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Farukh S. Sharopov
Institute of Pharmacy and
Molecular Biotechnology,
Heidelberg University
Im Neuenheimer Feld 364,
Heidelberg, Germany

William N. Setzer
Department of Chemistry,
University of Alabama in Huntsville
Huntsville, AL 35899, USA

Salomiddin J. Isupov
Tajik State Medical University
Rudaki str. 139, 734025, Dushanbe,
Tajikistan

Michael Wink
Institute of Pharmacy and
Molecular Biotechnology,
Heidelberg University
Im Neuenheimer Feld 364,
Heidelberg, Germany

Correspondence:
Michael Wink
Institute of Pharmacy and
Molecular Biotechnology,
Heidelberg University
Im Neuenheimer Feld 364,
Heidelberg, Germany

Composition and bioactivity of the essential oil of *Tanacetum parthenium* from a wild population growing in Tajikistan

Farukh S. Sharopov, William N. Setzer, Salomiddin J. Isupov, Michael Wink

Abstract

The essential oil of *Tanacetum parthenium* (L.) Schultz-Bip. was extracted by hydrodistillation and analyzed by gas-liquid chromatography - mass spectrometry (GLC-MS). Eight components were identified representing 99.8% of total oil composition. The major components were camphor (69.7-94.0%), camphene (1.7-12.2%), and bornyl acetate (4.2-8.7%). According to DPPH and ABTS analyses the volatile oil had an antioxidant activity with IC₅₀ values of 4.82 and 0.96 mg/ml, respectively. Lipid peroxidation was inhibited by 37.1% by 1.125 mg/ml oil. The cytotoxicity of the oil was tested against HeLa, CCRF-CEM and CEM/ADR5000 cancer cell lines: IC₅₀ values were 158.6 µg/ml for HeLa, 69.5 µg/ml for CCRF-CEM, and 83.9 µg/ml for CEM/ADR5000 cell lines. The essential oil of *T. parthenium* inhibits soybean 5-lipoxygenase (5-LOX) with an IC₅₀ value of 21.6 mg/ml indicating a low anti-inflammatory activity.

Keywords: *Tanacetum parthenium* (L.) Schultz-Bip., essential oil, camphor, chemotype, antioxidant, cytotoxicity.

1. Introduction

Feverfew, *Tanacetum parthenium* L. Schultz-Bip., is a member of the daisy family (Asteraceae); it has more than ten synonyms commonly used in the literature including *Chrysanthemum parthenium* and *Pyrethrum parthenium*. This herb is native to Eurasia and cultivated widely around the world. Feverfew is an old medicinal plant which has traditionally been used for reducing fever, women's ailments, inflammatory conditions, psoriasis, toothache, insect bites, rheumatism, asthma and stomachache [1-3]. It has been increasingly employed for the treatment of migraine [3].

Feverfew is an aromatic plant that is rich in essential oil. A number of studies have addressed the chemical composition of its essential oil [4-9]. According to these reports the essential oils which had been collected from different locations differed in their chemical profile. Two distinct chemotypes have been identified; one with camphor/chrysanthenyl acetate and the other with camphor/camphene as main constituents.

The aim of this study was to investigate the phytochemistry of the essential oil of *T. parthenium* from a wild population growing in Tajikistan in Central Asia. Furthermore, its antioxidant, anti-inflammatory activities, and cytotoxicity were investigated. A literature review indicated that there are no previous reports on the composition or bioactivity of the essential oil of *T. parthenium* from Tajikistan.

2. Materials and Methods

2.1 Plant Material

Aerial parts of *T. parthenium* were collected from the Chormaghzak village, Yovon region, Tajikistan, 1300 m above sea level, (38°24'45", 68°10'24") during its flowering season on 17 July 2012 and 8 June 2013. Plants were authenticated by Prof. S. Isupov and compared with the deposited voucher specimen in the herbarium of the Institute of Botany, Plant Physiology and Genetics of the Tajikistan Academy of Sciences.

