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# Study of Essential Oil Compounds from Three Iranian Artemisia Species and the Implications on Livestock Grazing Behavior

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**ABSTRACT**

**Aims** One of the herbal indices that effects on the livestock grazing behavior is the herbal compounds including the essential oils, so the aim of the present study is the comparison of three Artemisia species in terms of the composition of essential oils coincident and the relationship between livestock grazing time from these three plant species.

**Materials and Methods** Three Artemisia species (Artemisia sieberi Bess., Artemisia kermanensis Podl., and Artemisia khorassanica podl.) in the natural from habitats with similar conditions were studied. Five plant stands were selected randomly from each species and the essential oils were obtained by SDE of air-dried samples and the essential oil were analyzed by GC-MS.

**Findings** About 88% of essential oil compounds in these three Artemisia species were common and Artemisia kermanensis Podl. had more essential oil percentage (98.47%) and Artemisia sieberi had the lowest essential oil percentage (73.21%) while the rate of grazing on the Artemisia sieberi Bess. was higher compared to two other species in the same vegetative stage. Also, 6% of the compounds were observed only in the Artemisia kermanensis species, which is probably the presence of these compounds are the reason for the lack of feeding by the livestock of this species.

**Conclusion** The selection of livestock for grazing of these three species to be highly relevant to their essential oil compounds, although the amount of these compounds has decreased in the third phenological stage, preventing livestock feeding on these species in the first and second stage of vegetation. Livestock grazing planning based on essential oil combinations can be a new horizontal in rangeland management. It seems the biochemical defense in the plants with essential oil is a way to combat animal grazing.

**Keywords** Artemisia; Natural Products; GC-MS; Biochemical Defense; Livestock Grazing

**CITATION LINKS**

[1] Medicinal ...[2] Chemical composition and antimicrobial activity ... [3] Chemical composition and insecticidal activity ... [4] A generalization of the retention index system including linear ... [5] Quantitative and qualitative comparison of essential oils compounds ... [6] Antioxidant, anticholinesterase and antimicrobial constituents ... [7] Screening of chemical composition and antifungal and antioxidant activities of ... [8] The genus Ferulla: A comprehensive ... [9] Chemical control of stored product insects with fumigants ... [10] Biochemistry of plant secondary metabolites ... [11] Anticonvulsant activity and chemical composition of Artemisia ... [12] Screening of chemical composition, antimicrobial and antioxidant activities ... [13] Flora ... [14] Selenium Bioaccumulation and Bio-concentration Factors in some Plant Species ... [15] Chia seeds as a source of natural lipid ... [16] Identification of essential oil components by gas ... [17] Ethanol extract from Artemisia vestita, a traditional Tibetan medicine, exerts anti-sepsis action through down ... [18] Mining the essential oils of the ... [19] Composition and antimicrobial activity of the essential oil of ... [20] Effect of condensed tannins upon the performance of lambs grazing Lotus ... [21] Biologically active substances from the genus ... [22] Volatile constituents of Artemisia ... [23] Effects of monensin and increasing dose levels of a mixture of essential oil compounds ... [24] Determination of the chemical composition and antioxidant activity of the essential ... [25] Detection of sesqui terpenelactones in ten Artemisia species ... [26] The composition of the essential oil of Stachys iberica subsp stenostachya ... [27] Comparison of the volatile composition of Stachys persica ... [28] Medicinal and aromatic plants ... [29] Chemical composition of the essential oil from two wormwood species Artemisia frigida and Artemisia... [30] Production and Processing of Medicinal... [31] Effect of grazing intensity on soil moisture and vegetation ... [32] Threat of Copper, Zinc, Lead, and Cadmium in Alfalfa

## Introduction

Artemisia is one of the most important rangeland species in Iran which consist of a large part of Iran's vegetation. This plant genus, with the Astragalus, has more than 60% of Iran's natural vegetation. Artemisia L. is a genus of small herbs and shrubs that found in northern temperate regions, and belongs to the Compositae (Asteraceae) family, and it is one of the most numerous plant groupings, which comprises about 1,000 genera and over 20,000 species. In this family, Artemisia in the Anthemideae tribe and comprises over the 500 species, which are mainly found in Asia, Europe and North America [1]. A large number of members of the Anthemideae tribe are important as ornamental crops, and medicinal and aromatic plants. Some species of this family produce essential oils used in folk and modern medicine, and in the cosmetics and pharmaceutical industry [2, 3]. The genus of Artemisia L. (Commonly known as wormwood), one of the largest and most widely distributed genus of the Asteraceae family, includes perennial and annual herbs or small shrubs [4, 5].

