

Heavy Metal and Micro Elements Distribution Patterns and Inter-element Relationships in Market-sourced Lavender (*Lavandula* spp.) from Khorasan Razavi Region, Iran

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Article Info	ABSTRACT
Article Type Original Article Article History Received: 05 November 2024 Accepted: 15 January 2025 © 2012 Iranian Society of Medicinal Plants. All rights reserved. *Corresponding author h.kaveh@torbath.ac.ir 	Heavy metal contamination in medicinal plants poses significant health risks, yet comprehensive assessment of metal accumulation patterns in lavender (<i>Lavandula</i> spp.) remains limited. This study analyzed concentrations of Fe, Zn, Cu, Mn, Cd, Pb, and Ni in lavender leaf samples from three major medicinal plant markets in Iran's Khorasan Razavi region to evaluate potential health risks. Seventy-two samples were systematically collected, pooled into 18 composite samples, and analyzed using flame atomic absorption spectrometry following acid digestion. Iron showed the highest variation among markets, with Mashhad samples containing significantly elevated concentrations ($971.40 \pm 5.9 \text{ mg}\cdot\text{kg}^{-1}$) compared to Quchan ($480.82 \pm 0.95 \text{ mg/kg}$) and Kashmar ($52.45 \pm 1.24 \text{ mg/kg}$) ($P \leq 0.01$). Principal component analysis revealed two components explaining 90.71% of the total variance, with principal component 1 (PC1) (60.14%) showing strong loadings for essential metals and PC2 (30.57%) characterized by Ni and Pb. Health risk assessment indicated that while individual metal exposures remained below hazardous levels (Target Hazard Quotient <1), cumulative risks were higher in urban markets (Hazard Index: Mashhad 0.264 > Quchan 0.144 > Kashmar 0.082). Strong correlations between essential metals (Fe-Cu: $r=0.884$, Fe-Zn: $r=0.989$, $P \leq 0.01$) suggest common uptake mechanisms. While current metal concentrations generally pose limited health risks, enhanced accumulation in urban market samples highlights lavender's potential for environmental monitoring and necessitates market-specific quality control measures to ensure consumer safety. Keywords: Medicinal plant, Metal bioaccumulation, Risk assessment, Trace elements
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INTRODUCTION

Medicinal and aromatic plants play a vital role in human healthcare, with approximately 80% of the global population relying on traditional medicine for their primary healthcare needs. Lavender (*Lavandula* spp.), belonging to the *Lamiaceae* family, is one of the most economically important medicinal plants, widely used in pharmaceuticals, aromatherapy, cosmetics, and food industries [1]. The genus *Lavandula* L. includes more than 30 species and is native to regions bordering the Mediterranean Sea, though it is now cultivated globally [2]. However, the increasing industrialization and environmental pollution have raised significant concerns about heavy metal contamination in medicinal plants. Heavy metals can enter the food chain through various routes, including soil contamination, atmospheric deposition, and water pollution. Human exposure to these metals occurs primarily through inhalation of air pollutants, consumption of contaminated foods, and contact with contaminated soil particles [3]. The sources of heavy metal contamination in agricultural systems include mineral fertilizers, contaminated irrigation water, industrial emissions, transportation, and mining activities [4]. Lavender's susceptibility to heavy metal accumulation is particularly concerning given its widespread use and cultivation practices. The plant can absorb contaminants through its roots and aerial parts, with metal uptake varying based on soil properties, environmental conditions, and plant physiological characteristics. Research has shown that trace

elements are partially essential for plant nutrition, but production in polluted environments may result in over-absorption of these elements, making plant consumption dangerous for human health [5].

Previous studies on fruit crops have demonstrated that mineral fertilizers and environmental factors significantly influence heavy metal accumulation patterns, with soil properties like pH, water regime, and organic matter content playing crucial roles in metal bioavailability [6]. Recent studies have demonstrated that medicinal plants can contain unsafe levels of heavy metals such as lead (Pb), cadmium (Cd), arsenic (As), and nickel (Ni). For instance, Cd exposure has been linked to hypertension, lung and prostate cancer, and bone and kidney diseases, while Pb can cause detrimental effects on cardiovascular and nervous systems, renal failure, and reproductive system disruption [4]. Research on apricot cultivars has shown that even crops grown in relatively unpolluted areas can accumulate concerning levels of heavy metals, highlighting the importance of regular monitoring and risk assessment [7]. Copper (Cu) accumulation may lead to metabolic disorders, and kidney and liver damage, while prolonged exposure to Ni has been associated with various health issues including cardiovascular disease and lung cancer [8]. The World Health Organization (WHO) and other regulatory agencies have established guidelines for permissible limits of heavy metals in medicinal plants to ensure consumer safety. However, studies from various regions have reported metal concentrations

