Research in Pharmaceutical Sciences, June 2014; 9(2): 155-163
Received: Jan 2013
Accepted: Mar 2013

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***Short Communication***

# Chemical composition and antiprolifrative activity of *Artemisia persica* Boiss. and *Artemisia turcomanica* Gand. essential oils

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## *Abstract*

Essential oils obtained from aerial parts of *Artemisia persica* and *Artemisia turcomanica* were analyzed by GC/MS. While 28 components representing 91.01 % of *A. persica* were identified, the identity of 50 components, constituting 81.93 % of the total oil, was confirmed in *A. turcomanica*. β- thujone was the main compound (75.23%) in *A. persica* while the major identified phytochemicals in *A. turcomanica* were 1,8-cineol (19.23%), camphor (15.55%) and filifolone (15.53%). Both of the essential oils were predominantly made up of monoterpenes. Time- and dose-dependent cytotoxic effects of *A. persica* and *A. turcomanica* on MCF-7 cell line evaluated by MTT assay at 24, 48 and 72 h, showed that the highest cytotoxic effect of *A. persica* and *A. turcomanica* were appeared at 72 h incubation. At that incubation period, $CI_{50}$ of *A. persica* was found to be 0.15 µg/ml, while that of *A. turcomanica* was 0.1 µg/ml. Thus, cytotoxicity of *A. turcomanica* was slightly higher than *A. persica* which could be attributed to the higher content of sesquiterpene present in *A. turcomanica*. As a conclusion, these volatile oils could have chemotherapeutic potentials.

***Keywords:*** *Artemisia persica; Artemisia turcomanica;* Essential oil; Cytotoxicity; MCF-7

## INTRODUCTION

Essential oils as the secondary metabolites of medicinal plants, have indicated lots of biological effects and play a vital role as cytotoxic agents (1-2). *Artemisia* genus belonging to Asteraceae (Compositeae) family which contains 34 species growing in Iran (3), comprises aromatic plants known for their potent chemical constituents in their essential oils with antibacterial (4-5), antiviral (6), antifungal (4-5,7), insecticidal (8-9) and cytotoxic activity (10).

In prior studies, MTT assay, has been carried out on different species of *Artemisia* extracts on different cell lines including human breast carcinoma cell line (MCF-7) (11-12). To the best of our knowledge, cytotoxic activities of essential oils of *Artemisia persica* and *A. turcomanica* against cancer cell lines have not yet been reported. In previous studies, composition of volatile oils of these two species collected from different geographical locations has been reported (13-15).

Due to the effect of harvesting time and season, geographical location, altitude and climate on the yield and composition of volatile oils (16-17), firstly chemical composition of the essential oils of these two plants growing in Razavi Khorasan and North Khorasan provinces of Iran collected in September was determined. Considering both cytotoxic activity of volatile oils of other *Artemisia* species and existence of some cytotoxic components in *A. persica* and *A. turcomanica*, secondly we aimed to evaluate cytotoxic activity of *A. persica* and *A. turcomanica* against MCF-7 cancer cell line

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and to explore the relationship between chemical composition and cytotoxic activity of the essential oils of *A. persica* and *A. turcomanica.*

## MATERIALS AND METHODS

### *Plant material and preparation of essential oils*

Aerial parts of *A. persica* Boiss. and *A. turcomanica* Gand. were collected from Zaveh (Razavi Khorasan province, Iran) and Bojnourd (North Khorasan province, Iran), respectively in September 2010. Identification of these plants was carried out by Dr. Valiollah Mozaffarian (Research Institute of Forest and Rangelands, Tehran, Iran). Voucher specimens (No. 12502 for *A. persica* and 12573 for *A. turcomanica*) have been deposited in the herbarium of Department of Pharmacognosy, Faculty of Pharmacy, Mashhad University of Medical Sciences, Mashhad, Iran.

For the next step, air dried and ground stems and leaves of each species (90 g) were separately subjected to Clevenger-type apparatus by the aid of distilled water (1000 ml) for 4 h. After that the oils were measured and stored in dark glasses at 4 ºC for further studies.

