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Full Length Research Paper

Environmental adaptibility of tansy (*Tanacetum vulgare* L.)

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Ecological role of essential oils is reflected in the interaction of plants with environmental factors. Environmental adaptability of the plants can be assumed from essential oil contents. Essential oils are agents, which communicate with the plant environment. Tansy (*Tanacetum vulgare* L.) was selected for laboratory research since it belongs to urban flora and vegetation where the imperative to adapt is high. Sample plants were collected from two site locations: Ada Huja (industrial zone) and Topčider (green area) in Belgrade, Serbia. A GC-MS analyses, to determine quantitative and qualitative composition of essential oils were used. The results of the research indicate that tansy from both locations shows a high degree of adaptability. Tansy from Ada Huja had larger total amount of essential oils comparing to plants from Topčider. This study presents the comparative results of laboratory research. Specific components of the essential oil samples of plants from both site locations are identified.

Key words: Environmental adaptability, site location, tansy, essential oil.

INTRODUCTION

Among the plants that could become a potential source of usable substances is the *Tanacetum* genus comprising about 150 species. About 30 of them have been also practically utilized. Some of the most popular ones are *Tanacetum vulgare*, *T. parthenium* and *T. balsamita* (Vaverkova et al., 2008). The *Tanacetum* spieces are rich in essential oils, bitter components and sesquiterpene lactones. They are used widely because of their anti-inflammatory, antihistaminic and insecticide effects. Some species of this genus have been used traditionally in cosmetics, medicine and also in phytotherapy. The content of substances in species of the *Tanacetum* genus depends on many factors. Therefore, the precise identification of them is very difficult. Based on several studies the major content of substances of the drug seem to be essential oils, flavonoids, bitter compounds, tannins and alkaloids.

Essential oil and extracts from *Tanacetum* L. plants are compounds with various therapeutical uses. The biological activity of the *Tanacetum* L. genus is connected with terpenes in essential oils (Vaverkova et al., 2008). Many studies have been published about the composition of essential oils (Gallino, 1988; Uchio et al., 1981; De Pooter et al., 1988; Goren et al., 2001; Williams et al., 1999; Baser et al., 2001; Gallori et al., 2001; Akpulat et al., 2005; Fonseca et al., 2005). In one of them (Stoianova-Ivanova et al., 1983) the analysis of steam distilled flowers and leaves as well as the whole aerial part of *T. indicum* L. was described. The isolated oil was analyzed using the GC and GC/MS methods. It was found that main components were borneol, chrysanthenone and bornyl acetate. The most extensive investigation (on the composition of essential oils from tansy were performed by Gallino (1988) and Uchio et al. (1981).

The detailed analysis of volatile compounds from *T. vulgare* was conducted also by De Pooter et al. (1988) and Goren et al. (2001). The high biological activity between sesquiterpene lactone parthenolide and flavonoids was found in *T. parthenium* (Williams et al., 1999). Other studies reported the composition of essential oils from *Tanacetum* spp., *T. argyrophyllum, T. argenteum* and *T. praeteritum* (Goren et al., 2001; Baser et al., 2001).

Among recent studies, the most remarkable results were reported by Baser et al. (2001). The authors

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identified numerous components representing 72-91% of total essential oils amount.

The main component of *T. argyrophyllum* L. was α-thujone (Vaverkova et al., 2008). Caryophyllene oxide and α-thujone were the main components of *T. argenteum* L. oils. Borneol, 1,8-cineole and bornyl acetate are the main constituents in *T. praeteritum* subsp. *praeteritum* (Vaverkova et al., 2008).

Production of essential oil is an indicator of plant adaptation on habitat conditions. Ecological role of essential oils is reflected in the interaction of plants with environmental factors. It helps to plant that easier adopt to the environmental stress conditions: drought, intense radiation, high temperature and heavy metal contents (Abu-Darwish and Abu-Dieyeh, 2009). Essential oils are not constant in the qualitative and quantitative terms. They are changed continuously, due to the requirements of the environment and to individual survival. Concentrations and amount of essential oil depends on the external environmental condition (Schearer, 1984). Natural selection favors the survival of plants with the higher concentration of essential. It means a higher adaptive value.

In a variety of stressful conditions (e.g. unfavorable geological, pedological, climatic, or biotic conditions) there is an, increase in the number of plants rich in essential oils (Salamon, 2008). The role of essential oils is reflected in the importance of smell, which on the other hand, enable the survival of one plant, for attracting pollinators, or survival, as are other warning stimulus and refuse herbivore insects and surrender (Werker, 1993; Sangwan et al., 2001). *T. vulgare* (*Asteraceae/Compositae*, syn. *Chrysanthemum vulgare* L., common name tansy) has a variety of interesting pharmacological properties. Tansy is a common and widespread plant in Serbia. It can be cultivated and it also grows spontaneously.