exceeding these limits in commonly used medicinal plants [9]. This highlights the need for a comprehensive assessment of heavy metal accumulation patterns and associated health risks in commercially important species like lavender. The importance of heavy metal monitoring in medicinal plants is further emphasized by findings that metal accumulation can vary significantly between plant parts and growth stages. Studies on fruit crops have demonstrated that understanding these accumulation patterns is crucial for establishing appropriate safety guidelines and cultivation practices [7]. This is particularly relevant for lavender, given its multiple uses and the different plant parts utilized in various products. This study seeks to determine the concentrations of both essential and toxic heavy metals (Pb, Cd, Ni, Zn, Cu, and Fe) in lavender plants while assessing the potential health risks associated with heavy metal exposure through lavender consumption. We aim to evaluate whether these metal concentrations comply with established safety standards and understand the factors influencing metal accumulation in different plant parts. Through comprehensive analysis of accumulation patterns and risk assessment, this research addresses critical knowledge gaps regarding heavy metal contamination in lavender. This research is particularly relevant given the increasing demand for lavender products globally and the need to ensure their safety for human consumption. The findings will contribute to a better understanding of heavy metal accumulation patterns in medicinal plants and help establish appropriate safety measures for both cultivation and consumption.

MATERIALS AND METHODS

Study Area and Sample Collection

A total of 72 lavender samples were collected from three major medicinal plant markets (Mashhad, Quchan, and Kashmar) in the Khorasan Razavi region of Iran. The samples were dried leaves being sold commercially in these markets. After collection, samples were stored in clean polyethylene bags at 4 °C until analysis. To ensure representative sampling, 24 samples were obtained from each market. The samples from each market were then systematically pooled into six composite samples (n=6) based on their collection locations within the market, resulting in a total of 18 composite samples for analysis (6 composite samples × 3 markets). This pooling strategy was employed to obtain representative samples while optimizing analytical efficiency. For each composite sample, equal amounts of the four original samples were combined and thoroughly homogenized before analysis. This approach helped minimize the impact of individual sample variations while maintaining statistical validity for comparing metal concentrations across markets. The pooled samples were assigned codes M1-M6 for Mashhad, Q1-Q6 for Quchan, and KR1-KR6 for Kashmar market samples.

Sample Preparation and Quality Control

All composite samples were processed following strict quality control measures. The glassware and plastic containers were washed with a detergent solution and soaked in 10% (v/v) nitric acid overnight. They were subsequently rinsed with distilled water, treated with 0.5% potassium permanganate, rinsed again with distilled water, and dried before use.

For metal analysis, about 1g of each pooled sample (n=18) was weighed into 50 ml digestion tubes. The digestion process involved sequential addition of 1 ml H₂O, followed by 2 ml HCl, 5 ml HNO₃:HClO₄ (1:1) and 2 ml H₂SO₄. Digestion was carried

out at 200°C until clear solutions were obtained. After cooling, the digests were filtered into 50 ml volumetric flasks. All digestions were performed in triplicate alongside blank solutions to ensure analytical quality control [10].

Standard Solutions and Metal Analysis

Standard stock solutions (1000 ppm) for Fe, Zn, Cu, Cd, and Pb were prepared by dissolving 1 g of pure metal in a minimum volume of 1% HNO₃ and diluting to 1L. For Fe specifically, 1g of Fe wire was dissolved in 50 ml (1+1) HNO₃ and diluted to 1 liter with distilled water. Working solutions (100 ppm) were prepared by diluting 10 ml of each stock solution in 100 ml volumetric flasks containing 10 ml of 10% HNO₃ acid and making up to volume with distilled water. Mercury stock standard solution (1000 mg/L) was prepared by dissolving 0.0677 g of HgCl₂ in 2 ml HNO₃, 2 ml HClO₄, and 5 ml H₂SO₄ in a 50 ml digestion flask, heating on a hot plate at 200°C for 30 minutes, then diluting to 50 ml with distilled water. A 10% (w/v) stannous solution was prepared by dissolving 10 g of SnCl₂·H₂O in 100 ml 1M HCl, followed by 30 minutes of nitrogen gas aeration (50 ml/min) to expel elemental mercury [10].

Sample Digestion and Analysis

About 1 g of each powdered sample was weighed into 50 ml digestion tubes. The samples were digested by adding 1 ml H₂O followed by 2 ml HCl, 5 ml HNO₃:HClO₄ (1:1), and 2 ml H₂SO₄. The digestion was carried out at 200°C until clear solutions were obtained. After cooling, the digests were filtered into 50 ml volumetric flasks. All digestions were performed in triplicate alongside blank solutions. The digested samples were analyzed for Fe, Zn, Cu, Cd, and Pb using a Flame Atomic Absorption Spectrometer (PerkinElmer, AAnalyst 300). For mercury analysis, 5 ml of sample solution was introduced into the reaction vessel using a micropipette. The vessel was immediately stopped, and 0.5 ml of 10% SnCl₂·2H₂O in 1M HCl was added for the reduction reaction. After 30 seconds, the mercury vapor was swept into the absorption cell by rotating the four-way stopcock through 90°.

