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Elemental Profiles of Wild *Thymus* L. Plants Growing in Different Soil and Climate Conditions

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Featured Application: The data obtained can be useful for obtaining high-quality food and pharmacopoeia raw materials from wild thyme with a given chemical composition and controlled biological activity.

Abstract: Plants of the genus *Thymus* L. are traditionally used in medicine and cooking due to the presence of biologically active compounds in them that have fungicidal, antibacterial and other medicinal properties and original taste qualities. Genetic features and growing conditions cause the elemental composition, responsibly of the synthesised medicinal compounds. However, information on the contents and distributions of elements in the organs of *Thymus* L. is very limited. This study was to set and compare the elements in organs of wild thyme for different soil and climatic conditions. Two species of wild *Thymus* L. from Mongolian steppe and on the coast of Lake Baikal were collected during flowering. Twenty-four elements, including Si, in soils, roots, stems, leaves and flowers were simultaneously determined by atomic emission spectrometry. Elemental profiles of two species of wild *Thymus* L. are described. It is assumed that Si is a necessary element of the plant. The predominance of the genetic resistance of plants over the influence of soil and climatic conditions is shown.

Keywords: *Thymus serpyllum* L.; *Thymus baicalensis Serg*. L.; elemental profile; roots–stems–leaves–flowers; influence of soil and climatic conditions

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1. Introduction

Plants participate in the migration of chemical elements in natural ecosystems. Although the biological selectivity of plants in essential and toxic elements allows their chemical composition to be controlled within certain limits, the content of elements in plants is influenced by natural conditions such as soil type, climate, landscape, insolation, seasons and anthropogenic activity. Therefore, an investigation into the elemental composition of a soil–plant system coupled with environment discloses useful information [1–3], and variations in trace element concentrations can be used as a tool for examining specific features of plant growth conditions as well as the state of the environment [1–4]. The determination of a wide range of elements in plants is required for geoecological environmental monitoring [1,3], assessment of the quality and safety of food and medicinal plants [5,6], as well as regulation of their quantity in the diets of humans, domestic animals and poultry [7–9].

Currently, it is of commercial interest to study the biological activity of medicinal plants, herbs and spices that have been used for centuries in folk medicine and cuisine including thyme. The genus *Thymus* L. includes a large variety of species that are difficult to classify visually, as different species of the *Thymus* L. genus are similar in appearance [10,11]. Several hundred species of *Thymus* L. are widely distributed throughout the Eurasian continent (excluding the tropics) as well as North Africa and Greenland. More than 170 species of thyme grow on the territory of Russia and its neighbouring countries. In Eastern Siberia and Mongolia, *Thymus serpyllum* L. (creeping thyme) [11] and *Thymus baicalensis Serg.* L. [12] are among the most common species. Groundcover plants *Thymus serpyllum* L. and *Thymus baicalensis Serg.* L. spread intensively in steppe zones, filling gaps between boulders and creating a dense carpet-like groundcover that is completely covered with numerous purple–pink flowers during the flowering period, which lasts between 2 and 2.5 months. The specific pleasant smell of this plant is due to the essential oil present in this genus, the chemical composition of which varies from one plant species to another and is dependent on morphological characteristics, climatic conditions, the stage of plant maturity, the time of harvest and post-harvest processing [11–15].

The composition and concentration of organic compounds synthesised by plants, the mechanisms by which these organic compounds influence metabolic processes as well as their interaction with bacteria, viruses and fungi, were previously discussed in a number of publications [16–20]. In studies comparing essentials oils extracted from thyme with those obtained from other plant species, it was shown that thyme's essential oils were the most effective for the treatment of fungal and bacterial infections and also endocrine and tumour diseases [17–22]. Numerous publications have been devoted to the mechanisms of the synthesis of biologically active compounds in plants including those of a catalytic nature. The role of coenzymes, containing ions of essential elements and/or their organic compounds on the enzyme structure, are discussed in [14,23]. Although over 70 chemical elements have been identified in various plant species [1,3], the data concerning the abundances and specific roles of each trace element contained in the inorganic and organoelement compounds of *Thymus* L. species are very limited [15,23–31]. For instance, the composition of the essential oil of Hungarian *Thymus pannonicus* plants was studied, and the role elements, such as K, Na, Ca, Mg, Fe, Mn, Zn, Cu, Cr and Mo, played in this was described through investigation of the biosynthesis of certain volatile compounds [23]. Chemometric data processing revealed a correlation between Zn and citral, Mn and oxygenated monoterpenes, and between Mg and β-bourbonene, which explains the participation of these metals in the biosynthesis of the plant's essential oil. However, the initial analytical information used is incomplete, since the authors did not determine the high concentrations of Si and Al, which are generally characteristic of *Thymus* L. plants, and did not include them in the chemometric models. At the same time, it is known that mono- and dimmers of silicic acid (i.e., biogenic silica) are present in xylem, phloem and plant cell walls. Transport of soluble silicon compounds is carried out through the synthesis of a special Si-containing protein to ensure the mechanical strength of the cell wall, increase the natural resistance of plants to abiogenic stresses and fungal diseases, reduce the toxic effects of Fe, Mn, As, Al, ^{90}Sr and phenols [1,15,21,24,32]. In addition, numerous volatile organic compounds are characterised by strong variability [7,12,14,19] due to the presence of different factors including elemental composition. It is difficult to interpret unambiguously the complexity relationships between trace elements and organic compounds of *Thymus* L. species plants without knowing the distributions of a wide range of elements in their organs. For this reason, the aim of the study was to compile and compare the elemental profiles (also containing silicon information) of organs of two *Thymus* L. species growing wild under different soil types and climatic conditions.

