–0.28 %) and fresh fruiting stage (0.09–0.12 %) (Table 1). The results of this study showed that diurnal and ontogenetic variability were significantly affected by essential oils ($p < 0.01$).

Table 1. Total essential oils content (%) and changes during collecting times of the day and development stages of *Hypericum triquetrifolium* Turra.

Diurnal Collecting Times	Developmental Stages			
	Before flowering (%)	Full flowering (%)	Fresh fruiting (%)	Mean
09:00 am	0.28 b	0.20 e	0.10 g	0.19 b
12:00 am	0.25 cd	0.23 d	**0.09 h**	0.19 b
16:00 pm	0.26 c	**0.30 a**	0.12 f	**0.23 a**
Mean	**0.26 a**	0.24 b	0.10 c	0.20

Note: There is no statistically significant difference (p>0.05) between figures including the same letters in the columns.

Table 2. Variability of essential oils content of *H. triquetrifolium* within a day during the course of ontogenetic (%).

KI*	RT**	Compounds	Before flowering 09.00 am	Before flowering 12.00 am	Before flowering 4:00 pm	Full flowering 09:00 am	Full flowering 12:00 am	Full flowering 4:00 pm	Fresh fruiting 09.00 am	Fresh fruiting 12.00 am	Fresh fruiting 4.00 pm
939	5.44	α-pinene	3.8	2.7	3.5	5.8	4.5	4.7	17.6	16.4	16.5
971	6.87	3-methyl nonane	11.6	10.5	13.0	16.4	16.9	17.1	43.5	42.4	40.7
979	7.55	β-pinene	1.2	1.4	1.3	1.2	1.4	1.7	1.6	1.4	2.0
1003	7.90	α-phellandrene	0.2	0.2	0.4	0.5	0.3	0.5	0.8	0.7	0.9
1025	8.60	p-Cymene	-	-	-	0.1	0.1	0.1	-	0.1	0.1
1029	8.75	Limonene	0.3	0.2	0.3	0.6	0.4	0.2	1.2	1.7	1.3
1037	9.20	β-Ocimene	-	-	-	0.4	0.2	0.2	0.8	0.4	0.7
1063	10.10	2-methyl decane	0.1	0.2	0.2	3.2	4.1	3.0	4.6	4.9	5.0
1100	11.36	Undecane	0.8	1.1	1.4	1.5	2.5	2.8	3.8	3.2	4.0
1299	17.20	Carvacrol	7.1	5.9	7.6	1.2	0.7	0.4	0.2	-	-
1377	19.05	Copaene	3.2	2.8	2.4	3.3	3.8	4.9	0.5	0.7	0.5
1388	19.27	β-bourbonene	0.4	0.3	0.2	0.2	0.3	0.3	-	0.1	0.2
1409	20.12	Caryophyllene	32.9	30.6	28.4	14.9	14.2	12.4	10.4	11.8	11.8
1426	20.38	β-gurjunene	0.5	0.5	0.7	0.9	0.7	0.6	0.1	0.2	0.5
1455	20.97	α-humulene	3.2	2.9	3.1	2.9	2.8	2.4	1.7	1.9	1.5
1447	21.20	Aromadendrene	1.8	1.6	1.6	0.3	0.9	1.5	0.1	0.1	0.3
1475	21.57	α-amorphene	2.4	2.8	3.1	2.8	3.1	4.2	0.2	0.7	1.4
1485	21.65	Germacrene-D	7.9	7.6	7.6	13.6	12.6	10.4	3.1	2.9	3.0
1480	22.00	τ-muurolene	1.3	1.1	1.5	0.8	0.5	0.7	1.1	0.6	0.9
1500	22.20	α-muurolene	0.9	1.1	1.0	0.7	1.1	1.4	0.4	0.1	0.6
1514	22.50	Gamma-cadinene	1.1	1.0	1.7	1.2	1.8	2.3	1.3	1.4	1.4
1539	22.72	α-Cadinene	4.0	3.8	4.4	4.1	4.9	5.7	1.3	1.0	1.1
1583	24.03	Caryophyllene oxide	10.8	10.5	10.2	9.1	8.0	7.2	1.5	1.4	1.8
1660	25.63	α-cadinol	0.8	0.9	0.9	0.4	0.3	0.4	0.2	0.1	-
1675	26.00	valeranone	0.7	0.8	0.8	0.3	0.2	0.3	0.1	-	-
1686	26.30	Bisabolol	1.1	1.1	1.3	0.5	0.7	0.6	-	-	0.1
1943	29.71	Phytol	1.0	1.1	1.4	0.5	0.5	0.7	0.1	0.1	-

Note: *Kovats Index; ** Retention Time

Chemical concentrations vary considerably during ontogenesis in a medicinal plant, not only the concentrations of plant chemicals fluctuate through the season, but they can also be short-lived and experience rapid turnover (Smith *et al.* 1996).

