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EVALUATION OF CHEMICAL COMPOSITION OF ESSENTIAL OIL AND TOXIC METAL ACCUMULATION OF TARRAGON (*ARTEMISIA DRACUNCULUS* L.) CULTIVATED ON METAL-CONTAMINATED SOILS

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Abstract

This study evaluates the quality of tarragon essential oil, toxic metals content, and the growth potential of tarragon on heavy metal-contaminated soils. Tarragon demonstrated tolerance to toxic metals and can be cultivated on highly polluted soils. The levels of toxic metals in tarragon essential oil were below the Maximum Permissible Concentrations. Based on translocation and bioconcentration coefficients, tarragon is a Cd, Pb, Zn and Hg excluder and suitable for phytostabilization. Tarragon essential oil exhibits a mixed sabinene/elemicin/isoelemicin/methyleugenol chemotype and could be used in the pharmaceutical, cosmetic, and related industries.

Key words*: essential oil, phytoremediation, polluted soils, tarragon, toxic metals.*

INTRODUCTION

Artemisia dracunculus L., commonly known as tarragon, is a perennial plant belonging to the Asteraceae family. Tarragon is widespread across North America, Europe, Southeastern Russia, Central Asia, Turkey, and Mongolia (Aglarova et al., 2008; Fildan et al., 2019; Sharopov et al., 2020). It is cultivated in Europe, Russia, Asia, Africa, and North America and is widely used in food flavoring, medicine, and perfumery (Obolskiy et al., 2011; Fildan et al., 2019; Sharopov et al., 2020).

Tarragon is a small perennial shrub that grows 120-150 cm tall. Its alternate leaves measure 2-6 cm in length and 1-8 mm in width, often covered with fine hairs and featuring smooth or slightly toothed edges. The plant produces small yellow flowers with whitish petals, arranged in globose, inclined baskets that form broom-shaped racemes. The entire herb is green and mossy, with a pleasant aroma and taste, and flowers in the second half of summer. Tarragon thrives in various environments but produces the highest yields in moist, sandy loam soils with an alkaline reaction. Depending on the variety, it can be propagated by seed (Russian tarragon) or rhizome cuttings (French and German tarragon). In Europe, plantings are established in April, with cuttings placed 60 cm apart and covered with a thin layer of soil. The first harvest occurs in the same year, and in subsequent years, up to three harvests per season are possible. The harvested material is dried using natural air circulation or heated to 35°C before the leaves are separated from the stems (Aglarova et al., 2008; Obolskiy et al., 2011).

Tarragon essential oil, a pale yellow to amber liquid with a spicy scent, is extracted from the plant's aerial parts during flowering (Fildan et al., 2019). Traditionally, it has been valued for a wide range of therapeutic properties, including antioxidant, antimicrobial, antidiabetic, and anti-inflammatory effects (Obolskiy et al., 2011; Fildan et al., 2019; Sharopov et al., 2020).

Certain medicinal plants, such as mint, St. John's wort, and sage, are known to accumulate significant amounts of toxic heavy metals in their tissues. These plants can be used for phytoremediation, serving as an alternative to food crops grown in contaminated conditions. The concentrations of plant by-products depend on growing conditions, influencing metabolic pathways responsible for the synthesis of natural compounds (Akula & Ravishankar, 2011). Heavy metals can significantly alter the chemical composition of secondary metabolites in aerial plant parts, affecting the quality, safety, and efficacy of plant-derived products.

Although the composition of tarragon essential oil is well studied, limited information is available on toxic metal accumulation in tarragon's aboveground biomass and essential oil when grown on contaminated soils.
This study aims to conduct a comparative analysis to determine heavy metal accumulation, macro- and trace element deposition in the vegetative organs of tarragon, and the quality of its essential oil. Additionally, this study explores the feasibility of cultivating tarragon on heavy metal-contaminated soils.