2. Materials and Methods

2.1. Sample Collection

Two plants of the species *Thymus* L. were the objects of the study. The herbs were collected during a period of intensive flowering under different soil types and climatic conditions: (1) Mongolian thyme—*Thymus serpyllum* L., Tsongjin Boldog steppe area, Nalaikh District, 54 km from Ulaanbaatar, Mongolia; (2) Baikal thyme *Thymus baikalensis Serg*. L. on the coast of Lake Baikal near the village of Sakhyurta, approximately 250 km from the city of Irkutsk, Russia.

Both territories represent mountainous landscapes. Soils in Mongolia are mountain meadow–steppe soils that are formed on poorly leached soil-forming rocks (Mesozoic granites, carboniferous metamorphic clay shales and Neogene variegated clays) under conditions of a periodic washing water regime. The mountainous steppe soils of the Baikal coast are predominantly carbonate–clay, formed in the cold climate of Eastern Siberia (Russia) with moderate humidity. The Mongolian steppe territory, where the plants were selected, belongs to the eastern sector of the most continental part of the arid zone of Central Asia and has a mountainous climate with features of a sharply continental character. The climate in Sakhyurta is sharply continental.

In each district, arrays of 3 × 3 m in size were allocated. Soil samples were also taken at the plant collection sites using the "envelope" method: five points along the edges and in the middle of the site to a depth of 10–15 cm. Similarly, samples of plants of the species *Thymus* L. with roots and flowers were taken from these territories.

The collected plants were thoroughly washed with distilled water in order to remove soil particles; then divided into roots, stems, leaves and flowers; dried with filter paper. Samples of the plant organs and soils were dried at room temperature in a shaded place. The dry weight of the average sample for each type of thyme and soil were 200–300 g and 1 kg, accordingly. The produced material was crushed to a particle size of −0.08 mm in the KM-1 ball agate mill (Fritsch GmbH, Idar-Oberstein, Germany). The organic carbon content was approximately 3 wt.% in the soils from both places.

2.2. Atomic Emission Spectrometry with Direct Current Arc Discharge

Direct current arc atomic emission spectrometry (DC-arc AES) was used to determine the relative concentrations of 24 elements (Al, B, Ba, Be, Ca, Co, Cr, Cu, Fe, Ga, K, Li, Mg, Mn, Na, Ni, P, Pb, Si, Sr, Ti, V, Zn and Zr) [33]. This method does not require concentration or ashing of the samples. To reduce the negative impact of the organic matrix on the residence time of the substance's atoms in the excitation zone (in the arc discharge plasma), samples of plants and soils that had a high organic matter content were diluted with a spectroscopic buffer that slows down the oxidation process and maintains a constant arc temperature. Graphite powder (*ultra pure*) was used as a spectroscopic buffer. The sample, mixed in a 1:1 ratio with the buffer, was homogenised in an agate mortar for 5–7 min. The prepared samples were stored in a desiccator. Immediately before the analysis, two subsamples (10 ± 1 mg) of mixtures of either a calibration CRM or a probe were weighed and placed in the channel of pre-fired graphite electrodes. The following are the dimensions of the lower electrode (i.e., anode): base diameter—6 mm; height—35 mm; channel depth—4 mm; external diameter—5.4 mm; internal diameter—4 mm; wall thickness—0.7 mm. The upper electrode (i.e., cathode) was a graphite cylinder rod with plane ends: height—7 cm; diameter—6 mm. The mixture of the sample and graphite powder occupied no more than 80–90% of the electrode channel, providing a jet evaporation, primarily of highly volatile Pb, Zn and Li compounds. Plant and soil samples, graphite powder and their mixtures were weighed using the analytical scales LVA-210A (Sartogosm, Saint Petersburg, Russia).

2.3. Instrumentation

To record atomic emission spectra, an upgraded spectral setup was used. It consisted of the diffraction spectrograph DFS-458S (PO KOMZ, Kazan, Russia), the DC arc generator "Vesuvius" and photoelectric MAES analyser (VMK-Optoelectronics, Novosibirsk, Russia). The processing of the spectra was performed in the commercial program "ATOM" (VMK-Optoelectronics, Novosibirsk, Russia). It is described in detail in [33].

2.4. Calibration

The plant-matrix certified reference materials (CRMs) were used as calibration samples: GSO 8921-2007 and COOMET 0065-2008-RU (EC-1, Canadian pond weed), GSO 8922-2007 and COOMET 0066-2008-RU (Tr-1, meadowherbs mixture) and GSO 8923-2007 and COOMET 0067-2008-RU (LB-1, birch leaf) [34]; GSO 1483-78 (SBMK-01, potato tubers), GSO 1484-78

(SBMP-01, wheat grain) and GSO 1485-78 (SBMT-01, grass mixture) [35]; GWB07602, GWB07603 (GSV-1, GSV-2, bush branches and leaves), GWB07604 (GSV-3, black poplar leaves) and GWB07605 (GSV-4, tea leaves) [36].