### *GC-Mass analysis*

The essential oils of *A. persica* and *A. turcomanica* were analyzed using a Shimadzu GC/MS-QP5050A gas chromatography-mass spectrometer (GC-MS) fused with a capillary column of silica DB-1 (60 m × 0.25 mm i.d., 0.25 µm film thickness) with ionization potential of 70 ev. The injector temperature and split ratio were adjusted at 240 ºC and 1/31, respectively. The flow rate of nitrogen as a carrier gas was 1.3 ml/min and the oven temperature programming was as follows: temperature was kept at 60 ºC for 2 min then it raised to 260 ºC at a rate of 2 ºC/min. Afterwards it was maintained at 260 ºC for 2 min.

Components of essential oils were identified by comparison of their mass spectra with the NIST NBS54K Library. Kovats indices of components were obtained by the aid of standard n-alkanes (C8–C20) injection, under the same chromatographic conditions.

### MTT assay

*In vitro* cell growth inhibition of *A. persica* and *A. turcomanica* essential oils were evaluated by MTT assay against MCF-7 cell line. Alive cell's mitochondrial succinate dehydrogenase enzyme can reduce yellow colored MTT to a blue product of formazan. This process shows normal operation of mitochondria and cell viability as well (18).

The human breast cancer MCF-7 cell line, obtained from national cell bank of Iran (Pasteur institute, Iran), were cultivated in Roswell Park Memorial Institute (RPMI 1640) medium (Gibco, UK) enriched with 10% fetal bovine serum (FBS).

For prevention of bacterial contamination, 100 µg/ml streptomycin and 100 units/ml penicillin G were added. MCF-7 cells were maintained at 37 °C in a 5% $CO_2$ incubator. When cells reached ~ 90% confluency, they were detached from T-flask by the aid of 0.05% trypsin/EDTA. Cells were seeded in 96-well plate, at a density of 15000 cells/well.

### *Experimental design*

After 24 h, the cultivated cells were treated with several concentrations of *A. persica* and *A. turcomanica* (10, 1, 0.5, 0.2, 0.1, 0.05, 0.01, 0.005 µg/ml) prepared in 1% dimethyl sulfoxide (DMSO) and incubated for 24, 48 and 72 h. Varying concentrations of *A. persica* and *A. turcomanica* essential oils were tested in triplicate. Negative control groups treated with 1% DMSO, as a solubilizing agent, stayed untreated in four wells.

Afterwards, culture medium was changed with 150 µl RPMI-1640 medium plus 50 µl MTT solution (5 mg/ml in phosphate buffer solution). Cells were kept at 37 °C with 95% air, 5% $CO_2$ and complete humidity for 4 h. Then, the MTT solution was replaced with 200 µl DMSO, incubated for 15 min at 37 °C. Finally, the optical density of the wells was measured at 570 nm spectrophotometrically using a plate reader (Sunrise Tecan, Austria). The MTT assays were replicated twice in triplicate. Cytotoxicity index of *A. persica* and

*A. turcomanica* essential oils were calculated as CI%, according to following formula (19-22).

CI%=(1 - (optical-density of sample/optical density of control)) ×100

Cytotoxic potency of *A. persica* and *A. turcomanica* were determined as $CI_{50}$ from the curves of Cytotoxicity Index (CI %), versus different concentrations of *A. persica* and *A. turcomanica* (µg/ml) essential oils, respectively.

## RESULTS

Volatile oils yielded by hydrodistillation of aerial parts of *A. persica* and *A. turcomanica* were 0.62% and 0.78 % (v/w), respectively. Table 1 and 2 demonstrate components of essential oils listed in order of their elution from DB1 column. Phytochemicals were analyzed by GC-MS qualitatively and also quantitatively.

Twenty eight components consisting 91.01 % of total oil of *A. persica* were identified. Among identified constituents, β-thujone was the major and main compound (75.23%). Additionally, percentage of 1, 8-cineol (2.38%), α-thujone (2.84%) and 4-terpineol (2.16%) were more than other identified components. As it is apparent from Table 1, the majority of the components belong to monoterpene groups (88.01%). Moreover sesquiterpens (0.54%) and non- isoprenoid components (1.41%) exist in lower percentages.