MATERIALS AND METHODS

This experiment was conducted on an agricultural field contaminated with Zn, Pb, and Cd, located 0.5 km from the Non-Ferrous Metals Plant (NFMW) near Plovdiv, Bulgaria.
Tarragon seedlings were purchased and planted in the spring at sites 0.5 km from the pollution source. In the second and third years after planting, plants were collected and analyzed. Tarragon was hand-harvested at the flowering stage in late August. After transport to the laboratory, plants were separated into individual organs (roots, stems, leaves, and flowering tops) using scissors. Samples were air-dried at room temperature until reaching a stable dry mass, followed by further drying at 35ºC. The concentrations of metals in different plant parts - roots, aboveground mass (stems and leaves), and flowering tops - were determined.
Tarragon essential oil was extracted under laboratory conditions via steam distillation for 2 hours using a Clevenger-type apparatus.
The pseudo-total metal content of soils was determined following ISO 11466. The microwave mineralization method was used to measure metal concentrations in tarragon and its essential oil. Quantitative measurements were performed using ICP (Jobin Yvon Emission - JY 38 S, France). Hg content was analyzed without sample pretreatment using a mercury analyzer. The accuracy and performance of the ICP and mercury analyzer were validated using standard reference material from apple leaves (SRM 1515, National Institute of Standards and Technology, NIST).
The chemical composition of the essential oil (diluted in hexane, 1:1000) was analyzed using an Agilent 7890A gas chromatography (GC) system equipped with an FID detector and an Agilent 5975C mass spectrometer. Compounds were identified by comparing retention times and Kovats retention indices (RI) with standard substances and mass spectral data from the NIST'08 library (National Institute of Standards and Technology, USA).

RESULTS AND DISCUSSIONS

Soil Characteristics

The soils in the study area have a neutral to slightly alkaline reaction (pH = 7.5), with moderate organic matter content (2.1%) and average nutrient levels (P, K). The total Zn, Pb, and Cd concentrations are significantly elevated, measuring 2423.9 mg/kg Zn, 2509.1 mg/kg Pb, and 63.7 mg/kg Cd, far exceeding the maximum permissible concentrations (MPC) of 400 mg/kg Zn, 100 mg/kg Pb, and 3.0 mg/kg Cd (Table 1).

Table 1. The total content of Pb, Zn, Cd (mg/kg) and Hg (µg/kg) in soils

Element	Pb	Zn	Cd	Hg
Total, x±sd	2509.1±6.5	2423.9±4.5	63.7±1.8	488.0±13
MPC	100	400	3.0	1.5

x = mean value (mg/kg) from five repetitions; sd = standard deviation

Metal content in tarragon

Table 2 presents the results obtained for the toxic metals and nutrient contents in the organs of the essential oil crop studied. The Pb content in tarragon roots reached 141.8 mg/kg, while Zn, Cd, Hg, Cu, Fe, and Mn levels were 50.9 mg/kg, 3.9 mg/kg, 48.3 µg/kg, 7.1 mg/kg, 157.8 mg/kg, and 5.6 mg/kg, respectively. These results can be attributed to tarragon's anatomical and biological characteristics. The plant's rhizomatous root system consists of thick, lignified, serpentine rhizomes with a dense network of thin, branched roots extending 30-45 cm deep. This structure enhances nutrient and water absorption, allowing tarragon to thrive in various soil conditions.
The movement and accumulation of toxic metals in tarragon's vegetative organs varied significantly. Notably, Pb was highly accumulated in the aerial parts, reaching 1144.1 mg/kg in leaves. Typically, plants

absorb only a small fraction of Pb from soil, with most Pb retained in the roots. However, findings suggest that tarragon efficiently translocates Pb to aboveground tissues, likely due to its anatomical and morphological features. Leaf hairs may contribute to Pb fixation from airborne pollutants.

Cadmium, known for its high mobility in plants, was present at 39.0 mg/kg in tarragon leaves, exceeding the toxic threshold of 5.0 mg/kg (Kabata-Pendias, 2001). Similarly, Hg accumulation in tarragon was attributed primarily to aerosol deposition, as leaves are the primary uptake site, with soil absorption being secondary. Hg concentrations were higher in the aerial parts and flowering tops than in the roots.