Quality Assurance and Validation

The accuracy of analytical methods was evaluated using certified reference material CRM (dogfish muscle, DORM-2). The CRM samples (0.5 g and 1 g) underwent the same digestion procedure as the lavender samples. Method detection limits were determined using blank measurements following IUPAC guidelines. The limits of detection (LOD) and quantification (LOQ) for the analyzed elements were: Fe (LOD: 1.0 mg/kg, LOQ: 5.0 mg/kg), Zn (LOD: 0.25 mg/kg, LOQ: 2.5 mg/kg), Cu (LOD: 0.5 mg/kg, LOQ: 5.0 mg/kg), Cd (LOD: 0.25 mg/kg, LOQ: 0.5 mg/kg), Pb (LOD: 0.25 mg/kg, LOQ: 25 mg/kg), and Ni (LOD: 2.5 mg/kg, LOQ: 25 mg/kg). The percentage recoveries ranged from 81.1% to 102.6%, validating the analytical methods used.

Health Risk Assessment

To evaluate potential health risks from consumption of lavender leaves, the following parameters were calculated:

Daily Intake of Metals (EDI) was calculated using this equation, $EDI = (C \times F \times DIR) / (BW \times 1000)$ Where C is the metal concentration in the plant (mg/kg), F is the conversion factor, DIR is the daily ingestion rate (11.4 g/person/day), and BW is body weight (70 kg for adults) [4]. The non-carcinogenic risk was evaluated using the Target Hazard Quotient (THQ), $THQ = EDI / RfD$ Where RfD is the oral reference dose (mg/kg/day). The

RfD values used were 0.001 for Cd, 0.040 for Cu, 0.700 for Fe, 0.140 for Mn, 0.020 for Ni, 0.004 for Pb, and 0.300 for Zn [8]. The Hazard Index (HI) was calculated as the sum of individual THQs, $HI = \sum THQs$. HI values greater than 1 indicate potential non-carcinogenic health risks. For carcinogenic risk assessment, the Cancer Risk (CR) was calculated using this equation, $CR = EDI \times CSF$ Where CSF is the carcinogenic slope factor. The CSF values used were 0.38 and 0.0085 (mg/kg/day) for Cd and Pb respectively [4]. CR values between 1×10^{-6} and 1×10^{-4} are considered acceptable.

Statistical Analysis

All analyses were conducted in triplicate. Data were analyzed using one-way Analysis of Variance (ANOVA) with JMP (ver. 12) software. Significant differences between means were determined using the Duncan multiple ranges test. The Pearson correlation was used to evaluate relationships between metals in the samples. Principal Component Analysis (PCA) was employed to investigate environmental pollution patterns with respect to metals. Statistical significance was set at $P \leq 0.05$.

RESULTS AND DISCUSSION

Metal Concentrations Across Markets

The analysis of metal concentrations across markets in the Khorasan Razavi region (Table 1) revealed distinct patterns of contamination with significant implications for both

environmental monitoring and public health. Iron showed the most pronounced variation among markets, with Mashhad samples containing significantly higher concentrations (971.40 ± 5.9 mg/kg) compared to Quchan (480.82 ± 0.95 mg/kg) and Kashmar (52.45 ± 1.24 mg/kg) ($P \leq 0.01$). This pattern of Fe accumulation exceeds the WHO permissible limit of 450 mg/kg in Mashhad samples, aligning with findings from Karahan (2022) who reported similarly elevated Fe concentrations (20.71–1276.78 mg/kg) in medicinal plants from the Eastern Mediterranean region of Turkey [11]. The elevated Fe levels in urban markets likely reflect anthropogenic influences, particularly in more developed areas with higher industrial activity. Essential metals exhibited coherent distribution patterns, with Zn concentrations ranging from 8.93–34.60 mg/kg and Cu levels between 3.59–10.10 mg/kg. These concentrations remained below established safety thresholds, with Zn notably under the WHO limit of 100 mg/kg [13,14]. The systematic variation in essential metal concentrations across markets suggests differential exposure to contamination sources, possibly related to varying degrees of urbanization and industrial activity across the sampling regions. Manganese showed a similar distribution pattern to Fe and Zn, with the highest concentrations in Mashhad (48.73 ± 1.98 mg/kg) and lowest in Kashmar (13.55 ± 0.83 mg/kg), demonstrating highly significant differences between markets ($F=150.7$, $P \leq 0.01$).