This plant genus is particularly important because many Artemisia species have botanical and pharmaceutical properties, characterized scents, and tastes due to the content of monoterpenes and sesquiterpenes. These plants have folk and conventional medicine applications [6, 7]. The major classes of phytoconstituents of Artemisia species are terpenoids, flavonoids, coumarins, caffeoylquinic acids, and sterols.

Bora and Sharma reported that this genus is an important source of biological compounds used in insecticides, antimalarials, cytotoxins, antihepatotoxic, fungicides, antibacterials, and allelochemicals [8, 9]. Also, the important drug found from this genus is artemisinin that is the antimalarial drug isolated from *A. annua* [10, 8]. Other species of Artemisia have also been noted for their potential use at in-depth investigations on biological activities, especially those species that affect the central nervous and cardiovascular systems [3, 11].

This genus comprises more than 400 species and is predominantly distributed in the northern temperate region of the world in the 0-50cm precipitation area. Thirty-four species have been reported in Iran and among which two are endemic: *A. melanolepis* Boiss. and *A. kermanensis* Podl [12, 13].

*A. herba-alba* which is abundant in the central regions of Iran (Kashan city), the most frequent constituents in this species were 1,8-cineole=18.30% and camphor=42.50%; in another evaluation done in the northwest of Iran (West Azerbaijan province), the most frequent constituents were camphor=34.94% and beta-thujone=35.66% [14-16].

*A. kermanensis* mostly observed in southeastern of Iran (Kerman province), the most frequent constituents in this species were: 1,8-cineole=26.93% and camphor=16.97% [17].

*A. khorasanica* has a remarkable area in the northeast of Iran, the different constituents were found in this species; however, among them were some shared components. Those components consist of 1,8-cineole (17.75% in one experiment and 33.90% in another one), camphor (13.90% in one experiment and 12.60% in another one), alpha-thujone (43.40% in one and 11.90% in another one), beta-thujone (16.20% in one and 20.10% in another one) and davanone (12.20% in one and 36.40% in another one) [18, 19].

### Objective

Since the livestock feed of these three Artemisia species at the end of the phenological stage (Seedling) exclusively, so the compounds in the essential oils in these three species are a serious obstacle to the livestock grazing at the time of two previous vegetative stages. Therefore, this study aimed to evaluate the essential oils of these three species in the third vegetative stage to determine the contribution of the compounds in the essential oil and their effects on cattle behavior.

## Materials and Methods

**Livestock behavior:** In this research, the time method (Using a stopwatch) was used to determine the livestock behavior and the average time allocated for grazing of these three species was cited in the grazing season.

**Plant material:** The plant materials obtained from three Artemisia species (*Artemisia sieberi* Bess., *Artemisia kermanensis* Podl., and *Artemisia khorassanica* Podl.) from natural habitats with similar climatologically conditions in Iran. These three Artemisia species were taken from steppe rangelands in Natural habitats of Kerman province with 58° 10′ to 59° 13′ eastern longitudes and 33° 28′ to 43° 41′ northern latitudes with an area of 3700 hectares. The dominant species in this area is *Artemisia sieberi* Bess. and the average rainfall is 115mm, also, has an irregular distribution. According to the Doumarten methodology, the climate of this region is dry. In this study, the five plant stands were selected randomly from each species at the end of the phenological stage (Seedling), then the plant material dried at room temperature and shadow and they prepared for essential oil operation.

