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Original article

Distribution of Minerals and Some Heavy Metals in the Leaves and Stems of Salvia, Ziziphus, Thyme, and Artemisia Plants

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ABSTRACT
Depending on the species and the organ of the plant, medicinal and aromatic plants can vary significantly in their mineral make-up. This experiment compared sodium (Na), potassium (K), iron (Fe), zinc (Zn), copper (Cu), and manganese (Mn) in the leaves and stems of Salvia, Ziziphus, thyme and Artemisia growing in the Al-Jabal Al-Akhdar region of Libya in spring 2024. Measurement of sodium and potassium was done by flame photometry, and Fe, Zn, Cu, and Mn by atomic absorption spectrometry, after acid digestion. The most common macro element in all the samples was potassium. It had a mean concentration of 4.98 ± 0.84 mg/g in leaves and 5.39 ± 1.20 mg/g in stems, versus 1.28 ± 1.09 and 0.897 ± 0.485 mg/g in sodium, respectively. The mean concentration of the trace elements in both organs had the same order: Mn > Zn > Cu > Fe. Artemisia leaves had the highest Na (2.84 mg/g), Zn (37.07 µg/g), Cu (15.09 µg/g) and Mn (66.39 µg/g) and Salvia leaves had the highest K concentration (5.57 mg/g). Artemisia had the highest K (6.53 mg/g), Zn (18.27 µg/g), and Cu (9.21 µg/g), Ziziphus the highest Mn (77.50 µg/g) and thyme the highest Fe (1.02 µg/g). The findings show apparent species- and organ-based variations in the distribution of elements. Since no analytical replicates, recovery, detection limits or certified reference-material results are reported in the dataset, the results should be considered as descriptive and cannot be used independently to determine dietary adequacy, toxicity, or safety.

Introduction

Medicinal and aromatic plants play a significant role in the practice of traditional medicine and are still useful raw materials in food, herbal preparations, cosmetic and pharmaceutical studies. The comparatively moist Mediterranean weather and topography foster one of the richest floras in Libya in Al-Jabal Al-Akhdar. An area survey of Wadi Al-Kouf, in this region, recorded 107 medicinal species in 49 families, which not only reflects the rich botanical diversity of this region, but also the ongoing local use of medicinal plants [1]. Other genera or plant groups commonly known in Mediterranean ethnobotanical practice include Salvia, Ziziphus, thyme and Artemisia are commonly studied as sources of chemical constituents and biological activity.

Medicinal plants can be valued based on more than just their secondary metabolites. It also have an inorganic fraction that contributes to plant metabolism and could affect the quality of consumed or extracted material. Potassium is a primary plant nutrient that regulates osmotic potential, stomata, enzyme activation, and ionic balance, and sodium is typically not needed by most higher plants, but can sometimes help regulate osmotic potential in selected species and environments. Iron, zinc, copper and manganese are needed in lesser amounts and they are involved in electron transport, photosynthesis, antioxidant defense and many enzyme systems [2]. Their levels, however, are informative markers of plant mineral nutrition, but should not be automatically viewed as a marker of therapeutic efficacy. The interacting biological and environmental processes control the accumulation of elements in plant tissues. The availability of an element to roots is influenced by soil parent material, pH, organic matter, salinity, moisture, and the chemical form of an element. The final tissue concentration [2,3] is further determined by species-specific uptake mechanisms, root architecture, transport proteins, growth stage, and the redistribution of elements between vegetative organs. As a result, even samples of the same genus but at various times or habitats can have very different elemental profiles. The accumulation of mobile nutrients by leaves due to their high metabolic activity, and stems serving as a transportation path or storage tissues temporarily, can