**Table1.** GC/Mass data of *A. persica* essential oil.

| NO. | Compound | % Area | RI[a] | Molecular formula | Identification method[b] |
|---|---|---|---|---|---|
| 1 | Cumene | 0.14 | 898.44 | $C_9H_{12}$ | RI + MS |
| 2 | Sabinene | 0.22 | 960.51 | $C_{10}H_{16}$ | RI + MS |
| 3 | δ-4-Carene | 0.24 | 1003.22 | $C_{10}H_{16}$ | RI + MS |
| 4 | β-Cymene | 0.72 | 1005.91 | $C_{10}H_{14}$ | RI + MS |
| 5 | 1,8-Cineol | 2.38 | 1015.04 | $C_{10}H_{18}O$ | RI + MS |
| 6 | γ- Terpinene | 0.58 | 1044.95 | $C_{10}H_{16}$ | RI + MS |
| 7 | Terpinolene | 0.12 | 1076.73 | $C_{10}H_{16}$ | RI + MS |
| 8 | α-Thujone | 2.84 | 1085.26 | $C_{10}H_{16}O$ | RI + MS |
| 9 | β-Thujone | 75.23 | 1101.44 | $C_{10}H_{16}O$ | RI + MS |
| 10 | Thujyl alcohol | 0.68 | 1119.01 | $C_{10}H_{18}O$ | RI + MS |
| 11 | Pinocarvone | 0.24 | 1140.43 | $C_{10}H_{14}O$ | RI + MS |
| 12 | Borneol | 0.48 | 1151.91 | $C_{10}H_{18}O$ | RI + MS |
| 13 | α-Thujenal | 0.74 | 1160.73 | $C_{10}H_{14}O$ | RI + MS |
| 14 | 4-Terpineol | 2.16 | 1165.64 | $C_{10}H_{18}O$ | RI + MS |
| 15 | Myrtenal | 0.21 | 1173.46 | $C_{10}H_{14}O$ | RI + MS |
| 16 | Myrtenol | 0.12 | 1179.41 | $C_{10}H_{16}O$ | RI + MS |
| 17 | α-Pinene | 0.09 | 1195.03 | $C_{10}H_{16}$ | RI + MS |
| 18 | Cuminic aldehyde | 0.96 | 1218.32 | $C_{10}H_{12}O$ | RI + MS |
| 19 | (E)-Solanone | 0.55 | 1250.36 | $C_{13}H_{22}O$ | RI + MS |
| 20 | 2-Carene-10- al | 0.19 | 1265.37 | $C_{10}H_{14}O$ | RI + MS |
| 21 | (E)-3-Carene-2- ol | 0.15 | 1267.33 | $C_{10}H_{14}$ | RI + MS |
| 22 | Bornyl acetate | 0.22 | 1269.86 | $C_{12}H_{20}O_2$ | RI + MS |
| 23 | Isothymol | 0.48 | 1284.30 | $C_{10}H_{14}O$ | RI + MS |
| 24 | Z-Jasmone | 0.22 | 1360.06 | $C_{11}H_{16}O$ | RI + MS |
| 25 | (-)-Elemene | 0.19 | 1387.61 | $C_{15}H_{24}$ | RI + MS |
| 26 | Phenethyl isovalerate | 0.32 | 1457.17 | $C_{13}H_{18}O_2$ | RI + MS |
| 27 | (-)-Spathulenol | 0.34 | 1567.72 | $C_{15}H_{24}O$ | RI + MS |
| 28 | Hexahydrofarnesylacetone | 0.17 | 1831.83 | $C_{18}H_{36}O$ | RI + MS |
| Total compounds | 28 | | | | |
| Oxygenated monoterpenes | 86.94 | | | | |
| Monoterpene hydrocarbons | 2.12 | | | | |
| Oxygenated sesquiterpenes | 0.34 | | | | |
| Sesquiterpene hydrocarbons | 0.20 | | | | |
| Non-terpene hydrocarbons | 0.14 | | | | |
| Others | 1.27 | | | | |
| Total identified | 91.01 | | | | |

[a]Compounds listed in order of elution from a DB-1 column, [b]identification method (RI= Retention indices, MS= Mass spectroscopy.