**Isolation of the essential oil:**
The essential oil of air-dried samples (100g) of each species was isolated by hydrodistillation for 1h, using a Simultaneous Distillation Extraction (SDE). The air-dried aerial parts of the plant were powdered and the volatile fraction was prepared by a modified Likens-Nickerson's SDE method [12]. SDE device conducted the extraction of essential oils with an organic solvent such as pentane. A microscale simultaneous distillation extraction apparatus (Ashke Shishe; Tehran; Iran) was used. The dried powdered plant was homogenized with distilled water and the homogenate subjected to SDE apparatus for 1h using pentane (Chromatography grade reagent; Merck; German) as a solvent and then extract was concentrated with nitrogen.

**Gas Chromatography (GC) analysis:**
GC analysis was performed by using a Thermoquest gas chromatograph with a Flame Ionization Detector (FID). The analysis was carried out using fused silica capillary Innowax column (60m×0.25mm i.d.; film thickness=0.25μm). The operating conditions were as follows: Injector and detector temperatures were 230˚C and 250˚C, respectively. Nitrogen was used as carrier gas at a flow rate of 4ml/min; oven temperature program, 80˚C-230˚C at the rate of 3˚C/min, and finally held isothermally for 10min. GC-MS analysis GC-MS analysis was performed by using Thermoquest-Finnigan gas chromatograph equipped with an above-mentioned column and coupled to a TRACE mass quadrupole detector. Helium was used as carrier gas with the ionization voltage of 70ev. Ion source and interface temperatures were 200˚C and 250˚C, respectively. Mass range was from m/z 43-456. Gas chromatographic conditions were as given for GC.

**Identification of Compounds:**
The constituents of the essential oil were identified by calculation of their retention indices under temperature-programmed conditions for n-alkanes (C5-C10) and the oil on an Innowax column under the same chromatographic conditions. Identification of individual compounds was made by comparison of their mass spectra with those of the internal reference mass spectra library or with authentic compounds and confirmed by comparison of their retention indices with authentic compounds or with those of reported in the literature [19, 20]. For quantification purpose, relative area percentages obtained by FID were used without the use of correction factors.

## Findings
The results of essential oils analysis showed that the percentage of essential oil and the number of essential oil compounds were different in these three Artemisia species (Table 1), also the results of this research indicated that almost 88% of the essential oil composition of these three species was common, and 5 compounds were observed only in the *Artemisia khorassanica* Podl. species essential oil, while 3 of composition was observed only in the *Artemisia kermanensis* Podl. species essential oil (Diagram 1, Table 2), In the other hand, the tendency of livestock to feed on these three species showed that usually, the livestock feed *Artemisia sieberi* Bess. species more than two other species and doesn't show much interest for grazing from the *Artemisia kermanensis* Podl. species, of course, when the livestock have a normal physiological and metabolic conditions and haven't been hungry for a long time (Diagram 2) [21, 22]. It seems the selection of livestock from these three species to be highly relevant to their essential oil compounds, although the amount of these compounds has decreased in the third phenological stage, prevent livestock feeding on these species in the first and second stage of vegetation.