2.2 Oil Collection

Air-dried samples were crushed and 300 g were hydrodistilled for 3 hours according to the standard procedure of "a steam distillation technique" described in the Pharmacopoeia [10]. The yield of essential oil was 0.1-0.2%.

2.3 Gas-liquid Chromatography – Mass Spectrometry

The essential oils of *T. parthenium* were analyzed by GLC and GLC-MS using an Agilent 6890 GLC with an Agilent 5973 mass selective detector (EIMS, electron energy = 70 eV, scan range = 45-400 amu, and scan rate = 3.99 scans/s), and a fused silica capillary column (HP 5 ms, 30 m × 0.25 mm) coated with 5% phenyl-polymethylsiloxane (0.25 μm phase thickness). The carrier gas was helium with a flow rate of 1 ml/minute, and the injection temperature was 200 °C. The oven temperature was programmed to initially hold for 10 minutes at 4 °C, then ramp to 200 °C at 3 °C/minute and finally to 220 °C at 2°C/minute. The interface temperature was 280 °C. A 1% w/v solution of each sample in CH₂Cl₂ was prepared, and 1 μl was injected using a splitless injection technique. Identification of the oil components was based on their retention indices determined by reference to a homologous series of *n*-alkanes, and by comparison of their mass spectral fragmentation patterns with those reported in the literature [11], and stored in the MS library [NIST database (G1036A revision D.01.00)/Chem Station data system (G1701CA, version C.00.01.080)]. The percentages of each component are reported as raw percentages (total peak area = 100%) based on total ion current without standardization.

2.4 Bioactivity Assays

Antioxidant activity (via DPPH, ABTS, FTC assays), anti-inflammatory activity and cytotoxicity were analyzed as described earlier [12].

3. Results and Discussion

The aim of the present study was to investigate the phytochemistry and bioactivity of the volatile oil of *T. parthenium* growing wild in Tajikistan. The results obtained by GLC and GLC-MS analysis are presented in Table 1 and Figure 1. Eight components were identified representing 99.8% of total oil composition. The major components were the monoterpenes camphor (69.7-94.0%), camphene (1.7-12.2%), and bornyl acetate (4.2-8.7%). The composition differed between both years; in 2012 plants had a higher content of camphor than in 2013 (Table 1). The present study shows that in Tajikistan feverfew belongs to the camphor/camphene chemotype. Our results are in agreement with those of *T. parthenium* oil from Iran [7, 8], Kosova [4] and Turkey [13].

Table 1: Chemical composition the essential oil from of *Tanacetum parthenium* collected in Tajikistan in 2012 and 2013.

Compound	KI (retention indices)	% of total peak area	
		2012	2013
Camphene	954	1.7	12.2
<i>p</i> -Cymene	1021	-	1.7
Limonene	1029	-	1.6
γ -Terpinene	1059	-	1.3
Camphor	1144	94.0	69.7
Bornyl acetate	1285	4.2	8.7
β -Farnesene	1443	-	2.9
Germaacrene D	1484	-	1.9