Our study, the major constituents of the oil were 3-methyl nonane (10.5–43.5 %), carvacrol (0.2–7.6 %), caryophyllene (10.4–32.9 %), germacrene-D (2.9–13.6 %), α-pinene (2.7–17.6 %) and caryophyllene oxide (1.4–10.8 %). Most of them have been previously reported in the essential oil of *H. triquetrifolium* (Petrakis *et al.* 2005, Cirak *et al.* 2006, Hosni *et al.* 2011).

From a compositional standpoint, the chemical composition of *H. triquetrifolium* essential oils from different locations has been reported. Petrakis *et al.* (2005) studied the essential oil of Greece specimens and found that α-pinene, n-nonane, β-caryophyllene, 3-methylnonane and 2-methyloctane in phenological stage. Hosni *et al.* (2011) studied phenological variability of secondary metabolites from *H. triquetrifolium* and found that β-caryophyllene, α-pinene, n-nonane, 2-methyloctane, germacrene-D and n-octane. Bertolli *et al.* (2003) were investigated of volatile constituents of leaves and flowers of *H. triquetrifolium* in Italy and analyses results showed the major compounds of the leaf and flowers essential oils that were the α-pinene, β-pinene, β-caryophyllene, n-nonane, sabinene, myrcene, caryophyllene oxide and germacrene-D.

Variability of essential oils content of *H. triquetrifolium* within a day during the course of ontogenetic is listed table 2. As can be seen in table 2 the studied oils were resolved into 27 components at the before flowering, full flowering and fresh fruiting stage respectively.

At the before flowering, the oils consisted mainly of caryophyllene (32.9%), caryophyllene oxide (10.8%), 3-methyl nonane (10.5%), carvacrol (7.6%); germacrene-D (7.9%), α-cadinene (4.4%) and α-humulene (3.2%). At the full flowering stage, the oils consisted mainly of α-pinene (5.8%), 3-methyl nonane (17.1%), copaene (4.9%), caryophyllene (14.9%), α-amorphene (4.2%), germacrene-D (13.6%), α-cadinene (5.7%) and caryophyllene oxide (9.1%). At the fresh fruiting stage oils consisted mainly of α-pinene (17.6%), 3-methyl nonane (43.5%), 2-methyl decane (5.0%), undecane (4.0%), caryophyllene (11.8%) and germacrene-D (3.1%).

The diurnal variability of *H. triquetrifolium* has not been reported so far. However, same studies with Hypericum taxa by various authors have been described *H. triquetrifolium* (Schwob *et al.* 2004, Cirak *et al.* 2011, Hosni *et al.* 2011, Pasa 2013).

CONCLUSION

Ontogenetic and diurnal variabilities were applied on *H. triquetrifolium* and 27 essential oil components were detected. The major components of the oil of *H. triquetrifolium* aerial parts were caryophyllene and caryophyllene oxide.

The essential oils of the above-ground parts of the plant increased during flowering ontogenesis and reached the highest level in full flowering. Afterwards, it decreased at the beginning of the flowering phase. The highest level at full flowering 0.30% and the lowest level fresh fruiting 0.09%.

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REFERENCES

Aslan S, Ekim T, Vural M & Babac MT (eds) (2012) *Türkiye Bitkileri Listesi (Damarlı Bitkiler)*. Nezahat Gökyiğit Botanik Bahçesi ve Flora Araştırmaları Derneği Yayını, İstanbul.

Ayan AK, Yanar O, Cirak C & Bilginer M (2007) Morphogenetic and diurnal variability of total phenols in some *Hypericum* species from Turkey during their phonological cycles. *Bangladesh Journal of Botany* 36(1): 39–46.