GC/MS analysis of *A. turcomanica* essential oil afforded 50 phytochemicals representing 82.01% of total oil. As shown in Table 2, the major identified phytochemicals included 1, 8-cineol (19.23%), camphor (15.55%), filifolone (15.54%), brevifolin (6.19%) and *cis*-jasmone (4.31%). Table 2 also indicates that, the majority of components belongs to monoterpenes (68. 66%). Phyto-chemicals present with lower percentage belong to sesquiterpene (2.14%) and non-terpene components (11.13%).Tables 3 and 4 are reported for the sake of comparison of major compounds of *A. persica* and *A. turcomanica* volatile oils growing in different regions and climates. Table 3 exhibits percentages of major constituents of essential oils of Isfahan *A. persica*, Indian *A. persica,* Iranian *A. persica* and *A. persica* studied in the current work.

Table 4 shows percentages of major compositions of essential oils of Khorasan *A. turcomanica* leaves and stems, Iranian *A. turcomanica* leaves and stems and essential oil of *A. turcomanica* investigated in the present study.

**Table 2.** GC-Mass data of *A. torcomanica* essential oil.

| NO. | Compound | % Area | RI[a] | Molecular formula | Identification method[b] |
|---|---|---|---|---|---|
| 1 | Cumene | 0.14 | 899.51 | $C_9H_{12}$ | RI + MS |
| 2 | α-Thujene | 0.09 | 921.26 | $C_{10}H_{16}$ | RI + MS |
| 3 | α-Pinene | 1.17 | 928.75 | $C_{10}H_{16}$ | RI + MS |
| 4 | Propyl benzene | 0.01 | 930.91 | $C_9H_{12}$ | RI + MS |
| 5 | Camphen | 1.20 | 941.12 | $C_{10}H_{16}$ | RI + MS |
| 6 | Sabinene | 0.23 | 961.54 | $C_{10}H_{16}$ | RI + MS |
| 7 | β-pinene | 0.18 | 966.56 | $C_{10}H_{16}$ | RI + MS |
| 8 | Myrcene | 0.66 | 976.58 | $C_{10}H_{16}$ | RI + MS |
| 9 | 1-Decene | 0.08 | 982.03 | $C_{10}H_{20}$ | RI + MS |
| 10 | Cymenene | 0.06 | 993.95 | $C_{10}H_{12}$ | RI + MS |
| 11 | α-Tterpinene | 0.03 | 1004.41 | $C_{10}H_{16}$ | RI + MS |
| 12 | Para-cymene | 2.21 | 1008.22 | $C_{10}H_{14}$ | RI + MS |
| 13 | 1,8-Cineol | 19.23 | 1019.55 | $C_{10}H_{18}O$ | RI + MS |
| 14 | γ-Terpinene | 0.40 | 1046.06 | $C_{10}H_{16}$ | RI + MS |
| 15 | Verbenone | 0.05 | 1063.14 | $C_{10}H_{14}O$ | RI + MS |
| 16 | Terpinolene | 0.11 | 1077.72 | $C_{10}H_{16}$ | RI + MS |
| 17 | α-Thujone | 2.29 | 1086.81 | $C_{10}H_{16}O$ | RI + MS |
| 19 | α-Pinene oxide | 0.22 | 1106.15 | $C_{10}H_{16}O$ | RI + MS |
| 20 | Sabinene | 0.74 | 1110.06 | $C_{10}H_{18}O$ | RI + MS |
| 21 | Camphor | 15.55 | 1126.63 | $C_{10}H_{16}O$ | RI + MS |
| 22 | Pinocarvone | 0.83 | 1142.31 | $C_{10}H_{14}O$ | RI + MS |
| 23 | Borneol | 1.68 | 1154.03 | $C_{10}H_{18}O$ | RI + MS |
| 24 | 4-Terpineol | 2.22 | 1167.52 | $C_{10}H_{18}O$ | RI + MS |
| 25 | Myrtenal | 0.23 | 1174.74 | $C_{10}H_{14}O$ | RI + MS |
| 26 | (+)-α-Terpineol | 1.00 | 1178.03 | $C_{10}H_{18}O$ | RI + MS |
| 27 | Myrtenol | 0.06 | 1180.61 | $C_{10}H_{16}O$ | RI + MS |
| 28 | *cis*-(+)-Carveol | 0.22 | 1204.21 | $C_{10}H_{16}O$ | RI + MS |
| 29 | Nordavanone | 0.21 | 1204.73 | $C_{11}H_{18}O_2$ | RI + MS |
| 30 | Piperitone | 0.14 | 1234.06 | $C_{10}H_{16}O$ | RI + MS |
| 31 | s-(+)-Isopiperitenone | 0.32 | 1247.17 | $C_{10}H_{14}O$ | RI + MS |
| 32 | Thymol | 0.14 | 1269.78 | $C_{10}H_{14}O$ | RI + MS |
| 33 | Cumic alcohol | 0.08 | 1272.42 | $C_{10}H_{14}O$ | RI + MS |
| 34 | Carvacrole | 1.08 | 1277.21 | $C_{10}H_{14}O$ | RI + MS |
| 35 | (+)-Sabinol | 0.35 | 1282.23 | $C_{10}H_{16}O$ | RI + MS |
| 36 | 4-Terpineol acetate | 0.15 | 1284.21 | $C_{12}H_{20}O_2$ | RI + MS |
| 37 | 1,4-Paramenthadien-7- ol | 0.11 | 1313.66 | $C_{10}H_{16}O$ | RI + MS |
| 38 | Eugenol | 0.05 | 1338.07 | $C_{10}H_{12}O_2$ | RI + MS |
| 39 | *cis*-Jasmone | 4.31 | 1362.74 | $C_{11}H_{16}O$ | RI + MS |
| 40 | α-Copaene | 0.11 | 1376.42 | $C_{15}H_{24}$ | RI + MS |
| 41 | (-)-β-Elemene | 0.14 | 1388.29 | $C_{15}H_{24}$ | RI + MS |