The content of the elements in the calibration CRMs and their mixtures varied in wide ranges. However, they were not wide enough to analyse plants growing in areas with different climatic conditions and which had differing anthropogenic impacts. Therefore, for the analysis of plants from arid or heavily polluted areas, the upper limits of the measuring ranges were expanded by using mixtures of plant-matrix CRMs combined with CRMs of soils and loose sediments from the collection of IGC SB RAS (SGHM-1—GSO 3483-86, SGHM-2—GSO 3484-86 and SGHM-3—GSO 3485-86) and CRMs of sediments and silts (SGH-1—GSO 3131-85, BIL-1—GSO 7126-94 and BIL-2—GSO 7176-95), soil CRMs OOKO-151, -152 and -153; and other nature CRMs [37].

Examples of calibrations in bilogarithmic coordinates constructed in accordance with the empirical Lomakin–Scheibe equation (program "ATOM") are shown in the Supplementary Materials (Figure S1).

2.5. Quality Assurance and Quality Control

To assess the accuracy and traceability of the results by the DC-arc AES, in addition to the samples under study, we analysed the encrypted CRMs of the meadowherbs mixture Tr-1 GSO 8922 and tea leaves GSV-4 GWB07605. The accuracy, expressed as a relative standard deviation, amounted to 10–15% on average, varying from 1 to 35% for different analytes, depending on the element content. The reliability of the DC-arc AES was evaluated by comparing the results of the analysis of encrypted CRMs with the data obtained via the ICP-AES and ICP-MS methods [33].

The ICP-AES analysis was accomplished using the atomic emission spectrometer iCAP 6300 DUO (Thermo Scientific, Waltham, MA, USA); ICP-MS determinations were obtained via the mass spectrometer ELEMENT 2 (Finnigan MAT, Bremen, Germany). The solutions for analysis were prepared following the certified method [38]. After acid digestion, the sample solutions were evaporated with a small addition of hydrofluoric acid to concentrate the impurities. This procedure results in the removal of silicon from the solution. Therefore, silicon was not determined. Losses of other trace elements, such as Zr, Al and Ti, are also possible [39]. Perhaps also for this reason, the Zr content in the CRMs Tr-1 and GSV-4, obtained by the DC-arc AES method, was greater than that determined by the ICP-MS by 30% and more than 30 times, respectively (Tables S1 and S2, see Supplementary Materials). The concentrations of Be and Ga in the solutions were insufficient to determine them using the ICP-AES method (Table S1). For the DC-arc AES method, the determination of Be and Ga was accomplished without difficulty. The vanadium contents in GSV-4 (Table S2), obtained through different methods, were similar to each other, but significantly lower than the content recommended by the developer of this CRM. These results can be used to correct the V content in the GSV-4 CRM. The comparison between the results obtained using different methods and the certified contents in Tr-1 and GSV-4 CRMs indicates their good agreement for most of the elements (Tables S1 and S2). The 95% probability confidence intervals of the elemental contents obtained using different methods were comparable, and the means were close in accuracy.

3. Results and Discussion

The 24 element contents obtained by the DC-arc AES in the dried roots, stems, leaves and flowers of *Thymus serpyllum* L. and *Thymus baikalensis Serg*. L. plants as well as soils of Mongolia (1) and Russia (2) are given in Table 1. This Table also presents the literature data on the typical concentration ranges of these elements in the dry matter of plants of different species [1,2]. The observed contents of Al, Ba, Ti and Zr were found to exceed that recorded in the literature data (marked in italics in Table 1). Concentrations of elements (Si, K, P, Ti, Zn, Cu, V and B) that are close in value or differ by less than 30% for different *Thymus* L. species are shown in bold in the "Flowers" column. The simultaneous determination of both essential (P, K, Ca, Mg, Zn, Cu, B, Mn, Co, Na, V and Fe) [2] and toxic elements (Al, Be, Ba Cr, Ga, Li, Ni, Pb and Sr), as well as silicon, which is noted in a number of publications as an element necessary for plants in the maintenance of a normal life cycle [15,24], is an important advantage of the analytical method used.

Table 1. Element contents (mg/kg) in soils and the dried plant organs of *Thymus* L. species (n = 3).