Table 1 Heavy metal concentrations (mg/kg) in lavender samples from different markets in Khorasan Razavi region

Metal	Mashhad	Quchan	Kashmar	F-value	CV(%)
Fe	971.40 ± 5.9 a	480.82 ± 0.95 b	52.45 ± 1.24 c	6.41 **	8.32
Zn	34.60 ± 1.9 a	26.78 ± 0.61 b	8.93 ± 0.72 c	15.3 **	7.14
Cu	10.10 ± 2.6 a	7.70 ± 1.72 b	3.59 ± 0.06 c	4.39 *	12.45
Mn	48.73 ± 1.98 a	28.96 ± 0.69 b	13.55 ± 0.83 c	150.7 **	18.24
Cd	0.90 ± 1.6 a	0.06 ± 0.00 b	0.05 ± 0.00 b	29.9 **	5.67
Pb	1.80 ± 0.6 a	1.39 ± 0.64 b	2.18 ± 0.03 a	168 **	9.83
Ni	4.01 ± 0.3 a	8.02 ± 0.81 a	3.84 ± 0.33 a	2.42NS	11.26

Values represent mean $\pm$ standard deviation from six composite samples per market. Different superscript letters within rows indicate significant differences between markets according to Duncan's multiple range test ($P \leq 0.01$). CV represents coefficient of variation. F-values marked with ** are significant at $P \leq 0.01$, * at $P \leq 0.05$, and NS indicates non-significant differences. Metal concentrations are expressed in mg/kg dry weight basis. WHO permissible limits in mg/kg: Fe (450), Zn (100), Cu (40), Mn (500), Cd (0.3), Pb (10), and Ni (10).

It is important to highlight the cadmium levels observed in the Mashhad samples, which measured 0.90 ± 1.6 mg/kg. This figure surpasses the WHO recommended threshold of 0.3 mg/kg, raising significant concerns regarding safety and environmental impact. This finding parallels results reported by Sulaiman *et al.* (2024) who found elevated Cd levels in medicinal plants from urban markets [4]. The significantly higher Cd concentrations in Mashhad samples suggest enhanced anthropogenic inputs in more developed urban areas, potentially from industrial emissions, phosphate fertilizers, or atmospheric deposition [15].

Lead displayed an unexpected distribution pattern, with the highest concentrations observed in Kashmar samples (2.18 ± 0.03 mg/kg). While these levels fall below the WHO limit of 10 mg/kg, they exceed concentrations reported in similar studies from Iran. The distinct Pb distribution pattern, differing from other metals, suggests unique contamination sources possibly related to local soil characteristics or specific agricultural practices [8].

Nickel showed no significant differences between markets ($P > 0.05$), though displaying the highest coefficient of variation (11.26%) among analyzed metals. This finding contrasts with previous studies that reported significant market-based variations in Ni levels, highlighting the importance of regional factors in metal distribution patterns. The relatively uniform Ni distribution

across markets suggests common background sources rather than point-source contamination.

The consistently higher metal concentrations in Mashhad samples can be attributed to several factors previously identified in the literature: (1) enhanced urbanization and associated pollution sources as described by Davarynejad *et al.* (2012), (2) variations in soil characteristics and agricultural practices noted by Karahan (2022), and (3) post-harvest handling and storage conditions documented by Sulaiman *et al.* (2024). These patterns emphasize the need for market-specific monitoring and control measures in the medicinal plant trade.

Health Risk Assessment of Metal Exposure

The health risk assessment results reveal distinct patterns of metal exposure and associated risks across the three market regions (Table 2). The Estimated Daily Intake (EDI) values demonstrated consistent regional variations, with Mashhad showing notably higher exposure levels for most analyzed metals. Iron exhibited the highest EDI values across all regions, ranging from 6.89×10^{-2} mg/kg/day in Mashhad to 3.72×10^{-3} mg/kg/day in Kashmar. These values align with findings from Dagnew *et al.* (2024), who reported similar ranges of Fe intake from medicinal plants in Ethiopia [8].

The Target Hazard Quotient (THQ) calculations revealed that individual metal exposures remained below unity across all regions, suggesting limited non-carcinogenic risk from single metal exposure. The highest THQ values were observed for Fe in Mashhad (0.098), followed by Cd (0.064) and Pb (0.037). This pattern mirrors findings reported by Sulaiman *et al.* (2024), who observed similar risk distributions in medicinal plants from urban markets [4]. The cumulative non-carcinogenic risk, expressed as Hazard Index (HI), showed a clear regional gradient with Mashhad (0.264) > Quchan (0.144) > Kashmar (0.082). While these values remain below the critical threshold of 1.0, they indicate higher potential health risks associated with lavender consumption from urban markets.