**Table 1)** Percentage of essential oil compositions of three Artemisia species

| N | Compound | RI | *A. khorassanica* | *A. kermansis* | *A. sieberi* |
|---|---|---|---|---|---|
| 1 | 7-methyl-1-octene | 860 | 2.8 | 1.9 | 3.5 |
| 2 | 2,5-divinyl-2-methyl-tetrahydrofuran | 909 | 6.9 | 6.3 | 5.6 |
| 3 | 4,4-dimethyl but-2-enolide | 919 | 1.9 | 1.2 | 2.3 |
| 4 | Tricyclene | 925 | 1.8 | 1.9 | 2.5 |
| 5 | alpha-thujene | 929 | 10.3 | 7.3 | 6.2 |
| 6 | alpha-pinene | 937 | 5.8 | 5.4 | 3.0 |
| 7 | 6-methyl-5-octene-2-one | 940 | 3.1 | 3.0 | 0.2 |
| 8 | Camphene | 951 | 16.9 | 14.3 | 11.2 |
| 9 | Sabinene | 969 | 1.3 | 1.1 | 4.9 |
| 10 | beta-pinene | 976 | 5.3 | 4.9 | 2.4 |
| 11 | Myrcene | 980 | 2.8 | 2.4 | 1.9 |
| 12 | alpha-phelandrene | 999 | 6.9 | 5.7 | 11.6 |
| 13 | alpha-terpinene | 1011 | 11.6 | 8.6 | 6.2 |
| 14 | 3-none-2-one | 1000 | 11.6 | 9.8 | 6.7 |
| 15 | 4-methy-4-vinylbutyrolactone | 1004 | 2.8 | 2.5 | 1.4 |
| 16 | p-cymene | 1014 | 6.9 | - | - |
| 17 | 1,8cineol | 1023 | 1.9 | 1.4 | 9.3 |
| 18 | lialic alchol | 1028 | 11.6 | 8.5 | 5.6 |
| 19 | gama-terpinene | 1049 | - | 9.3 | - |
| 20 | cis-sabinene hydrate | 1055 | - | 5.6 | - |
| 21 | Verbenol | 1076 | 3.1 | 2.7 | 2.5 |
| 22 | Linalool | 1081 | 2.5 | - | - |
| 23 | trans-sabinene hydrate | 1085 | 3.1 | 2.7 | 0.2 |
| 24 | alpha-thujone | 1100 | 5.5 | 5.5 | 17.7 |
| 25 | Cis-p-menth-2-en-1-ol | 1108 | 26.6 | 25.4 | 25.8 |
| 26 | 1-terpineol | 1109 | - | 3.1 | - |
| 27 | Ipsdienol | 1125 | 2.1 | 5.5 | 5.5 |
| 28 | Camphor | 1126 | 5.3 | 28 | 28 |
| 31 | Borneol | 1152 | 2.8 | - | - |
| 32 | Menthol | 1156 | 6.9 | 2.1 | 2.1 |
| 33 | p-cymene-8-ol | 1158 | 1.9 | 3.5 | 3.5 |
| 34 | terpin-4-ol | 1161 | 11.6 | 9.8 | 9.8 |
| 35 | fenchyl alchol | 1169 | 3.1 | 4.6 | 4.6 |
| 36 | p-menth-2-en-8-ol | 1171 | 5.5 | 5.8 | 5.8 |
| 37 | Nordavanone | 1199 | 2.2 | 2.3 | 1.7 |
| 38 | Citronellol | 1201 | 4.3 | - | - |
| 39 | Neral | 1210 | 4.9 | 5.8 | 5.8 |
| 40 | Carvone | 1214 | - | 3.1 | 3.1 |
| 41 | 4-tepinyl acetate | 1216 | 2.1 | 2.9 | 2.9 |
| 42 | Pipertone | 1228 | 5.3 | 4.7 | 4.7 |
| 43 | linalyl acetate | 1229 | 3.1 | 3.9 | 3.9 |
| 44 | Geranial | 1237 | 5.5 | 5.6 | 5.6 |
| 45 | Decanol | 1245 | 6.8 | - | - |
| | Others | - | 7.8 | 6.9 | 5.9 |
| | Total | - | 98.47 | 85.34 | 73.21 |

**Table 2)** The essential oil compositions that only abserved in *A. khorassanica* Podl and *A. kermanesis* Podl species

| *A. khorassanica* | *A. kermanesis* | *A. sieberi* | **Compound** (%) |
|---|---|---|---|
| p-cymene | NA | *NA* | 6.9 |
| Linalool | NA | *NA* | 2.5 |
| Citronellol | NA | *NA* | 4.3 |
| Decanol | NA | *NA* | 6.8 |
| Borneol | NA | *NA* | 2.8 |
| NA | gama-terpinene | *NA* | 9.3 |
| NA | cis-sabinene hydrate | *NA* | 5.6 |
| NA | 1-terpineol | *NA* | 3.1 |