T. vulgare is principally used in tradition Maroccon medicine as antihypertensive remedy. It is used as an antispasmodic, antihelminthic, carminative, stimulant to abdominal viscera, tonic, emmenagogue, antidiabetic, diuretic and antihypertensive. Its extract has been reported to exhibit antitumor (Konopa et al., 1967), anti-inflammatory (Williams et al., 1999), antioxidant (Bandoniene et al., 2000; Mantle et al., 2000), antimicrobial activity (Holetz et al., 2002) and antimalarial effect (Jansen, 2006). It has also been reported to have an effect on experimental gastric ulcer (Tournier et al., 1999) and to be used in the treatment of various infectious diseases (Holetz et al., 2002). *Tanacetum* water extract is also prescribed to treat hypertension. It contributes to coronary artery disease and infarction, stroke, cardiac failure, renal insufficiency, which can be prevented by effective antihypertensive therapy. Also, tansy has a lot of use in the treatment of wounds. The aqueous extract of *Tanacetum* possesses vasorelaxing properties *in vitro* (Lahlou et al., 2008).

Primary research aim of this study was to investigate adaptibility of the tansy to different stress environmental condition. Plant has been collected from two specific location in Belgrade, Serbia. One of them (Ada Huja) is a polluted industrial zone, but the other one (Topčider) is unpolluted park area.

The goal was to measure the quantity and quality of essential oil, as adaptibility indicator and to prove the statement that the plants answer on environmental stress by production of essential oil.

MATERIALS AND METHODS

Whole plants of *T. vulgare* were collected from two different ecological habitats: Ada Huja (industrial zone) and Topčider (green park zone). It was the time at the end of August when the blossom stage of development was optimal for essential oil extraction. The plant reaches a height of 150 - 200 cm at that time. Tansy was growing on damp hill and waste materials on Ada Huja and next to the Topčider river in Topčider park. Plant materials were harvested in three different places on each locality. The sample size was 10 plants per the place. Fresh mass of one plant was approximately 300 g. Plants were harvested three times during the one flowering season. Total sample size was 90 per the site locality. Fresh plant material was air dried between the sheets of porous paper, for about a month. All subsequent investigations were made with this prepared material marked as "air-dry matter".

Methods for determination of the content and composition of essential oil

Essential oil content was determined by distillation with water steam according to pharmacopoeia (Radulović et al., 2007). The composition of essential oil was determined by the method of gas chromatography (GC) and a combination of gas chromatography-mass spectrometry (GC-MS). GC was performed on the instrument of production Varian (USA), model 3400, which was equipped with split/splitless injector (1:20), working temperature 266^{0}C. Column production JW (USA) type DB-5, length 30 m, internal diameter of 0.25 mm and film thickness of 0.25 m. Bearing gas was hydrogen. Flow 1 ml/min at 210^{0}C. Temperature of column was liner programmed from 60 to 285^{0}C rate of 4.3^{0}C/min. Flame ionization detector (feed) to 300^{0}C.

GC-MS was performed on the production of mass spectrometers Finnigan Mat (Germany) model 8230, with the ion trap detector (ITD) model of ITD-705 the same manufacturer. Hromatograf gas associated with the mass spectrometers, as well as working conditions were the same as for GC, with the feed off. Used a column of production Supelco (USA), the type of PTE-5 the same dimensions, as well as columns for GC. Output column is connected through the interface directly from the ITD at a temperature of 240^{0}C. Scanning area of 39-333D, the 1 scan/s. Bekgraund the ion mass 33D. Used the software ITDS version 3rd GC-MS data were analyzed using AMDIS, version 2 and compilation of photoelectron spectra library for the instrumental analysis of chemical faculties and the Institute of Chemistry, technology and metallurgy in Belgrade in 3833 with the spectrum of 1666 compounds with retention (Kovats) index (RI). RI is calibrated using a series of *n*-alkanes C_8H_{18}-$C_{24}H_{50}$ under the same experimental conditions as for GC analysis (Stein, 1999).

Particular essential oil components were identified by standards as well as by comparison of their relative retention times, retention indices and mass spectra with those of the WILEY 275 spectral library (2001).

Table 1. Comparative GC-MS analysis of fresh oil of *T. vulgare* plants, collected at polluted and non polluted sites in Belgrade, Serbia.