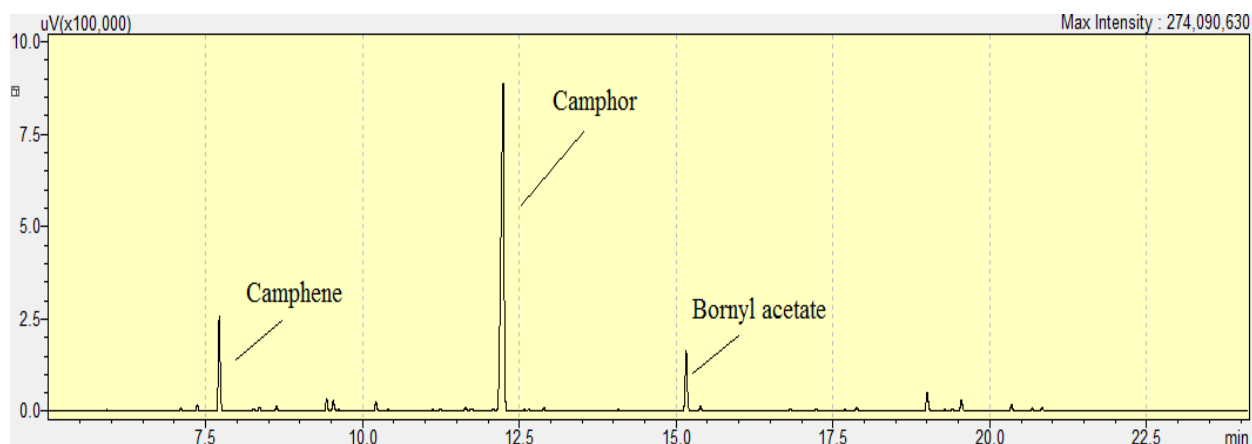


Fig 1: GLC profile of the essential oil from *Tanacetum parthenium*.

The antioxidant activity of the volatile oil was evaluated by using the radical scavenging DPPH and ABTS assays and by the lipid peroxidation test using the ferric thiocyanate method (FTC). Relatively long-lived DPPH[•] and ABTS^{•+} radicals can be scavenged by antioxidants. The volatile oil exhibited weak DPPH and ABTS scavenging activities with an IC₅₀ of 4.82 and 0.96 mg/ml, respectively. As compared to the standards caffeic acid and trolox which had IC₅₀ values of 1.7 and 1.1 μg/ml, respectively. Furthermore, lipid peroxidation was inhibited by 37.1% by the volatile oil (1.125 mg/ml). Lipid peroxidation can lead to loss of membrane function and integrity resulting in cell necrosis and death. Hydroxyl radicals can also react with DNA bases and cause mutations [14]. Some essential oils have anti-inflammatory activity. For example chamomile oil has been used for centuries as an anti-inflammatory agents [15, 16]. In contrast to other essential oils [16, 17], the *T. parthenium* oil inhibits soybean 5-lipoxygenase (5-LOX) weakly. Its IC₅₀ value was 21.6 mg/ml.

The cytotoxicity of the essential oil was investigated against HeLa, CCRF-CEM and CEM/ADR5000 cell lines using the MTT assay. IC₅₀ values were 158.6 μg/ml for HeLa, 69.5 μg/ml for CCRF-CEM, and 83.9 μg/ml CEM/ADR5000 cell lines. Based on these results, the *T. parthenium* oil exhibits a medium cytotoxicity as compared to doxorubicin (a standard chemotherapeutic) (Fig. 2). Monoterpenes are lipophilic and can dissolve in biomembranes, which disturb their fluidity and permeability [15]. It is likely that the cytotoxicity of feverfew oil, which is similar to that of other essential oils [18], is a consequence of such membrane interactions. CCRF-CEM and CEM/ADR5000 differ by the degree of P-gp (an important ABC transporter) expression (being over-expressed in CEM/ADR5000). The higher IC₅₀ value in CEM/ADR5000 suggests that the components of the essential oil can modulate P-gp activity, which would be plausible for lipophilic compounds.

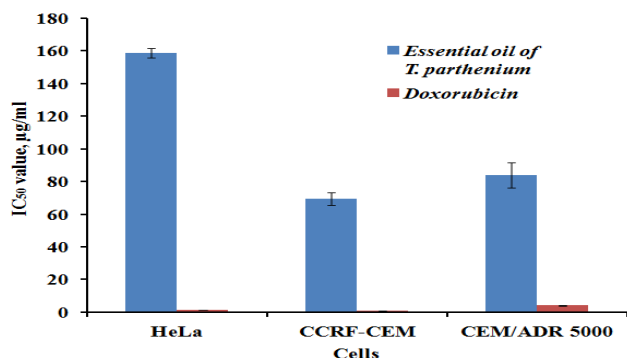


Fig 1: Cytotoxicity: IC₅₀ values (µg/ml) of the essential oil from *Tanacetum parthenium* in HeLa, CCRF-CEM and CEM/ADR 5000 cell lines.

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