**Table 2. (Continued)**

| NO. | Compound | % Area | RI[a] | Molecular formula | Identification method[b] |
|---|---|---|---|---|---|
| 42 | β-Caryophyllene | 0.06 | 1420.98 | $C_{15}H_{24}$ | RI + MS |
| 43 | Neryl acetone | 0.04 | 1426.89 | $C_{13}H_{22}O$ | RI + MS |
| 44 | Davanone | 0.06 | 1450.91 | $C_{15}H_{24}O_2$ | RI + MS |
| 45 | Germacrene- D | 0.11 | 1480.81 | $C_{15}H_{24}$ | RI + MS |
| 46 | δ-Cadinene | 0.08 | 1518.82 | $C_{15}H_{24}$ | RI + MS |
| 47 | (+)-Spathulenole | 0.80 | 1568.72 | $C_{15}H_{24}O$ | RI + MS |
| 48 | Caryophyllene oxide | 0.37 | 1575.03 | $C_{15}H_{24}O$ | RI + MS |
| 49 | Viridiflorol | 0.42 | 1586.01 | $C_{15}H_{26}O$ | RI + MS |
| 50 | Methyl jasmonate | 0.06 | 1589.05 | $C_{13}H_{20}O_3$ | RI + MS |
| 51 | Brevifolin | 6.20 | 1624.74 | $C_{10}H_{12}O_4$ | RI + MS |
| 52 | Hexahydrofarnesyl aceton | 0.15 | 1832.15 | $C_{18}H_{36}O$ | RI + MS |
| Total compounds | 52 | | | | |
| Oxygenated monoterpenes | 62.30% | | | | |
| Monoterpene hydrocarbons | 6.36% | | | | |
| Oxygenated sesquiterpenes | 1.65% | | | | |
| Sesquiterpene hydrocarbons | 0.49% | | | | |
| Non-terpene hydrocarbons | 0.23% | | | | |
| Others | 10.90% | | | | |
| Total identified | 81.93% | | | | |

[a]Compounds listed in order of elution from a DB-1 column, [b]Identification method (RI= Retention indices, MS= Mass spectroscopy.