Constituents	Relative content [%]				
	RI	Ada Huja	Percent	Topčider	Percent[c]
α-phelandrene	1005	tr.	/	0.6	>80
p-cumene	1023	tr.	/	0.5	60-70
β-phelandrene	1030	tr.	/	0.5	>80
santolina alcohol	1035	1.7	60-70	tr.	/
Linalool oxide	1050	tr.	/	1.7	75-80
artemisia ketone	1065	1,1	>80	0.7	75-80
sabinene hydrate	1080	tr.	/	0.8	70-75
undecane	1100	2.7	70-75	tr.	/
α -thujone	1110	0.9	>80	tr.	/
β-thujone	1116	8.3	>80	1.3	60-70
pinocarvone	1163	0.8	75-80	tr.	/
p-cresol acetate	1168	tr.	/	0.8	60-70
cis-pinochamphone	1176	tr.	/	2.8	>80
trans-carveol acetate	1203	5.1	60-70	14.5	60-70
piperitone	1235	0.9	>80	3.1	>80
Linalool oxide acetate	1389	0.8	70-75	9.9	60-70
Germacrene-D	1480	1.1	>80	tr.	/
trans-chrysanthenol	1504	tr.	/	1.4	70-75
spathulenol	1562	5.5	75-80	tr.	/
trans-chrysanthenyl acetate	1588	47.9	>80	36.9	>80
longiborneol	1594	0.9	60-70	tr.	/
α -cadinol	1655	3	60-70	tr.	/
Total identified		80.7		75.5	

RI – retention index (Kovats); tr. - traces
All contents are relative, comparing according to total amount of essencial oil, which is found in the plant.

Statistics and repetition

Three measurement repetitions had been performed per each of the 180 samples. The results were analysed using completely randomized design and tested according to least significant difference (LSD) test.

RESULTS

Essential oil contents of *T. vulgare* from two different localitetes Ada Huja and Topčider were investigated and presented in this paper. Twenty two different major and minor constituents from tansy, using the GC/MC chromatography, were separated and identyfied. Retention indices (RI) and percentage are showen in Table 1.
Results in percentage (according to total amount of essential oil) and identified specific components of the essential oil of plants from both sites are presented numerically in Table 1.

The results showed that only six components (artemisia ketone, *trans*-chrysanthenyl acetate, *trans*-carveol-acetate, linalool-okside-acetate, piperitone and β-thujone), of about 14 identified oil compounds, were found in the plants from both sites. The total content of identified essential oil in plants originated on Ada Huja site was 80.7%, while in Topčider plants was 75.5%, related to total amount (Table 1). Essential oil of *Tanacetum vulgare* plants from Ada Huja and Topčider showed differences in the contents of an eight characteristic compounds.

Specific essential oil compounds of *T. vulgare* from Ada Huja were: germacrene D, α-cadinol, longiborneol, pinocarvone, santolina alcohol, spathelenol, α-thujone and undecane. However, compounds characteristic from *T. vulgare* growth on Topčider were: α-phelandrene, β-phelandrene, *trans*-chrysanthenol, *p*-cresol-acetate, *p*-cumene, linalool oxide, *cis*-pinochamphone and sabinene-hydrate.

Trans-chrysanthenyl acetate and *trans*-carveol-acetate (more than 50%), according to total amount of essential oil were main compounds in plants from both localities.

DISCUSSION

Tansy plants from Ada Huja showed higher total amount of essential oil than plants from Topčider (Table 1). Tansy plants growth on Ada Huja and Topčider locations belong to the *trans*-chrysanthenyl acetate chemotype (Table 1), which is one of four different chemotypes of this species identified in Serbia (thujone, *trans*-chrysanthenyl acetate, chrysanthenyl and camphoric chemotype) (Popov et al., 2001).

Trans-chrysanthenyl acetate is the main component of *T. vulgare* L. from the investigated sites. The results presented in this paper showed that the *trans*-chrysanthenyl acetate is the most important component of tansy essential oil and that it is in correlation with the results of Popov et al. (2001). They performed a research on different localities in Serbia (Bor, Deligrad, Aleksinac, Stig, Brezovica, Sevce, Aradac, Zlatar) and showed that the *T. vulgare* in all localities had the same chemotype, but in a very wide range of 0.09 to 83.23%, referring to total amount of essential oil. Toxic β-thujone was found in the essential oil of tansy plants growth on contaminated Ada Huja site (8.3%), while in the essential oil of Topčider plants the concentration was much less (1.3%), both comparing to total amount. This results was expected and it was in accordance with the difference between contamination of the industrial and green area zone. Extremely high content of toxic thujone in the tansy is very dangerous for human health (Gallino, 1988). That is why the site location, from where the plants are harvested for medical purposes, is important. *Trans*-chrysanthenol was found in the essential oil from Topčider plants in very small concentration (1.4% of total amount). Camphor is not found in the essential oil plant *T. vulgare* L. of any investigated habitats in the territory of Belgrade.

The comparision of the results in this experiments with the data published by other authors (Schearer, 1984; Vaverova et al., 2008) point out that the values obtained in this paper are compatible with the published one.

In conclusion, the research presented in this paper demonstrates that the specific essential oils compounds are adequate to represent the *T. vulgare* environmental adaptability. The total amount of essential oil in the tansy found from both industrial and green area locations, analyzed in the laboratory, proves a higher degree of representative compounds in the plants from Ada Huja contaminated area. It could serve as an indicator of the adaptability of this species on anthropocentric and anthropogenic environmental conditions, as well as its usability. Further experiments are required to isolated and to identify all other active principles also responsible for tansy ecological plasticity.

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