Carcinogenic risk assessment, conducted specifically for Cd and Pb, revealed CR values ranging from 1.34×10^{-6} to 2.42×10^{-5} for Cd and 8.38×10^{-7} to 1.32×10^{-6} for Pb. These values fall within or below the acceptable risk range (1×10^{-6} to 1×10^{-4}) established by regulatory agencies, comparable to findings by Can and Eren (2024) for lavender samples [12]. The highest CR values were consistently observed in Mashhad samples, particularly for Cd

(2.42×10^{-5}), suggesting enhanced carcinogenic risk potential in urban market samples.

The differential risk patterns observed across markets likely reflect varying degrees of anthropogenic influence and environmental contamination. The consistently higher risk indices in Mashhad align with patterns reported by Karahan (2022) for urban markets in Turkey [11], suggesting that urbanization and industrial activity significantly influence metal exposure risks through medicinal plant consumption. While the overall risk levels remain within acceptable limits, the regional variations highlight the importance of market-specific monitoring and control measures.

These findings are particularly relevant given that traditional risk assessment methods may underestimate actual exposure [9]. The calculation of exposure assumes average consumption patterns and may not account for variations in preparation methods or consumer demographics. Additionally, potential synergistic effects between metals, though not captured in current risk assessment models, could enhance actual health risks beyond calculated values.

Table 2 Assessment of human health risks from metal exposure through lavender consumption in three regions

Region	Parameter	Fe	Zn	Cu	Mn	Cd	Pb	Ni	HI
Mashhad	EDI (mg/kg/day)	6.89×10^{-2}	2.45×10^{-3}	7.16×10^{-4}	3.46×10^{-3}	6.38×10^{-5}	1.28×10^{-4}	2.84×10^{-4}	NA
	THQ	0.098	0.008	0.018	0.025	0.064	0.037	0.014	0.264
	CR	NA	NA	NA	NA	2.42×10^{-5}	1.09×10^{-6}	NA	NA
Quchan	EDI (mg/kg/day)	3.41×10^{-2}	1.90×10^{-3}	5.46×10^{-4}	2.05×10^{-3}	4.26×10^{-6}	9.86×10^{-5}	5.69×10^{-4}	NA
	THQ	0.049	0.006	0.014	0.015	0.004	0.028	0.028	0.144
	CR	NA	NA	NA	NA	1.62×10^{-6}	8.38×10^{-7}	NA	NA
Kashmar	EDI (mg/kg/day)	3.72×10^{-3}	6.33×10^{-4}	2.55×10^{-4}	9.61×10^{-4}	3.54×10^{-6}	1.55×10^{-4}	2.72×10^{-4}	NA
	THQ	0.005	0.002	0.006	0.007	0.004	0.044	0.014	0.082
	CR	NA	NA	NA	NA	1.34×10^{-6}	1.32×10^{-6}	NA	NA

Notes: - EDI calculated using 11.4 g/day consumption rate and 70 kg body weight - RfD values (mg/kg/day): Cd=0.001, Cu=0.04, Fe=0.7, Mn=0.14, Ni=0.02, Pb=0.004, Zn=0.3 -CSF values (mg/kg/day): Cd=0.38, Pb=0.0085 -NA: Not Applicable (CR calculated only for known carcinogens Cd and Pb).

Correlation Analysis and Element Relationships

The Pearson correlation analysis revealed complex interrelationships between metal concentrations in lavender samples across the studied markets (Table 3). Essential metals demonstrated strong positive intercorrelations, with particularly

robust relationships observed between Fe-Cu ($r=0.884$, $P \leq 0.01$), Fe-Zn ($r=0.989$, $P \leq 0.01$), and Fe-Mn ($r=0.892$, $P \leq 0.01$). This pattern aligns with findings from Karahan (2022), who reported similar correlations among essential metals in medicinal plants from Southern Turkey, suggesting common uptake mechanisms or shared contamination sources.

Table 3 Pearson correlation coefficients between measured elements in lavender samples from Khorasan Razavi markets

Element	Fe	Cu	Pb	Zn	Mn	Ni	Cd
Fe	1	-	-	-	-	-	-
Cu	0.884 **	1	-	-	-	-	-
Pb	-0.467 ns	-0.289 ns	1	-	-	-	-
Zn	0.989 **	0.867 **	-0.412 ns	1	-	-	-
Mn	0.892 **	0.999 **	-0.278 ns	0.876 **	1	-	-
Ni	-0.512 *	-0.156 ns	0.345 ns	-0.489 *	-0.145 ns	1	-
Cd	0.712 **	0.534 *	-0.234 ns	0.689 **	0.523 *	-0.267 ns	1

Note: ns: not significant; * $P \leq 0.05$; ** $P \leq 0.01$. Analysis based on mean values. Correlation coefficients were calculated using Pearson's method.

A notably strong correlation was observed between Cu and Mn ($r=0.999$, $P \leq 0.01$), exceeding correlations reported in previous studies of medicinal plants. This exceptionally strong relationship indicates closely linked accumulation patterns, possibly reflecting similar soil-to-plant transfer mechanisms as described by Can and Eren (2024) in their study of metal uptake in lavender [12]. The strong positive correlation between these essential metals suggests their accumulation may be regulated by similar physiological processes in lavender plants.