The Zn content in tarragon leaves reached 556.1 mg/kg, below the phytotoxic threshold of 500-1500 mg/kg (Kabata-Pendias, 2001). The Cu concentration in leaves was 74.3 mg/kg, exceeding the toxic range for most plants (25-40 mg/kg). Fe accumulated mainly in leaves (396.3 mg/kg), surpassing normal plant values (50-250 mg/kg), while Mn levels (56.5 mg/kg) remained well below phytotoxic thresholds (400-2000 mg/kg).

Most macroelements accumulated in tarragon's aerial parts. In leaves, P, K, Mg, and Ca levels reached 1590.1 mg/kg, 9843.0 mg/kg, 540.4 mg/kg, and 2387.0 mg/kg, respectively. In flowering tops, these concentrations were significantly higher: 5062.1 mg/kg P, 24,233.6 mg/kg K, 2536.6 mg/kg Mg, and 16,915.4 mg/kg Ca.

Despite accumulating heavy metals at levels exceeding critical toxicity thresholds, tarragon exhibited no visible symptoms of heavy metal toxicity, indicating a high level of tolerance. Heavy metal and nutrient accumulation was significantly higher in flowering tops than in leaves, except for Pb and Zn. In flowering tops, Pb reached 932.8 mg/kg, Zn 304.8 mg/kg, Cd 35.1 mg/kg, and Hg 78.2 µg/kg.

Overall, metal distribution in tarragon was selective, primarily depending on the plant organ and its surface characteristics. Heavy metals and micro- and macroelements were predominantly accumulated in leaves and flowering tops (Figure 1).

Table 2. Content of toxic metals and nutrients (mg/kg) in tarragon

	Roots	Stems	Leaves	Flowering tops
Pb	148.8±1.2	45.3±0.3	1084.7±2.6	932.8±2.0
Cd	3.9±0.1	15.5±0.2	39.0±0,,5	35.1±0.5
Zn	50.9±1.3	68.5±1.5	556.1±8.7	304.6±4.6
Cu	7.1±0.8	13.3±1.0	74.3±3.5	79.5±3.8
Fe	57.8±0.8	166.9±3.4	396.3±4.6	4790.1±8.9
Mn	5.6±'0.5	16.0±1.0	56.5±1.8	232.2±2.6
P	493.0±5.5	675.8±5.9	1590.1±9.8	5062.1±12.7
Ca	1420.4±8.8	2387.0±10.8	16915.3±25.0	19226.8±26.0
Mg	216.7±1.9	540.4±2.8	2536.6±10.6	4616.8±12.7
K	5751.5±8.7	9843.0±9.8	24233.6±23.0	36562.5±26.9
Hg*	48.3±3.8	45.0±4.5	284.7±10.0	78.2±7.1

x - average value (mg/kg) from 5 repetitions; sd - mean standard deviation; *- µg/kg.

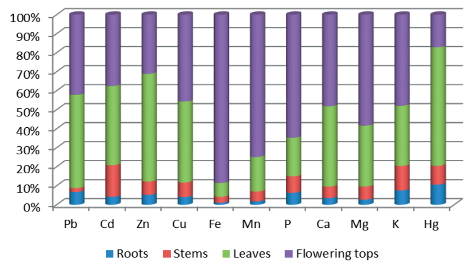

Figure 1. Distribution of toxic metals and nutrients in tarragon

Toxic Metal Content in Tarragon Essential Oil

The toxic metal content of tarragon essential oil was also analyzed. The results indicate that most metals present in flowering tops do not transfer into the oil during distillation. Consequently, their concentrations in the oil are significantly lower. The Pb content in tarragon essential oil reached 0.27 mg/kg, and Zn 1.09 mg/kg, while Cd and Hg levels were below the detection limits of the analytical method.

These findings strongly suggest that a substantial portion of metals present in tarragon flowering tops does not pass into the essential oil. Furthermore, the levels in the oil remain below the accepted maximum limits for an environmentally safe product (5 mg/kg Pb, 1 mg/kg Cd, 0.1 mg/kg Hg) as per the Council of Europe (2021).