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depend upon the direction and magnitude of the difference between the two depending on the element and species. Quality-control criteria of herbal materials also include reliable elemental characterization.
To differentiate between endogenous mineral material and soil particles, atmospheric deposition or analytical contamination, standardized sampling, thorough washing, complete digestion, calibration, blank correction, and suitable quality-control samples are required. The World Health Organization stresses validated techniques and contaminant evaluation as the key elements of herbal-material quality control [4]. Alkali metals (Na and K) are typically determined by flame photometry, and many trace metals can be determined by element-specific atomic absorption spectrometry with proper control of digestion and calibration. Past studies in Al-Jabal Al-Akhdar have indicated that leaves and stems of locally growing medicinal plants vary in terms of their macro- and trace-element contents [5-42]. The same studies point out that there is no species-wide uniformity of organ-specific patterns. Nevertheless, unbiased data on comparisons of Salvia with Ziziphus, thyme, and Artemisia under the same sampling and analytical parameters are scarce.
It is practical to establish a local baseline to facilitate future ecological, nutritional and quality-control studies, especially when parts of plants are dissected and utilized independently. Thus, the objective of the current research was to identify Na, K, Fe, Zn, Cu and Mn in the leaves and stems of four medicinal plants growing at Al-Jabal Al-Akhdar and to characterize the difference between the species and the organs. The research also aimed at comparing the observed trends with the reports in the region. The analysis itself is descriptive: unless biological and analytical replication has been documented, it is not testing the statistical significance or setting safe consumption limits.

Methods

Sampling

During spring 2024, plant samples were taken in the Al-Jabal Al-Akhdar area of Libya. The samples were washed using distilled water and were dried at room temperature over two weeks. Leaves and stems were further torn apart to be analyzed.

Samples preparation

About 0.5 g of each sample was moved to a glass beaker. A number of mL of distilled water was added (25 mL) and concentrated nitric acid (5 mL), and the mixture was then heated on a hot plate for 2 h until it was almost dry. Following filtration, the volume of the filtrate was quantitatively transferred into a 100 mL volumetric flask and diluted to volume with distilled water.

Determination of Minerals and Metals:

A JENWAY flame photometer was used to measure sodium and potassium, according to the general procedures mentioned in references [60-70]. A Thermo atomic absorption spectrometer in the Central Laboratory, Faculty of Science, Omar Al-Mukhtar University was used to measure iron, zinc, copper, and manganese.

Results

The results of the measurements are shown in Tables 1-4. Na and K values are given in mg/g, but Fe, Zn, Cu and Mn values are given in µg/g. The mean and standard deviation shown in each table describe the difference between the four plant species; they do not represent replicate-based estimates for individual species. Based on this, the following description justifies concentration patterns without any claim of statistical significance.
Minerals

Table 1. Sodium and potassium concentrations (mg/g) in leaves of the studied plants.

Plant Species	Sodium	Potassium
Salvia	1.15	5.57
Ziziphus	0.28	5.14
Thyme	0.87	3.75
Artemisia	2.84	5.49
Average	1.28	4.98
±SD	1.09	0.84

Leaf Na ranged from 0.28 mg/g in Ziziphus to 2.84 mg/g in Artemisia, with an across-species mean of 1.28 ± 1.09 mg/g. Leaf K was higher than Na in all four plants and varied from 3.75 mg/g in thyme to 5.57 mg/g in Salvia (mean 4.98 ± 0.84 mg/g). Artemisia also contained comparatively high K (5.49 mg/g), close to the Salvia value.

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Table 2. Sodium and potassium concentrations (mg/g) in stems of the studied plants.

Plant Species	Sodium	Potassium
Salvia	0.98	4.57
Ziziphus	0.48	6.32
Thyme	0.58	4.17
Artemisia	1.55	6.53
Average	0.897	5.39
±SD	0.485	1.20

In stems, Na concentrations ranged from 0.48 mg/g in Ziziphus to 1.55 mg/g in Artemisia (mean 0.897 ± 0.485 mg/g). Potassium again exceeded Na in every species, ranging from 4.17 mg/g in thyme to 6.53 mg/g in Artemisia, with a mean of 5.39 ± 1.20 mg/g. The stem K concentration was also high in Ziziphus (6.32 mg/g).

Table 3. Iron, zinc, copper, and manganese concentrations (μg/g) in leaves of the studied plants.