Baytop T (1999) *Therapy with Medicinal plants in Turkey.* Istanbul University press, Istanbul, pp. 66–167.

Bertolli A, Menchini F, Mazzetti M, Spinelli G & Morelli I (2003) Volatile constituents of leaves and flowers of *Hypericum triquetrifolium* Turra. *Flavour Fragrance Journal* 18: 91–94.

Campbell MH & Delfosse ES (1984) The biology of Australian weeds *Hypericum perforatum* L. *The Journal of the Australian Institute of Agricultural Science* 50: 50–63.

Ciccarelli D, Andreucci AC & Pagni AM (2001) Translucent glands and secretory canals in *Hypericum perforatum* L. (Hypericaceae): morphological, anatomical and histochemical studies during the course of ontogenesis. *Annals of Botany* 88: 637–644.

Cirak C, Sağlam B, Ayan AK & Kevseroğlu K (2006) Morphogenetic and diurnal variability of hypericin in

some *Hypericum* species from Turkey during the course of ontogenesis. *Biochemical Systematics and Ecology* 34: 1–13.

Cirak C, Radusiene J, Janulis V, Ivanauskas L, Camas N & Ayan AK (2011) Phenolic constituents of *Hypericum triquetrifolium* Turra (Guttiferae) growing in Turkey: variability among populations and plant parts. *Turkish Journal of Biology* 35: 449–456.

Crockett SL & Robson NKB (2010) Taxonomy and chemotaxonomy of the genus Hypericum. *Medicinal and Aromatic Plant Science and Biotechnology* 1: 1–13.

Conforti F, Statti GA, Tundis R, Menichini F & Houghton P (2002) Antioxidant activity of methanolic *Hypericum triquetrifolium* Turra extract of aerial part. *Fitoterapia* 73: 479–483.

Couladis M, Baziou P, Verykokidedou E & Loukis A (2002) Antioxidant activity of polyphenols from *Hypericum triquetrifolium* Turra. *Flavour Frangrance* 16: 769–770.

Hosni K, Msaada K, Taarit MB & Marzouk B (2011) Phenological variabilitys of secondary metabolites from *Hypericum triquetrifolium* Turra. *Biochemical Systematics and Ecology* 39: 43–50.

Kizil G, Toker Z, Özen HC & Aytekin C (2004). The antimicrobial activity of essential oils of *Hyperiricum scabrum, Hypeicum scabroides* and *Hypericum triquetrifolium. Phytotherapy Research* 18: 339–341.

Petrakis PV, Couladis M & Roussis V (2005) A method for detecting the biosystematics significance of the essential oil composition: the case of five Hellenic *Hypericum* L. species. *Biochemical Systematics and Ecology* 33: 873–898.

Pistelli L, Bertolli A, Morelli I, Menichini F, Musmannu RA, Di Maggio T & Coratza G (2005) Chemical and antibacterial evaluation of *Hypericum triquetrifolium* Turra. *Phytotherapy Research* 19: 787–791.

Robson NKB (1967) *Hypercium. Flora of Turkey and the East Aegean Islands*, Vol 2. Edinburgh University Press, Edinburgh.

Selvi S & Paşa C (2011) Türkiye Florasında Bulunan *Hypericum* (Guttiferae) Türleri ve Tehlike Durumları. IX. Tarla Bitkileri Kongresi, 12–15 Eylül 2011, Bursa, pp. 1458–1461.

Schwob L, Bessiere JM, Masotti V & Viano J (2004) Changes in essential oil composition in Saint John's wort (*Hypericum perforatum* L.) aerial parts during its phonological cycle. *Biochemical Systematics and Ecology* 32: 735–745.

Smith R, Caswell D, Carriere A & Zielke B (1996) Variability in the ginsenoside content of American gingseng, Panax quinquefolius L. roots. *Canadian Journal of Botany* 74: 1616–1620.

Pasa C (2013) Kazdağlarında yayılış gösteren bazı *Hypericum* türlerinde uçucu yağ oranı ve bileşenlerinin diurnal, ontogenetik ve morfogenetik varyasyonunun belirlenmesi üzerine bir araştırma, *Namık Kemal Universitesi, Fen Bilimleri Enstitüsü, Doktora Tezi, Namık Kemal Universitesi, Fen Bilimleri Enstitüsü, Tekirdağ*, pp. 135–142.