These results confirm previous findings by Angelova et al. (2015), which demonstrated that the heavy metal content in essential oils remains minimal, regardless of soil contamination levels. This is primarily due to the high molecular weight and non-volatile nature of heavy metals, preventing their concentration during the distillation process.

Phytoremediation potential of tarragon

The phytoremediation efficiency of tarragon was evaluated using the Bioaccumulation Factor (BCF) and Translocation Factor (TF). These indices are commonly used to assess plant-based remediation potential and depend on factors such as soil conditions, plant species, and heavy metal bioavailability.

The coefficients were calculated using the following equations:

- Translocation Factor (TF):

$$TF = \frac{Cshoots}{\text{Croots}}$$

where:

- *Cshoots* represents the heavy metal content (mg/kg) in the aerial parts (stems, leaves, flowering tops);

- *Croots* represents the concentration in the roots (mg/kg).

- Bioaccumulation Factor (BCF):

$$BCF = \frac{Cplant\ parts}{\text{Csoil}}$$

where:

- *Cplant parts* represents the heavy metal content (mg/kg) in various plant organs (roots, stems, leaves, flowering tops);

- *Csoil* represents the heavy metal concentration in the corresponding soil (mg/kg).

A plant species is considered suitable for phytoextraction when its translocation factor (TF) > 1. If TF < 1, the plant is more suitable for phytostabilization.

The results indicate that tarragon exhibits high translocation potential for heavy metals: Pb: TF = 8.15; Cd: TF = 9.96; Zn: TF = 10.93; Hg: TF = 5.89.

The bioaccumulation factor for roots (BCFroots) was calculated as follows: Pb: BCFroots = 0.059; Cd: BCFroots = 0.061; Zn: BCFroots = 0.021; Hg: BCFroots = 0.099 and is presentet in Tabel 3.

Table 3. Bioaccumulation (BCF) and translocation (TF) factors for tarragon

	TF	BCFroots	BCFleaves	BCF tops
Pb	8.15	0.059	0.43	0.37
Cd	9.96	0.061	0.61	0.55
Zn	10.93	0.021	0.22	0.13
Hg	5.89	0.0995	0.58	0.16

For flowering tops, the BCF values were all below 1, indicating limited metal accumulation in this plant part (Table 4).

Table 4. Composition of tarragon oil (%)

	Component	RI	% of TIC
1	α-Thujene	924	0.233
2	α-Pinene	939	0.428
3	**Sabinene**	969	**21.414**
4	β-Pinene	979	0.671
5	β-Myrcene	991	1.615
6	α-Phellandrene	1005	0.119
7	α-Terpinene	1018	0.412
8	p-Cymene	1026	0.191
9	Limonene	1029	0.963
10	β-Phellandrene	1032	0.146
11	cis-beta-Ocimene	1040	2.244
12	trans-beta-Ocimene	1050	3.529
13	γ-Terpinene	1062	0.745
14	cis-Sabinene hydrate	1065	0.363
15	Terpinolene	1086	2.342
16	beta-Linalool	1097	0.179
17	trans-Verbenol	1141	0.068
18	Terpinene-4-ol	1177	0.761
19	Estragol	1186	0.446
20	β-Citronellol	1211	0.227
21	Citronellyl acetate	1354	2.565
22	Geranyl acetate	1383	1.411
23	**Methyleugenol**	1402	**13.107**
24	β-Caryophyllene	1419	0.236
25	α-Humulene	1454	0.115
26	γ-Curcumene	1481	0.382
27	Germacrene D	1484	3.923
28	Methyl isoeugenol	1490	2.734
29	Bicyclogermacrene	1500	2.597
30	**Elemicin**	1557	**17.429**
31	trans-Nerolidol	1563	0.587
32	**Isoelemicin**	1570	**15.259**
33	(-)-Spathulenol	1578	1.076
34	Caryophyllene oxide	1581	0.162
35	α-Cadinol	1652	0.136
36	Farnesol	1722	0.244
Total			99.059
Oil content, %			1.1

RI - Kovacs relative indices

BCF for leaves reflects the plant's ability to absorb and transfer metals to the leaves, which can be harvested for remediation purposes. The classification of plants based on BCF values is as follows: BCF < 1: Excluder; BCF = 1: Indicator; 1 < BCF < 10: Accumulator; BCF > 10: Hyperaccumulator. The BCF values for tarragon leaves were: Pb: 0.43; Cd: 0.61; Zn: 0.23; Hg:0.58. These results suggest that tarragon functions as an excluder of Cd, Pb, Zn, and Hg when grown in contaminated soils.