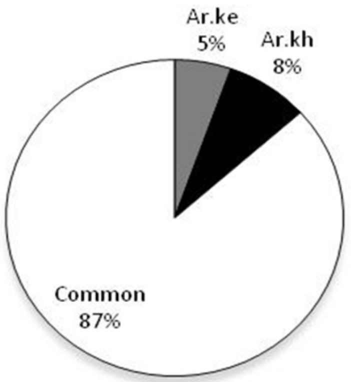


**Diagram 1)** Distribution essential oil composition in three A. species

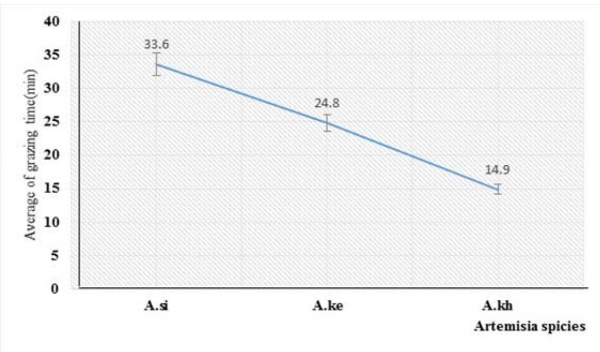


**Diagram 2)** Average grazing time of the livestock from the three Artemisia species during a grazing

## Discussion and Conclusion

According to a study carried out by Benchaar *et al.*, [23] α-pinene, α-tujune, and camphene were the major constituents of Artemisia essential oil which are natives of Iran, Which is consistent with the results of this research. Also, Dehghani Bidgoli [5] studied the effect of different grazing intensities on the essential oils of *Artemisia sieberi* that their results showed that the different grazing intensities effect on essential oil composition in this species.

To the best of our knowledge, there are many reports on the chemical composition of the essential oils isolated from the plants belonging to the genus Artemisia [11, 21, 22]. Most of these reports indicate that α-tujune and α-Pinene are the main and/or characteristic constituents of Artemisia essential oils, which are indicated in the present study. These results might have arisen from several differences in climatically, seasonal, geographical, and environmental factors as mentioned by Kordali *et al.* [24].

The percentage and retention indices of the essential oil components obtained from aerial parts of Artemisia species are listed in Table 1. As shown in Table 1, the analysis essential oil from the aerial parts of A. species indicated the 45 constituents, representing 73.21%, 85.34%, and 98.47% of the (*Artemisia sieberi* Bess., *Artemisia kermanensis* Podl., and *Artemisia khorassanica* Podl.) essential oil respectively [25-27].

The main components of these three Artemisia species essential oil were alpha-terpinen, alpha-thujene, and camphen (Diagram 3). The aerial parts of (*Artemisia sieberi* Bess. *Artemisia kermanensis* Podl., and *Artemisia khorassanica* Podl.) essential oil were found to be rich in regards to oxygenated monoterpenes (68.2%, 75.5%, and 80.3%) [28, 29].

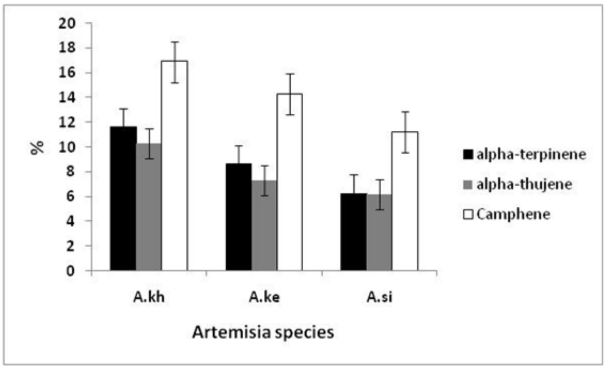


**Diagram 3)** The percentage of main essential oil components in three A. species

Of course, the percentage of compounds, 1,8-Cineol, Camphon, Chrysanthenone, Alpha-Painan, Sabine, Para-cymene, cis-chrysanthenone, 4-terpinene, had not significant differences in the three Artemisia species essential oil [30-32].

Livestock grazing planning based on essential oil combinations can be a new horizontal in rangeland management. Although the amount of essential oil compounds in the third phenological stage has in the three species of Artemisia decreased, the judging about of grazing livestock from these plant species needs further research, in order to know exactly which compounds cause the herd to not feed these species. Perhaps biochemical defense in the plants with essential oil is a way to combat animal grazing.

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