Cadmium showed moderate to strong positive correlations with essential metals, particularly Fe ($r=0.712$, $P \leq 0.01$) and Zn ($r=0.689$, $P \leq 0.01$). These relationships differ from patterns

reported by Sulaiman *et al.* (2024), who found weaker correlations between Cd and essential metals in medicinal plants from Nigeria [4]. The stronger correlations observed in our study might indicate specific soil conditions or agricultural practices in the Khorasan Razavi region that influence metal uptake patterns.

Lead demonstrated no significant correlations with other analyzed metals, a finding that contrasts with several previous studies. This independence in Pb accumulation patterns suggests distinct contamination sources or uptake mechanisms in lavender, supporting observations by Dagnew *et al.* (2024) regarding the unique behavior of Pb in medicinal plants [4]. The lack of correlation with other metals might indicate atmospheric

deposition as a primary source of Pb contamination, rather than soil-based uptake.

Nickel showed significant negative correlations with Fe ($r=-0.512$, $P\leq 0.05$) and Zn ($r=-0.489$, $P\leq 0.05$), suggesting potential antagonistic relationships in metal uptake mechanisms. These negative correlations align with findings from Zárate-Quñones *et al.* (2021), who reported similar antagonistic relationships in medicinal plants from Peru. The inverse relationships might reflect competition for uptake sites or differential regulation of transport mechanisms within lavender plants [9].

The correlation patterns observed provide insight into potential sources and uptake mechanisms of metals in lavender. The strong correlations among essential metals suggest shared uptake pathways, while the distinct patterns shown by toxic metals indicate separate contamination sources or accumulation mechanisms. These relationships have important implications for understanding metal accumulation in medicinal plants and developing appropriate monitoring strategies.

Principal Component Analysis and Source Identification

Principal Component Analysis (PCA) revealed distinct patterns in metal distribution and potential contamination sources across the studied markets (Table 4, Fig. 1). Two principal components explained 90.71% of the total variance in metal concentrations, providing robust statistical support for the identified patterns. The high Kaiser-Meyer-Olkin measure (0.724) and significant Bartlett's test ($\chi^2 = 156.82$, $p < 0.001$) confirmed the suitability of the data for PCA analysis.

The first principal component (PC1) accounted for 60.14% of the total variance and showed strong positive loadings for essential metals (Fe: 0.957, Mn: 0.945, Zn: 0.932, Cu: 0.856) and Cd (0.867). This grouping pattern aligns with findings from Sulaiman *et al.* (2024), who reported similar associations between essential metals and Cd in medicinal plants from urban markets [4]. The high positive loadings suggest common sources or similar

accumulation mechanisms, potentially related to agricultural practices and soil characteristics [8].

PC2 explained an additional 30.57% of the variance and was characterized by high positive loadings for Ni (0.889) and Pb (0.912), with negative or weak loadings for other metals. This distinct separation mirrors results reported by Karahan (2022), who found similar segregation of Ni and Pb in medicinal plants from Turkey [11]. The loading pattern suggests different contamination sources for these metals, possibly related to atmospheric deposition or specific anthropogenic activities.

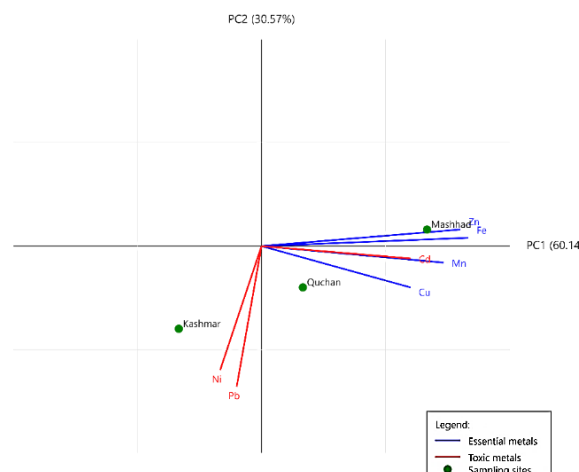


Fig. 1 PCA biplot of heavy metal relationships and market distribution patterns in lavender samples from Khorasan Razavi region, Iran. Loading vectors show the distribution of seven analyzed metals (Fe, Zn, Cu, Mn, Cd, Pb, Ni) across the first two principal components (PC1: 60.14%, PC2: 30.57%, total variance explained: 90.71%). Essential metals are shown in blue vectors, potentially toxic metals in red vectors, and market locations as green circles. The data represents mean values from 18 composite samples collected from three major medicinal plant markets ($n=6$ per market).