Previous studies support the findings of Kocaman (2022) classified tarragon as an accumulator of Pb and Cd (1<Cd-Pb<10) and an excluder of Hg (Hg<1). Ghasemidehkordi et al. (2018) reported Pb bioaccumulation in Artemisia dracunculus L. in Iran. Ozyigit et al. (2018) found that tarragon accumulates low levels of Cd and has not been identified as a heavy metal accumulator.

Essential oil quality

The quantity and composition of tarragon essential oil are influenced by genetic factors, phenological stage, and environmental conditions (e.g., soil composition and light intensity).

The essential oil yield varied between 0.15% and 3.1%, with the highest concentrations occurring at the beginning of flowering (Fildan et al., 2019). The results from this study align with previous reports.

In total, 36 components were identified in tarragon essential oil, primarily belonging to the terpene group (monoterpenes and sesquiterpenes) and phenylpropanoids, which together accounted for 99.059% of total oil composition (Table 4). The dominant compounds (>10%) were: Sabinene (21.414%), Elemicin (17.429%) Isoelemicin (15.259%), Methyleugenol (13.107%).

Based on functional group classification, tarragon oil was composed of:

- Phenylpropanoids: 48.975%
- Monoterpene hydrocarbons: 35.052%
- Sesquiterpene hydrocarbons: 7.253%
- Oxygenated monoterpenes: 5.574%
- Oxygenated sesquiterpenes: 2.205%.

The main phenylpropanoids in essential oil are elemicin (17.429%), isoelemicin (15.259%), methyleugenol (13.107%), methyl isoeugenol (2.734%). The amount of phenylpropanoid compounds in tarragon oils varies widely (25.4% (Sharopov et al., 2020), 36.41% (Fildan et al., 2019), 51.93% (Varban et al., 2023), 52.2%, (Lopez-Lutz et al.,2008), 62.2% (Zawislak and Dzida, 2012), 73.3% (Fraternale et al., 2015).

Variation in methyleugenol composition was found (0.2% (Fraternale et al., 2015), 6.2% (Zawislak and Dzida, 2012), 9.09% (Fildan et al., 2019), 13.107% (this study) 29.19% (Varban et al., 2023). The content of methyleugenol depends on the phenophase, with the highest content identified in *A. dracunculus* in mass flowering (Khodakov et al. 2009). The content of elemicin also varies widely (7.95% (Fildan et al., 2019), 11.43% (Varban et al., 2023), 17.429% (this study), 56.0% (Zawislak and Dzida, 2012). Elemicin has antimicrobial, antioxidant, antiviral and psychoactive effects, and there is no significant difference in its content at the growth stages of tarragon (Varban et al., 2023). Significant differences were also found in the content of estragole (methyl chavicol) (phenylpropanoid compound) (0.446% (this study), 3.9% (Russia), 16.2% (Canada) (Obolskiy et al., 2011), 24.6% (Sharopov et al., 2020), 73.3% (Italy, Fraternale et al., 2015), 82% (USA, Obolskiy et al., 2011). This compound is considered a key component of French tarragon oil (>80%) (Sharopov et al., 2020).

Monoterpenes belong to the second primary chemical class of oil constituents, which reach 40.626% of the total oil composition. The amount of monoterpene hydrocarbons reaches 35.052%, of which sabinene (21.414%), trans-beta-ocimene (3.529%), terpinolene (2.342%), cis-beta-ocimene (2.244%), β-myrcene (1.615%).