**Table 3.** Percent of major compounds in different *A. persica* essential oils growing in different areas.

| Compounds | % in Is.A.P. [1] | % in In.A.P.[2] | % in Ir.A.P.[3] | % in A.P.[4] |
|---|---|---|---|---|
| 4-Terpineol | - | - | - | 2.16 |
| α-Thujone | 3.6 | - | - | 2.84 |
| 1, 8-Cineol | - | - | - | 2.38 |
| β-Thujone | - | - | - | 75.23 |
| *trans*-Ascaridol | - | - | 16.1 | - |
| Ethyl 2-nonynoate | - | - | 24.4 | - |
| Davanone | 60.56 | - | - | - |
| *cis*-Chrysanthenyl acetate | 8.65 | - | - | - |
| Limonene | 5.68 | - | - | - |
| α-Pinene | 3.74 | - | - | - |
| Davanone ether isomer | 3.6 | - | - | - |
| α-Thujene | 3.6 | - | - | - |
| Sabinen hydrate acetate | - | 76.74 | - | - |
| *cis*-Ocimenone | - | - | 39.6 | - |
| Ascaridole | - | - | 16 | - |
| α-Terpinene | - | - | 10 | - |
| *cis*-Sabinene hydrate | - | - | 38.8 | - |
| Terpinolene | - | - | 13.3 | - |

[1]Percent of major compounds in essential oil of Isfahan *A. persica*. [2]Percent of major compounds in essential oil of Indian *A. persica.* [3]Percent of major compounds in essential oil of Iranian *A. persica*. [4]Percent of major compounds in essential oil of *A. persica* in current study.

Results of time and dose dependent cytotoxic effects of *A. persica* and *A. turcomanica* essential oils on MCF-7 cell line evaluated by using MTT assay are shown in Figs. 1 and 2 which were extracted from the plots of cytotoxicity percentages versus essential oils concentrations at 24, 48 and 72 h.

As it is evident in Fig. 1, when the results of control group were compared to those of *A. persica* essential oil-treated cells, dose and time dependent cytotoxicity was clearly demonstrated. At concentration of 0.5 µg/ml, cytotoxicity of the essential oil of *A. persica* reached 100% at times greater than 24 h.

**Table 4.** Percent of major compounds in different parts of *A. turcomanica* essential oils growing in different areas.

| Compounds | % in KATL[1] | % in KATS[2] | % in IATL[3] | % in IATS[4] | % in AT[5] |
|---|---|---|---|---|---|
| Linalool | 16.5 | 16.5 | - | - | - |
| *cis*-Chrysanthenyl acetate | 15.3 | 29 | 12 | 29 | - |
| 1, 8-Cineol | 13.4 | 6.9 | - | 6.9 | 19.2 |
| Camphor | 13 | 9 | 36 | 9 | 15.5 |
| Bornyl acetate | 9.2 | 18 | 8 | 18 | - |
| *trans*-Nerodilol | - | 6.2 | - | - | - |
| *cis*-Jasmone | - | - | 4 | - | 4.3 |
| Selin-11-en-4-a-ol | - | - | 8.7 | - | - |
| Brevifolin | - | - | - | - | 6.2 |
| Filifolone | - | - | - | - | 15.5 |

[1]Percent of major compounds in essential oil of khorasan *A. turcomanica* leaves.[2]Percent of major compounds in essential oil of khorasan *A. turcomanica* stems.[3]Percent of major compounds in essential oil of Iranian *A. turcomanica* leaves.[4]Percent of major compounds in essential oil of Iranian *A. turcomanica* stems. [5]Percent of major compounds in essential oil of *A. turcomanica* in current study.