Element	Soil	Roots	Stems	Leaves	Flowers	Gathering Place (1 or 2) *	Range of Element Content in the Dried Plants [1,2]
Al	85,850	*21,550*	5540	6700	6280	1	*<100–10,000*
	56,950	1710	1900	2140	9500	2	
B	25.1	18.0	20.0	27.1	**22.5**	1	2–800
	9.5	15.1	13.6	17.6	**18.0**	2	
Ba	411	*271*	*236*	*220*	126	1	*1–160*
	93	73	112	117	113	2	
Be	2.85	0.57	0.26	0.20	0.25	1	<0.001–7
	0.61	0.03	0.06	0.07	0.10	2	
Ca	8495	7720	7245	8715	8820	1	3000–100,000
	44,250	6035	7795	10,450	20,850	2	
Co	4.2	2.2	1.3	1.0	0.9	1	0.05–10
	28.7	1.7	2.4	1.6	4.2	2	
Cr	36.4	10.5	4.6	3.3	2.3	1	1–1100
	153	9.6	10	10	54	2	
Cu	23	16	20	13	**29**	1	1–500
	60	29	24	32	**39**	2	
Fe	12,950	7095	2565	2520	1945	1	300–100,000
	49,500	1475	2000	1975	6795	2	
Ga	13.0	4.2	2.3	1.3	1.6	1	0.02–16
	11.8	0.7	1.2	0.9	2.4	2	
K	38,400	8050	16,100	5040	**16,100**	1	5000–80,000
	32,500	8480	26,700	19,200	**13,900**	2	
Li	9.2	2.9	2.6	2.0	4.0	1	0.02–1000
	11.7	2.0	2.0	2.0	9.0	2	
Mg	2450	2010	1380	2150	2100	1	200–60,000
	34,800	2500	3200	5025	12,700	2	
Mn	262	284	155	132	122	1	15–330
	1043	70	96	114	240	2	
Na	29,400	5050	4365	2140	**3930**	1	200–100,000
	8545	386	1730	1315	**3290**	2	
Ni	17.8	14.7	10.1	10.8	7.8	1	0.05–50
	62.6	15.2	9.3	9.4	30.2	2	
P	278	651	995	1165	**1600**	1	100–70,000
	360	820	995	1155	**1760**	2	
Pb	21.0	6.1	3.2	1.3	1.9	1	0.01–2500
	25.4	0.5	1.2	0.6	1.1	2	
Si	316,500	80,000	29,100	29,100	**37,000**	1	1000–100,000
	258,000	7865	12,950	10,900	**37,000**	2	
Sr	103	110	89	86	64	1	1.5–600
	149	49	45	57	52	2	
Ti	1640	*1007*	*498*	*451*	*400*	1	*0.15–80*
	2640	*98*	*120*	*133*	*396*	2	
V	23	16	20	13	**29**	1	0.2–1000
	60	29	24	32	**39**	2	
Zn	71	61	52	45	**61**	1	5–250
	52	49	50	48	**51**	2	
Zr	455	*62*	*27*	*23*	*50*	1	*0.005–2.6*
	30.4	*1.0*	*5.5*	*2.5*	*4*	2	

* **Gathering Place:** 1—Mongolia, Ulaanbaatar; 2—Russia, Irkutsk.

Soil samples from the sites where we collected the two *Thymus* species differed in the contents of major elements, such as Fe, Ca, Mg, Na, Mn and Ti, by 3.5, 4.9, 18, 2.5, 4.2 and 1.7 times, respectively (Figure 1). The contents of trace elements, such as P, Co, Ga, Li, Pb, Sr and Zn, in soil samples varied by less than two times and were regarded as similar. The concentrations of trace elements in the Mongolian soil samples were higher compared to the Baikal ones as follows: 4 times for barium, 4.7 times for beryllium and 16 times for zirconium. Compared to the Mongolian soil, the Baikal soil samples were more enriched in boron (2.6 times), cobalt (5 times), Cr (3 times), Cu (2.3 times), V (5.8 times) and Ni (3.4 times). However, these features of the soils' elemental composition did not affect the absorption by different *Thymus* species of elements such as P, B, Zn, Cu, Ca and Mg, which are traditionally regarded as essential elements (Figure 2a).

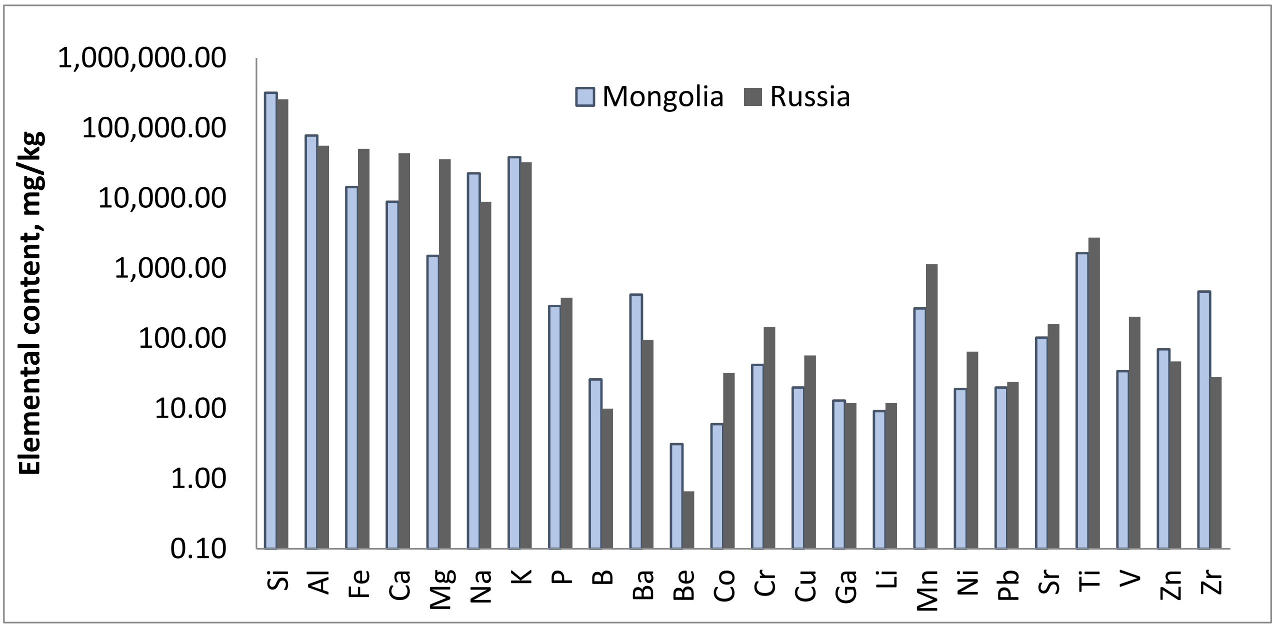

Figure 1. Elemental profiles of the soils.