Sabinene is a significant component in tarragon essential oils from different geographical origins (Sharopov et al., 2020). It was found in high concentrations in the essential oil of *A. dracunculus* growing in Tibet (China| (19.2%, Liu et al., 2018), Lithuania (14-25%, Sharopov et al., 2020), Romania (9.44-42.4%, Fildan et al., 2019; Varban et al., 2023), Kazakhstan (20.2%, Suleimenov et al., 2010), Poland (20.9%, Zawislak and Dzida, 2012), Tajikistan (29.1%, Sharopov et al., 2020), while in India – trace amount (Verma et al., 2010).

The number of oxygenated monoterpenes reaches 5.574 % of the total composition of the oil. The main oxygenated monoterpenes are citronellyl acetate (2.565%) and geranyl acetate (1.411%).

The sesquiterpenes reach 9.458%. The amount of sesquiterpene hydrocarbons reaches 7.253%. Germacrene D (3.923%) and bicyclogermacrene (2.597%) are the main components that predominate in this group.

The amount of oxygenated sesquiterpenes reaches 2.205% of the total oil composition.

The oil contains (> 1.0 %) caryophyllene oxide (1.076%).

The results show significant differences in the composition of oils obtained from different geographical locations. Studies have shown significant differences in the composition of tarragon essential oil. These differences in the chemical composition of the essential oil extracted from the aerial parts of *A. dracunculus* may be due to environmental conditions (Fildan et al., 2019), habitat, soil pH, plant part used, extraction method applied, and other factors such as genotypes and ontogeny (Obolskiy et al., 2011; Fraternale et al., 2015; Fildan et al., 2019). According to Ekiert et al. (2021), the main components in the essential oil of *A. dracunculus* are estragole (40-85%), sabinene (up to 35%), methyleugenol (up to 25%) and elemicin (up to 57%). The essential oil of tarragon from Tajikistan is characterised mainly by sabinene (29.1%), and estragole (24.6%), the oil from Turkey contains Z anethole (81.0%) (Kordali et al., 2005), the oil from Cuba - elemicin (53.0%) and methyleugenol (17.6%) (Pino et al., 1996), the oil from Iran - anethole (21.2%), E-(β)-ocimene (22.6%) and limonene (12.4%) (Sayyah et al., 2004). The Russian tarragon oil is rich in sabinene (39.4%), elemicin (16.0%), methyl eugenol (14.7%) and iso-elemicin (7.7%), while the French tarragon oil has the main component methyl chavicol (68.6%) (Arabhosseini et al., 2006).

Based on 30 components of 105 essential oils of different geographical origins, Sharopov et al. (2020) established 7 main chemotypes of tarragon essential oil. The results show that the essential oil of *A. dracunculus* belongs to the mixed chemotype variation without a dominant component. Tarragon essential oil is characterised by a mixed sabinene/elemicin/isoelemicin/methyleugenol chemotype.

CONCLUSIONS

Based on the results obtained, the following key conclusions can be drawn:

1. Toxic metal tolerance and phytoremediation: Tarragon is tolerant to toxic metals and can be cultivated on highly contaminated soils (2423.9 mg/kg Zn, 2509.1 mg/kg Pb, and 63.7 mg/kg Cd). It is effective for phytoremediation of soils contaminated with heavy metals.
2. Heavy metal excluder: Tarragon functions as a Cd, Pb, Zn and Hg excluder when grown in contaminated environments.
3. Metal and nutrient uptake: There is a distinct pattern in the absorption and distribution of heavy metals, microelements, and macroelements between the vegetative and reproductive organs of tarragon
4. Essential oil safety: The levels of Pb, Cd, Zn, and Hg in tarragon essential oil, cultivated 0.5 km from NFMW-Plovdiv, were below the Maximum Permissible Concentrations. This makes the oil suitable for use in the pharmaceutical, cosmetic, and related industries.
5. Essential oil composition: phenylpropanoids (48.975%) are the predominant components in tarragon oil, followed by monoterpene hydrocarbons (35.052%), sesquiterpene hydrocarbons (7.253%), oxygenated monoterpenes (5.574%), and oxygenated sesquiterpenes (2.205%).
6. Chemotype classification: Tarragon essential oil from the NFMW-Plovdiv region exhibits a mixed sabinene/elemicin/isoelemicin/methyleugenol chemotype.

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