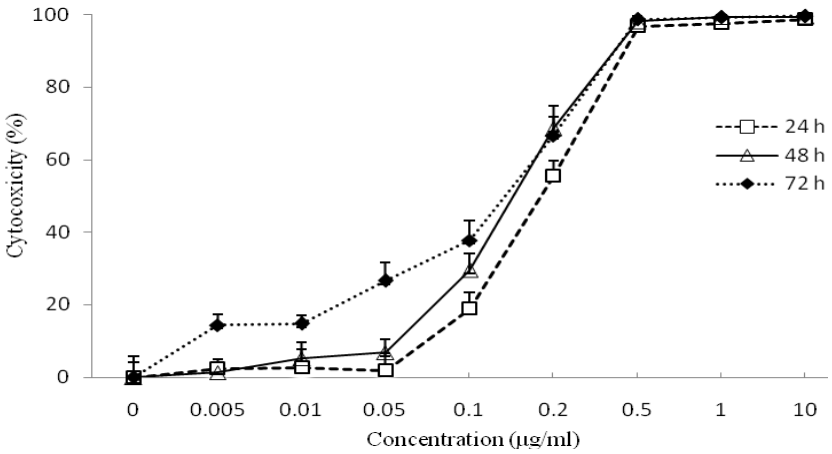


**Fig. 1.** Effect of essential oil of *A. persica* on cell proliferation of MCF-7 cell line displayed as percentage of cytotoxicity index, versus concentration of *A. persica* at 24, 48 and 72 h.

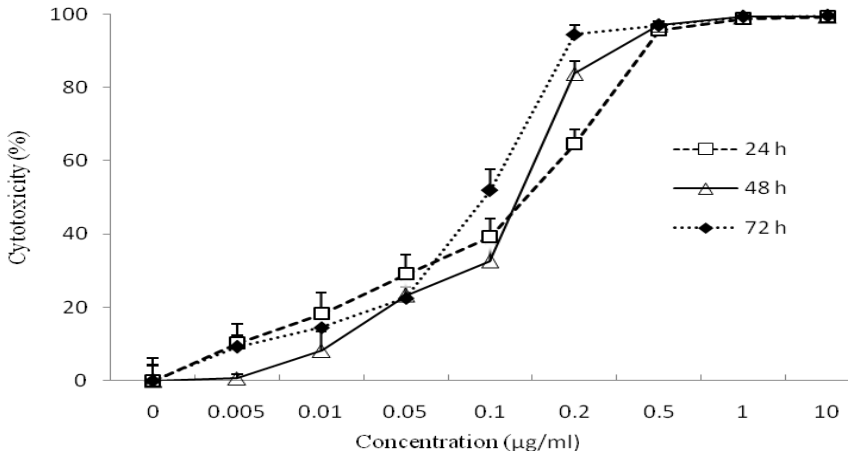


**Fig. 2.** Effect of essential oil of *A. turcomanica* on cell proliferation of MCF-7 cell Line displayed as percentage of cytotoxicity index, versus concentration of *A. turcomanica* at 24, 48 and 72 h.

As it is apparent in Fig. 2, when the results of control group were compared with those of *A. turcomanica* essential oil-treated cells, dose and time dependent cytotoxicity could be clearly observed. Therefore, at 1 µg/ml concentration, cytotoxicity reached to 100% at times higher than 24 h. Thus, *A. persica* and *A. turcomanica* essential oils were significantly cytotoxic on MCF-7 cell line in a dose and time dependent manner.

Highest cytotoxic effect of *A. persica* and *A. turcomanica* essential oils was appeared at 72 h incubation. At that incubation period, $CI_{50}$ of *A. persica* and *A. turcomanica* was 0.15 and 0.1 µg/ml, respectively. Thus, cytotoxicity of *A. turcomanica* was slightly higher than that of *A. persica*.

## DISCUSSION

*Artemisia* species have always been of interest in Iranian traditional medicine as antipyretic**,** wound healer, vermifuge, tonic of stomach, antiflatulent and for their high essence content with pleasant odor and valuable pharmacological effects. Biological activities of *Artemisia* species could be due to secondary metabolites such as monoterpenes, sesquiterpenes, especially sesquiterpene lactones (23-24). For instance, artemisinin, derived from *A. annua*, is a sesquiterpene lactone and considered as a novel antimalarial agent (25). Bisabololoxides as a rare type of sesquiterpenoids and antiplasmodial constituents as well as scopoletin sesquiterpene ethers have been purified from *A. persica* (26-27). Aerial parts of *A. turcomanica* yielded several known mono- and sesquiterpenes (28). Similar to the extracts of *Artemisia* species, wide range of biological effects especially cytotoxic activity (10) which is of great importance as chemotherapeutic agents has been observed in volatile oils of these plants (4-10).