Plant Species	Iron	Zinc	Copper	Manganese
Salvia	0.48	17.29	5.50	14.31
Ziziphus	0.07	18.87	4.43	40.60
Thyme	0.76	9.22	4.64	20.67
Artemisia	0.69	37.07	15.09	66.39
Average	0.50	20.61	7.41	35.49
±SD	0.31	11.75	5.137	23.44

Trace-element concentrations in leaves followed the mean order Mn (35.49 ± 23.44 μg/g) > Zn (20.61 ± 11.75 μg/g) > Cu (7.41 ± 5.13 μg/g) > Fe (0.50 ± 0.31 μg/g). Artemisia leaves had the highest Zn, Cu, and Mn concentrations (37.07, 15.09, and 66.39 μg/g, respectively), while thyme recorded the highest Fe value (0.76 μg/g). Ziziphus had the lowest leaf Fe concentration (0.07 μg/g), and thyme had the lowest leaf Zn value (9.22 μg/g).

Table 4. Iron, zinc, copper, and manganese concentrations (μg/g) in stems of the studied plants.

Plant Species	Iron	Zinc	Copper	Manganese
Salvia	0.12	12.43	4.41	8.35
Ziziphus	0.10	12.66	4.77	77.50
Thyme	1.02	12.50	5.76	26.44
Artemisia	0.67	18.27	9.21	41.16
Average	0.47	13.96	6.03	38.36
±SD	0.44	2.87	2.190	29.33

The same overall order occurred in stems: Mn (38.36 ± 29.33 μg/g) > Zn (13.96 ± 2.87 μg/g) > Cu (6.03 ± 2.19 μg/g) > Fe (0.47 ± 0.44 μg/g). Ziziphus stems contained the highest Mn concentration (77.50 μg/g), whereas Artemisia stems had the highest Zn and Cu values (18.27 and 9.21 μg/g). Thyme stems contained the highest Fe concentration (1.02 μg/g). Salvia stems showed the lowest Mn value (8.35 μg/g).

Comparison of organ means showed that leaves contained 43.2% more Na, 47.6% more Zn, and 22.8% more Cu than stems. Mean Fe was similar between organs (0.50 versus 0.47 μg/g), whereas mean K and Mn were modestly higher in stems by approximately 7.6% and 7.5%, respectively. These mean patterns concealed species-specific differences: all measured elements were higher in Salvia leaves than stems, while Ziziphus and thyme showed several higher stem values. Artemisia generally accumulated Na and trace elements in leaves, but its stem K concentration was higher than its leaf value. In Libya, different studies used different chemical methods and instruments for estimating chemical constituents in soils, plants, rocks, and marine samples; these studies showed the effect of selected methods of analysis on accuracy and validation of these methods [43-110].

Discussion

The current data display the following two patterns: K was the most common measured macro element, and the most common measured trace elements were Mn and Zn. An existing abundance of K above Na is physiologically plausible due to the relatively high concentrations of K needed for enzyme activation, osmotic regulation, and stomatal function, whereas Na is not a universal essential element for higher plants [2]. However, the physiological demand cannot be used to explain