The element ratio in the roots and soil indicates the intake of individual elements by the root system. The value of the ratio depends both on the specific physiological characteristics of the plant and the total element content in the soil. The two plants of the *Thymus* species growing in different climatic conditions demonstrate unrestricted uptake of only such elements as P, B, Cu and Zn. A similar phenomenon was observed in the drier climate of Mongolia for both the essential elements (i.e., Ca, Co, Mg, Mn and V) and conditionally toxic elements, such as Ni and Sr, the concentrations in the soil of which were very low. Around Lake Baikal, the contents of Mg, Mn, Ni, V and Sr in the soil are higher, and the root system limits their uptake into the plant. Only the accumulation of Ba, which is scarce in the soil, is unrestricted by the root system. In the Mongolian soil samples, the contents of Ca, Mg, Fe, Mn, Co, Cr, Ni and Ti were lower compared with the Baikal samples. Despite this, the distribution profiles of these elements in plant organs differed slightly. The Si, Al, Be, Ga, Li, Na, Pb and Zr concentrations were found to be high in the Mongolian soil, but their uptake through the roots was limited (Figure 2), and they hardly accumulated at all in plant tissues. Though the total contents of Si, Na and Be in the Baikal soil were lower, the distribution profiles of these elements in the soils and organs of the two plants were similar to each other: the concentration of the element in the roots decreased sharply, remained approximately the same in the stems, leaves and flowers or increased slightly in the leaves and flowers (Figure 2).

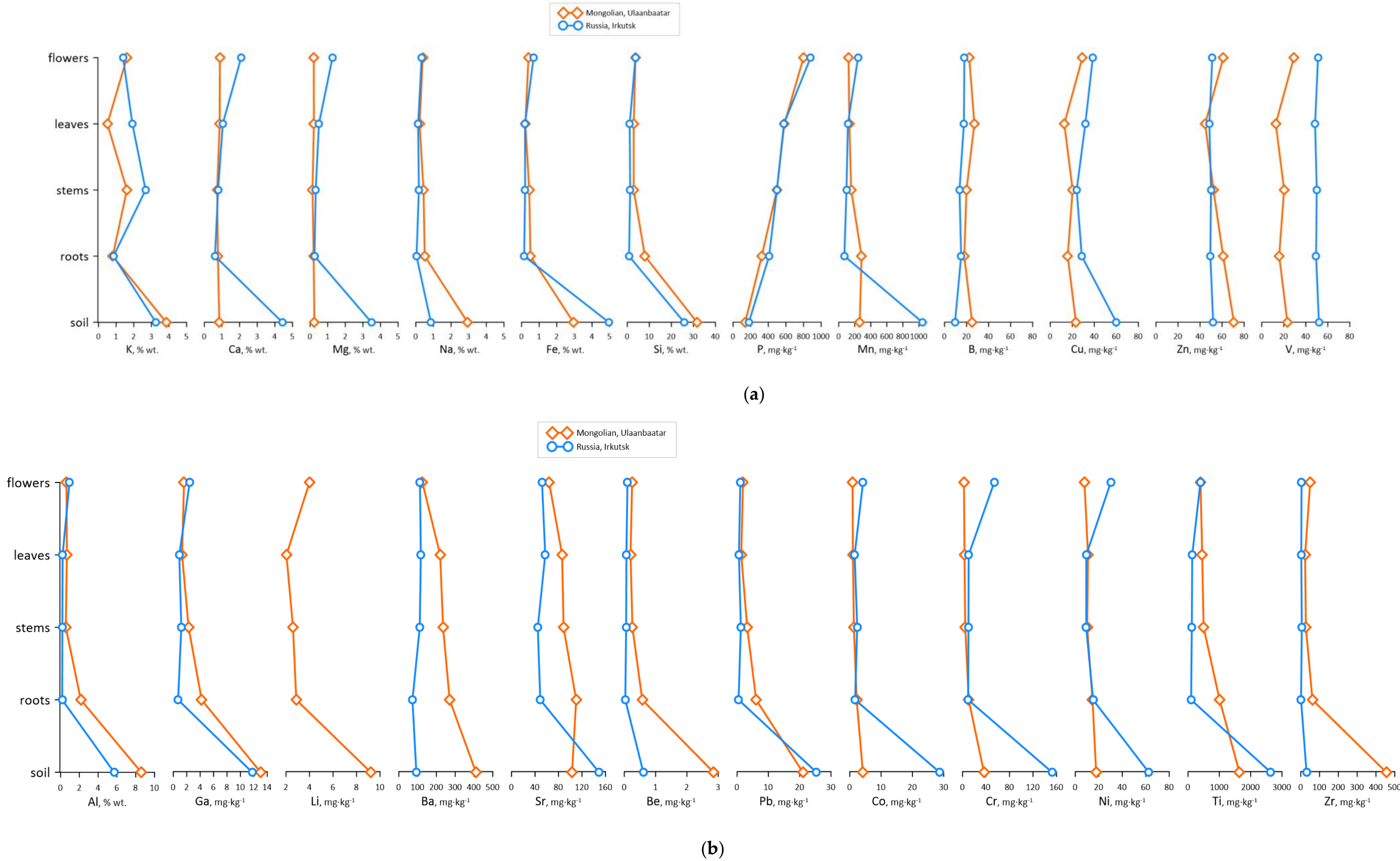

Figure 2. The elemental profiles: (**a**) essential elements; (**b**) toxic and sub-toxic (or sub-essential) elements.