Table 3 shows major constituents of *A. persica* essential oils grown in different regions. As evident in this table, main constituents of essential oils are not in common. β-thujone (75.23%) is the major component of *A. persica* as found in the current study which can be called β-thujone chemotype, while the main constituents of Isfahan *A. persica*, Indian *A .persica* and Iranian *A. persica* are davanone (60.56%), sabinen hydrate acetate (76.74%) and *cis*-ocimenone (39.60%) respectively. Furthermore, other major components presents in *A. persica* investigated in the current study do not exist in Isfahan *A. persica* , Indian *A .persica* and Iranian *A. persica* except α-thujone which showed higher percentage in Isfahan *A. persica* (3.60%) than *A. persica* (2.84%) (15,29-31).

Table 4 demonstrates major composition of *A. turcomanica* essential oils grown in different regions. As seen in this table, Khorasan *A. turcomanica* leaves, Khorasan *A. turcomanica* stems, Iranian *A. turcomanica* leaves, Iranian *A. turcomanica* stems and *A. turcomanica* assessed in the current study are in common by containing camphor. All of the volatile oils in table 4 contain 1,8 cineol except Iranian *A. turcomanica* leaves. in the percentage of this substance in *A. turcomanica*is more than other essential oils constituents, so it can be called 1,8 cineol chemotype (13,32).

According to previous studies, different factors such as harvesting time and season, geographical location, altitude, climate and other factors including geno type, reproductive stage, cutting height, drying condition could extremely affect the yield and composition of volatile oils of the same species (33-34). In this study, the effect of geographical location, climate and month of collection are shown on the phytochemical composition of *A. persica* and *A. turcomanica* essential oils

Cytotoxicity of *A. persica* and *A. turcomanica* were evaluated by MTT assay, a simple and reliable experiment which measures cell viability and cytotoxicity for screening cytotoxic agents (35). Results of this test could be a basis for finding further chemotherapeutic agents. Findings of this experiment demonstrated that *A. persica* and *A. turcomanica* were strongly cytotoxic on MCF-7 cell line in a dose and time dependent manner. Chemical composition of *A. persica*, indicated that its cytotoxic effect could be attributed in part to the high content of β-thujone (75.23%) and low content of α-thujone (more toxic than β-stereoisomer). Previous

studies have reported cytotoxic effect of thujone on A375 , monkey's kidney cell lines and its anticancer potentials (36-39). Cytotoxicity of *A. turcomanica,* to some extent, could be attributed to the high content of 1, 8-cineol (19.23%) and camphor (15.55%), which are toxic constituents as previously reported (40). Furthermore, it has been indicated that fractions with higher content of sesquiterpenes show anticancer activity (41), so higher toxicity of *A. turcomanica* could be somehow explained by its higher sesquiterpene content (2.14%) compared to *A. persica* (0.53%). Comparison of $CI_{50}$ of *A. persica* and *A. turcomanica* with $CI_{50}$ of extracts and essential oils of some species of *Artemisia* and Asteraceae family against MCF-7 cells is very promising and illustrates higher anticancer capacity of *A. persica* and *A. turcomanica* against breast cancer carcinoma cell line (11,42-45).

## CONCLUSION

It can be concluded that the volatile oils of *A. persica* and *A. turcomanica* could have chemotherapeutic potentials. Further investigations are needed to isolate cytotoxic constituents from the essential oils of these two plants and clarify their molecular mechanisms as well as understanding the effects of *A. persica* and *A. turcomanica* on other cell lines.

## ACKNOWLEDGMENTS?

We are thankful to Dr. Mahdi Mojarab and Dr. Solmaz Esnaashari for their kind cooperation with this work.

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