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tissue concentrations. The quantity held in a harvested organ depends on soil availability, salinity, moisture, rhizosphere chemistry and species-specific transport [2,3].
The relative dispersion of the leaf K data was lower than that of the sodium, indicating that potassium was less different in the leaf samples. Artemisia contained the highest Na concentration in both organs and Ziziphus contained lowest in leaves and stems. This contrast could be attributed to variations in ion selectivity or response to habitat, but the study did not quantify soil chemistry or salinity; thus, a specific mechanism cannot be attributed. The increased average K in stems was mainly due to Artemisia and Ziziphus; otherwise, Salvia exhibited the reverse trend. Such species-specific behavior justifies the treatment of leaves and stems as separate units of analysis, but not as a synthesis of aerial components.
Of particular informative use is the local comparison. A previous Al-Jabal Al-Akhdar study had a report of K values of 9.21 µg/g/ thyme leaves and 4.01 µg/g/ stems, and 7.00 and 3.11 µg/g for Salvia leaves and stems, respectively [33]. The current tables provide K in mg/g and provide 3.75 and 4.17 mg/g of thyme and 5.57 and 4.57 mg/g of Salvia. Direct numerical comparison should be approached with caution because the earlier article was inconsistent in using unit language throughout its text and tables and a different method of digestion was used. Nonetheless, both articles demonstrate that there is a significant organ- and species-related variance and support the necessity to present units and analysis guidelines in a clear manner.
Zn, Cu, and Mn were preferentially accumulated in Artemisia leaves, implying that these elements are either highly retained or translocated to photosynthetically active tissue. Many metalloenzymes are supported by Zinc and Cu, Mn is involved in the oxygen-evolving complex of photosystem II and antioxidant metabolism [2]. The accumulation of these micronutrients in leaves can be described by this functional context, but does not demonstrate a nutritional benefit. The bioavailability to humans is dependent on chemical form, processing, extraction efficiency, serving size and interactions with plant constituents. The data hence are a description of total concentrations in digested material and not the proportion absorbed following consumption.
The highest variations were registered by manganese, especially in stems. The 77.50 µg/g of the Ziziphus stems was almost nine times the Salvia stem value. Artemisia on the other hand, contained the most Mn in leaves. This inversion means that the distribution of the organs is element- and species-specific. It can be caused by variations in xylem transport, sequestration, age of leaves, or local soil availability [2,3]. The loosely attached material could have been washed away by distilled water, so environmental deposition might have contributed to the values of leaves. Taking source confirmation would involve paired soil analysis and calculation of soil-to-plant transfer factors.
Special care is needed with the Fe results. Leaves concentrations of 0.07-0.76 µg/g and Stem concentrations of 0.10-1.02 µg/g are significantly lower than those commonly reported with medicinal plants. An example is a Jordanian study of Salvia officinalis that discovered the levels of Fe of 834.5-1743 mg/kg along with Zn of 27.80-42.72 mg/kg and Mn of 26.10-53.70 mg/kg [5]. Part of this contrast could be due to differences in species, soil, climate, organ composition, and sample preparation. But the size of the Fe anomaly also leads to the need to check the reported unit, dilution factor, instrument calibration, and the completeness of digestion prior to publication as well.
The present Zn results agree more closely with the broad range reported in regional medicinal-plant studies. Salvia leaves Zn (17.29 µg/g) was about twice the value reported in the earlier Al-Jabal Al-Akhdar study (8.91 µg/g), while thyme leaves Zn (9.22 µg/g) was close to its previous value (10.40 µg/g) [33]. Stem Zn was higher in the present samples for both plants. These differences are plausible because trace-element concentrations respond to site, season, developmental stage, and geochemical conditions [3,5]. They may also reflect analytical differences, so comparisons across studies are most useful for identifying general patterns rather than declaring one population intrinsically richer.
Quality-control-wise, the elements measured in this work are key nutrients other than the major toxic contaminants that are typically of interest in herbal-safety evaluations. There were no Pb, Cd, As, or Hg data and no intake scenario was considered. It would be scientifically inappropriate to come to any conclusion about whether these plant materials are safe or unsafe for medicinal use only using the present tables. WHO guidelines suggest proven analytical techniques and clear evaluation of the pertinent contaminants on herbal substances [4]. Future work should include toxic elements, dry-weight reporting with corrections on the basis of moisture, and the indication of a direct relationship between the concentration measured and a realistic exposure.

Conclusion

The majority of trace elements measured were Mn and Zn, with potassium prevailing over sodium in all the examined leaves and stems. The most enriched elements in Artemisia were the widest, though Ziziphus stems had the highest concentration of Mn and thyme stems had the highest concentration of Fe. The organ-specific differences indicate that leaves and stems ought to be considered separately. Such findings are descriptive, and this needs to be confirmed by replicated sampling and complete analytical validation before nutritional or safety conclusions can be drawn.

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