The element content in plants usually reflects their biological necessity for plant survival. The element concentration is also an indication of the composition of the soil in which they grow. As was previously shown [26], higher Al content in the organs of *Thymus serpyllum* L. may result from the increased abundance of this element in the soil. A similar phenomenon was also observed in plants growing in soils with a higher chromium content [25]. Chromium and nickel are usually considered toxic elements, but plants can use these metals in the biosynthesis of organic compounds for electron charge transfer, just as Cu, Fe, Mn, Co, Mo and V are used [1,2,40]. The behaviour of Ba and Sr in plant organs depends on their content in the soil. If the element is present in large quantities in the soil, then its concentration decreases towards the flowers. If the element is not present in large quantities, then its accumulation in flowers will remain constant. Thus, when the concentration of Ba in the soil is 400 and 93 mg/kg, concentrations of 126 and 113 mg/kg in flowers are observed; when the concentration of Sr in the soil is 100 and 150 mg/kg, concentrations of 126–113 mg/kg and 64–52 mg/kg are observed in flowers. The Ca/Sr ratio in plant organs is 70–138 for *Thymus serpyllum* L. and 123–402 for *Thymus baikalensis Serg*. L., accordingly. The increase in the ratio of these elements indicates an increase in the contribution of enzymes to the transfer of elements from the soil in the Baikal plant, situated in a wetter climate [41]. The increased content of Cr, Ni and V in *Thymus baikalensis Serg*. L. flowers is likely associated with the same effect. However, it is difficult to unambiguously attribute biophilic behaviour to Ni, Co and Cr. Such elements as Be, Ga and Pb were evaluated as toxic, since even at low concentrations in soils, their abundances decreased sharply in the root system, although they accumulated in small quantities in the flowers.

We observed an intense accumulation of phosphorus from the roots to the leaves and flowers even at low P levels in the soil. The food supply of biophilic K, Ca, Mg, Fe, Mn and Na from the soils to the plants was partially limited but reached the maximum concentration in the flowers. Similar elemental profiles of the "roots–stems–leaves–flowers" sequences for biophilic B, Cu, V and Zn for the different *Thymus* species were obtained (Figure 2).

It is known that silicon compounds are involved in the synthesis of proteins, which allows for the transport of soluble silicon compounds required for the mechanical strength of the cell wall. In gymnosperms and aquatic plants, the silicon content is often higher than 1 wt.%. The element is difficult to analyse by ICP-AES and ICP-MS. During polymerisation and hydrolysis of silicon, large particles are formed in the solutions of the plant samples, which are filtered out before the analysis or are distilled through heating with hydrofluoric acid [39]. These Si contents are not determined by method of X-ray fluorescence spectrometry because it has a high detection limit for silicon. The factors mentioned above usually cause the exclusion of silicon from the analyte list. For plants grown in the studied climatic conditions, the concentrations of Si and Ti in the "roots–stems–leaves–flowers" sequence are significantly different, though in flowers their abundances are similar: Si—3.70 wt.% and Ti—400 mg/kg (Table 1). These are probably the maximum concentrations of Si and Ti that *Thymus* plants can accumulate and use in essential processes safely. In the shoots of *Thymus marschallianus* L., the silicon content was found to be 2.21 wt.% [15], which is in good agreement with the data obtained for the studied plants, averaging for stems and leaves 2.91 and 1.19 wt.%, respectively. Data concerning the Ti content in *Thymus* plants were not found in the literature.

The 24-element profile of Baikal *Thymus* indicates that flowers accumulate more trace elements than vegetative organs (i.e., roots, stems and leaves) (Figure 3a). This pattern was not observed for Mongolian *Thymus* (Figure 3b). It is likely that the dry steppe climate of Mongolia and the specific features of this plant limit the transfer of some trace elements from the soil to the flowers [41]. As seen in Table 1, the assimilation of dominant and trace elements in the flowers of the two *Thymus* L. species differs as follows:

Thymus serpyllum L.

Si > K > Al > Ca > Na > Mg = Fe > **P > Ti > Mn = Ba > Sr = Zn** > Zr > **Cu = V > B > Ni > Li** > **Cr > Pb > Ga > Co > Be**;

Thymus baikalensis Serg. L.

Si > Ca > K > Mg > Al > Fe > Na > **P > Ti > Mn > Ba > Sr = Zn**= Cr > **Cu = V > Ni > B > Li** > Zr = **Co > Ga > Pb > Be**.