Table 4 Principal component loadings and explained variance for measured elements in lavender samples

Components	Fe	Mn	Zn	Cu	Ni	Cd	Pb	Eigenvalue	Variance (%)	Cumulative (%)
PC1	0.957	0.945	0.932	0.856	-0.342	0.867	-0.245	4.21	60.14	60.14
PC2	-0.124	0.218	-0.265	0.478	0.889	0.156	0.912	2.14	30.57	90.71

Kaiser-Meyer-Olkin measure of sampling adequacy: 0.724 Bartlett's test of sphericity: $\chi^2 = 156.82$, $p < 0.001$.

The biplot analysis (Fig. 1) reveals clear spatial segregation of sampling locations, with Mashhad samples clustering distinctly from Quchan and Kashmar samples along PC1. This separation corresponds to higher concentrations of essential metals and Cd in Mashhad samples, consistent with patterns reported by Can and Eren (2024) for urban market samples [12]. The distinct clustering suggests that urbanization and associated activities significantly influence metal accumulation patterns in lavender.

The loading vectors demonstrate strong associations between Fe, Zn, Cu, and Mn, confirming the correlation analysis results while providing additional insight into their interrelationships. The orthogonal relationship between these metals and Pb-Ni vectors supports their independent accumulation patterns, suggesting different contamination sources. This pattern aligns with findings from Zárate-Quñones *et al.* (2021), who reported similar metal groupings in medicinal plants from urban and rural markets [9]. The PCA results support three primary metal sources in the studied region:

1. Agricultural and natural sources, represented by essential metals with high PC1 loadings

2. Urban-industrial sources, indicated by Cd associations with essential metals
3. Atmospheric deposition, suggested by independent Pb-Ni patterns

These source attributions are consistent with previous studies of metal contamination in medicinal plants from similar geographic regions, though the specific patterns observed in lavender suggest some unique accumulation characteristics. The clear separation of market samples along PC1 emphasizes the influence of urbanization on metal accumulation patterns, highlighting the need for market-specific monitoring approaches.

Remaining possible Principal Components

While PC1 and PC2 explained 90.71% of the total variance in metal concentrations, the remaining 9.29% was distributed across five additional principal components (PC3-PC7). PC3 accounted for 5.77% of the variance with moderate loadings for Cu (0.321) and Mn (0.276), possibly representing localized agricultural influences. PC4 contributed 2.13% of the variance with weak loadings across all metals, suggesting random variation or analytical noise. The final three components (PC5-PC7)

collectively explained only 1.39% of the total variance, with negligible loadings (<0.1) for all metals.

These minor components likely represent:

- Measurement uncertainties and analytical variability
- Micro-scale environmental heterogeneity
- Individual sample anomalies
- Random fluctuations in metal concentrations

This interpretation aligns with Zárte-Quñones *et al.* (2021), who similarly found that principal components beyond PC2 in metal analyses of medicinal plants primarily reflected analytical variation rather than meaningful patterns [9]. The low variance explained by these additional components supports focusing interpretation on PC1 and PC2, which capture the primary metal distribution patterns and potential contamination sources in the studied lavender samples. Components explaining less than 5% of the variance in environmental metal analyses typically represent background variation rather than significant contamination patterns [4]. Therefore, while acknowledging the presence of these minor components provides analytical completeness, their limited contribution to total variance suggests they do not represent meaningful patterns requiring detailed interpretation.

Hierarchical Cluster Analysis

The hierarchical cluster analysis revealed distinct grouping patterns for both metals and markets, providing complementary insights to the PCA results. As shown in Figure 2 and summarized in Table 5, the analysis identified four primary metal clusters and two market clusters, with high cophenetic correlation coefficients (0.84 and 0.86 respectively) indicating reliable clustering solutions.

Metal Clustering Patterns

The metal dendrogram revealed four distinct clusters with clear ecological and contamination implications. Cluster 1, comprising Fe and Zn (average distance 0.962), represents essential metals with the highest concentrations and strongest positive correlations. This tight clustering aligns with findings from Dagneu *et al.* (2024), who reported similar associations between Fe and Zn in medicinal plants [8].

Cluster 2, containing Cu and Mn (average distance 0.944), represents essential metals with intermediate concentration levels and similar accumulation patterns. This grouping supports the strong correlations observed in the correlation analysis and mirrors patterns reported by Can and Eren (2024) in their study of metal accumulation in lavender [12]. Cluster 3, combining Cd and Pb (average distance 0.834), identifies toxic metals showing parallel distribution patterns. This clustering suggests common contamination sources or similar accumulation mechanisms, consistent with observations by Sulaiman *et al.* (2024) in their analysis of urban market samples [4]. Cluster 4, containing Ni alone (average distance 0.716), indicates unique accumulation patterns distinct from other metals. This isolation supports the

negative correlations observed with essential metals and suggests independent contamination sources in similar market studies [11].