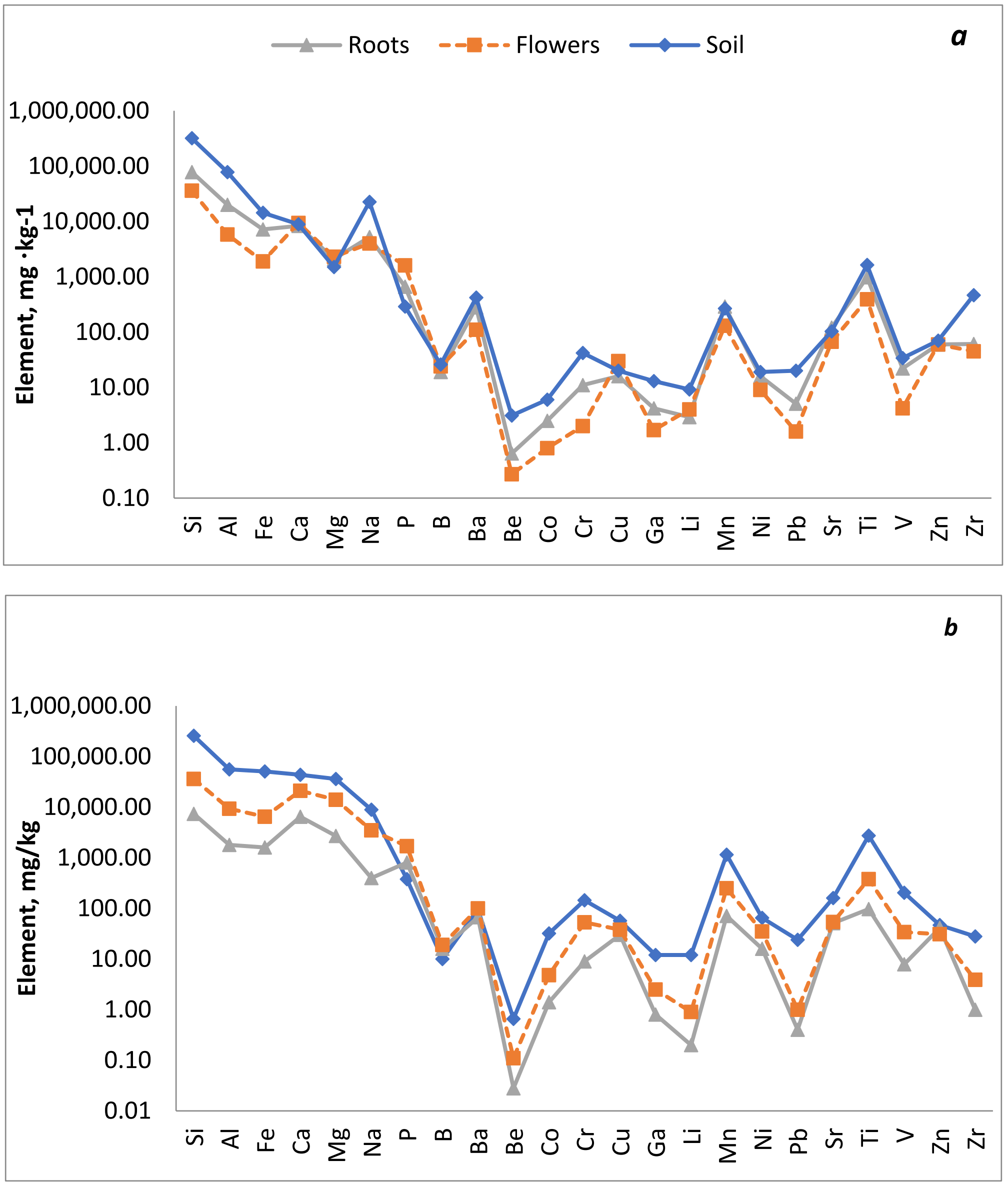

Figure 3. Element contents in the soils, roots and flowers of plants: (**a**) *Thymus serpyllum* L. (Mongolia, Ulaanbaatar); (**b**) *Thymus baikalensis Serg*. L. (Russia, Irkutsk).

Silicon is probably a necessary element of *Thymus* species due to the fact that its content is the largest in flowers and comparable to the essential elements (i.e., potassium and calcium). The mineral contents of plants are quite variable, but their composition is strongly controlled through genetic means. This is evidenced by the near-identical contents in the flowers of the biophilic K, Na, P, B, Cu and Zn but also Si and Ti as conditionally biophilic elements, which are rarely determined in plants (Table 1, Figure 2). The V, Mn, Ba, Sr and Al contents in the flowers of the two species, growing in different natural and climatic conditions, varied no more than twice. Some of the elements found were redox-active, which makes them indispensable as catalytically active cofactors in enzymes; the others demonstrate enzyme-activating functions, while the third group of elements play a structural role in protein stabilisation. The role of each element in the biosynthesis of numerous volatile organic compounds in plants of this species is yet to be fully determined.

For the Mongolian species, the accumulation of Si, Al, Fe, Na, Ba, Be, Co, Cr, Ga, Ni, Pb, Sr and V in the flowers was lower than in the roots (Figure 3a). The concentration of most elements in the flowers of Baikal thyme was lower than the soil but higher than the roots (Figure 3b). Compared to the dry steppe climate of Mongolia, the high moisture levels near Lake Baikal promotes the uptake of elements from the soil to the plants.

The published elemental concentrations in the thyme leaves, measured using a number of analytical methods and obtained for the two studied plants, were collected and compared (Table 2).

Table 2. The elemental contents (mg/kg) in the leaves and spices of the Thymus plant, found by different analytical methods.

Element	*Thymus vulgaris*	*Thymus vulgaris*	*Thymus serpyllum*	*Thymus marschallianus*	*Thyme (Thymbra spicata)*	*Thymus vulgaris*
Al	6.35–7.90					
Ba					81.6	
Ca					**7759**	**21,100**
Co					0.15	
Cr	0.83				0.57	
Cu		4.1	**7.2**		6.1	**8.8**
Fe		111.5	267.3		440	427
K					14,708	**14,700**
Mg					**2115**	
Mn		60.9	84.9		**116**	19.3
Na					106.5	
Ni					1.5	
Pb		**0.62**	**1.12**			
Si				**22,100**		
Sr					45.6	26.8
Zn		**32.8**	14.4		22.4	**35.1**
Gathering place	Spain, supermarket	Austria, near Vienna			Turkey, supermarket	Syria, supermarket
Method	ETA–AAS	ETA–AAS, FAAS			ICP–MS	PIXE
Reference	[25,26]	[27]		[15]	[29]	[28]

Element	*Thymus vulgaris, Labiatae*	*Thyme pannonicus All. (Lamiaceae)*	*Thymus vulgaris*	*Thymus serpyllum*	*Thymus baikalensis*
Al				6700	2140
Ba	18.06			200	117
Ca	**13,810**	**7886.1–26,580.94**	**18,066**	**8715**	10,450
Co	0.193			1.0	1.6
Cr	0.97	0.41–1.41		3.3	10
Cu	**12.17**	**7.04–14.59**	**15**	**13**	32
Fe	301	89.5–749.91	**1194**	**2520**	**1975**
K		**8006.06–23,066.93**	**10,160**	**5040**	**19,200**
Mg	**1670**	**1820.34–3802.95**		**2150**	5025
Mn	44.5	20.54–219.09	**150**	**132**	**114**
Na		2.53–114.01		2140	1315
Ni	2.34	1.35–**16.22**		**10.8**	**9.4**
Pb				**1.30**	**0.60**
Si				**29,100**	**10,900**
Sr	27.60			86	57
Zn	20.4	**33.85–106.66**	**51**	**44.7**	**48.5**
Gathering place	Turkey, supermarket	Serbia	Spain	Mongolia, Ulaanbaatar	Russia, Irkutsk
Method	ICP–MS, ICP–AES	ICP–AES, FAAS, GFAAS	TRXF	DC–arc AES	
Reference	[30]	[23]	[31]	This article	

Note. Empty cells–no data.

The elements were determined in the leaves of wild and cultivated *Thymus* species [15,23,31], or their "herb" form, and sold in supermarkets and markets [25,27–30]. Similar element abundances are displayed in bold in this table. The values of Ca, Cu, K, Mg, Mn, Ni, Zn, Si and Pb concentrations agree satisfactorily, despite differences in plant species, variations in growing conditions and, possibly, specific pre-treatment of the leaves before being sold for use as a dried herb. Thus, the genetic relationship linking plants from different regions of the world is observed. Analytical traceability is also present for the results of elements from 1 to 6, obtained mainly using different methods of atomic absorption spectrometry (i.e., FAAS, ETA-AAS and GFAAS), for elements 11–18 found using one or more analytical methods, such as ICP-AES, ICP-MS, FAES, TRXF or X-ray analysis, including different options for chemical preparation of plant samples (acid digestion, melting, etc.). However, the highest number of simultaneous element concentrations (24 elements) was determined using the DC-arc AES method without the use of ashing or acid digestion of plant samples.

4. Conclusions

Two *Thymus* L. plant species growing wild in different soil types and climatic conditions were collected and studied: *Thymus serpyllum* L.—in the Mongolian steppe—and *Thymus baikalensis Serg*. L.—on the coast of Lake Baikal in Russia. The concentrations of 24 elements were determined in the underground and aboveground parts of the two *Thymus* L. species. To obtain the element concentrations, we used the method of atomic emission spectrometry with arc discharge, which does not include ashing or acid digestion of plant samples. The average RSD values amounted to 10–20%. The traceability of the results was evaluated based on the data for the certified reference samples (i.e., CRMs Tr-1 and GSV-4) obtained by ICP-AES and ICP-MS.

For *Thymus* species plants growing in different soil types and climatic conditions, the accumulation profiles of 24 elements in the sequence "soil–roots–stems–leaves–flowers" was obtained. The maximum Si accumulation in the aboveground organs of the plants indicates the essential nature of this element in the plant's life cycle gained during the plant's evolution. Although the absolute values of element concentrations varied, the middle members of the obtained sequence of trace elements (i.e., element profiles) remained constant: … P > Ti > Mn ≥ Ba > Sr = Zn … Cu = V > B > Ni > Li … . The similar concentrations of Si, K, P, Ti, Zn, Cu, V and B found to have accumulated in plant flowers, along with very similar values of Na, Mn, Ba and Sr, also indicate the high level of genetic resistance in *Thymus* plants to external element concentrations and a less significant influence of soil and climatic conditions. Such elements as Pb, Ga and Be were found to be toxic to plants. It is likely that, depending on the accumulated content of Fe, Co, Ni and Cr, these elements can characterise both essential behaviour and toxic processes. It was shown that the behaviour of Zr in aboveground plant organs was similar to the behaviour of Fe, Ca, Mg, Cr, Co and Ni: the higher the content of the element in the soil, the more it accumulated in aboveground plant organs (leaves and flowers).

The data obtained can be useful for collecting wild thyme and obtaining high-quality food and pharmaceutical raw materials with a given chemical composition and controlled biological activity.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/app12083904/s1, Figure S1: Calibrations for determination of phosphorus (a) and silicon (b) contents in the soils and plant organs by the DC-arc AES technique, Table S1: Certified (A ± ΔA) and defined (C ± ΔC) element contents in GSO Tr-1 CRM by DC-arc AES, ICP-AES and ICP-MS (P = 95%, *n* = 3), Table S2: Certified (A ± ΔA) and defined (C ± ΔC) element contents (mg/kg) in GSV-4 CRM by DC-arc AES and ICP-MS (P = 95%, *n* = 3).

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