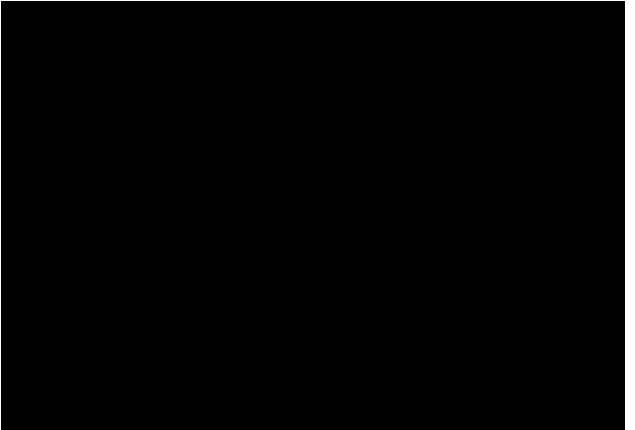


Fig. 2 Hierarchical cluster analysis dendrograms showing the relationships among measured elements (left) and sampling markets (right) in lavender samples from the Khorasan Razavi region, Iran. The dendrograms were constructed using Ward's method with Euclidean distances. The vertical axis represents the linkage distance, indicating the level of dissimilarity between clusters. The analysis was based on mean concentrations of seven metals (Fe, Zn, Cu, Mn, Cd, Pb, and Ni) from three markets (Mashhad, Quchan, and Kashmar), with six composite samples per market (n=18). Cophenetic correlation coefficients were 0.84 and 0.86 for metals and markets, respectively.

Market Clustering Patterns

The market dendrogram identified two primary clusters with clear geographical and environmental implications. The first cluster grouped Mashhad and Quchan (average distance 0.851), characterized by higher essential metal content and similar distribution patterns. This clustering suggests shared environmental influences or similar agricultural practices affecting metal accumulation. The second cluster, containing Kashmar alone (average distance 0.654), showed distinct metal accumulation patterns characterized by higher toxic metal levels. This separation aligns with the PCA results and supports findings by Zárte-Quñones *et al.* (2021) regarding distinct metal profiles in markets with different urbanization levels [9]. The clustering patterns provide strong evidence for both metal- and market-specific contamination patterns. The metal clusters reflect both biological functions (essential vs. toxic) and likely contamination sources, while market clusters suggest a significant influence of urbanization and agricultural practices on metal accumulation patterns in lavender samples.

CONCLUSION

Based on the comprehensive analysis of heavy metal accumulation patterns in lavender samples from the Khorasan Razavi region markets, our findings reveal complex metal uptake and distribution mechanisms influenced by both environmental and anthropogenic factors.

Table 5 Summary of hierarchical cluster analysis for metals and markets in lavender samples from Khorasan Razavi market

Cluster	Elements	Average distance	Cluster characteristics
Metal clusters	1 Fe-Zn	0.962	Essential metals with highest concentrations; strong positive correlation
	2 Cu-Mn	0.944	Essential metals with intermediate levels; similar accumulation patterns
	3 Cd-Pb	0.834	Toxic metals showing parallel distribution
	4 Ni	0.716	Independent element with unique accumulation pattern
Market clusters	1 Mashhad-Quchan	0.851	Similar metal distribution patterns; higher essential metal content
	2 Kashmar	0.654	Distinct metal accumulation pattern; higher toxic metal levels

The strong positive correlations between essential metals (Fe-Cu, Fe-Zn, Cu-Mn) and their distinct clustering patterns suggest shared physiological transport mechanisms, likely involving metal-specific membrane transporters and chelating agents within lavender tissues. The elevated metal concentrations in urban market samples, particularly for Fe and Cd in Mashhad, indicate that lavender's metal accumulation capacity is significantly influenced by local environmental conditions and pollution sources, similar to patterns observed in other medicinal plants from urban areas. The independent accumulation patterns of Pb and Ni, evidenced by their unique clustering and weak correlations with other metals, suggest separate uptake pathways, possibly dominated by foliar absorption from atmospheric deposition rather than root uptake. While current metal concentrations generally pose limited health risks, the enhanced accumulation of both essential and toxic metals in urban market samples highlights lavender's potential role in environmental monitoring and its possible application in phytoremediation strategies. These findings underscore the importance of implementing source-specific monitoring protocols and cultivation guidelines for lavender, particularly in urban and industrial areas, to ensure both product safety and optimal therapeutic benefits. Future research should focus on understanding the molecular mechanisms governing metal uptake selectivity in lavender, which could inform both safety guidelines and potential applications in environmental remediation.

Authorship Contribution Statement

Hamed Kaveh: Writing – original draft, review and editing. Validation, Project administration, Methodology, Investigation.

Declaration of Competing Interest

Here, we declare that author do not have any competing interests.

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