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Chemical Compositions and Some Heavy Metal Contents of Five *Lathyrus* Species (Fabaceae) from Türkiye

Bekir Yildirim[1]

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ABSTRACT

Background: In today's modern world, heavy metal accumulation, one of the consequences of rapid and excessive environmental pollution, is a serious threat to the health of organisms. Many diseases and disorders are linked to the excessive accumulation of heavy metals, defined as metals with a specific gravity greater than 5 g/cm^3. This study aims to investigate the elemental composition of *Lathyrus annuus*, *L. hierosolymitanus*, *L. hirsutus*, *L. undulatus* and *L. tuberosus* species, which have potential for human consumption as food and to determine the concentrations of Na, Mg, K, Ca, Fe, Zn, Mn, Cr, Ni, Cd and Cu, the last seven of which are classified heavy metals.

Methods: Plant materials were obtained from their natural distribution areas and identified according to the Flora of Türkiye. In the chemical analyses, sample preparation was carried out using the wet digestion method in accordance with EPA Method 3015A and ICP-OES measurements were performed according to EPA Method 6010.

Result: The highest concentrations of Ca, Mn, Cd, Zn and Cu were found in *L. hirsutus*, while Na and Fe were most abundant in *L. hierosolymitanus*, K in *L. tuberosus* and Mg in *L. annuus*. In all species, Cr and Ni amounts were found to be lower than 0.008 and 0.006 ppm, respectively. Cd and Cu levels were less than 0.005 and 0.003 ppm, respectively, in all species except *L. hirsutus*. Compared to other species examined in the current study and to literature data, it was found that the aerial parts of *L. hirsutus* contain very high amounts of Mn and Cd.

Key words: Chemical composition, Elements, Heavy metals, *Lathyrus*.

INTRODUCTION

Environmental pollution has significantly increased in the last hundred years due to population growth, the transition to modern agriculture practices, rapid industrialisation and urbanisation. Among the major pollutants, heavy metals, the most important pollution factor, affect all organisms through the food chain, increasing the danger of pollution even more (Öktüren-Asri *et al*., 2007). The consumption of food contaminated with heavy metals poses significant toxicological risks to human health (Mathew and Teenamol, 2019). Heavy metal accumulation and toxicity impair the function of various organs and physiological systems and have been linked to a wide range of diseases and disorders (Mahurpawar, 2015; Ohiagu *et al*., 2022).

Heavy metals, typically defined as elements with densities at least five times greater than that of water, pose significant threats to public health (Özbolat and Tuli, 2016; Ohiagu *et al*., 2022). Although more than 60 heavy metals have been identified, the most commonly encountered and well- known include arsenic (As), cadmium (Cd), chromium (Cr), cobalt (Co), copper (Cu), iron (Fe), lead (Pb), manganese (Mn), mercury (Hg), molybdenum (Mo), nickel (Ni), silver (Ag), tin (Sn) and zinc (Zn). Heavy metals are typically classified into two groups, essential (Cr, Co, Cu, Fe, Mn, Mo, Ni and Zn) and non-essential (As, Cd, Hg, Pb and Sn), according to their effects on biological processes (Saad *et al*., 2014; Özbolat and Tuli, 2016; Dökmeci *et al*., 2019; Ohiagu *et al*., 2022; Soomro *et al*., 2024). Essential heavy metals, which are crucial for life and present in trace amounts in the organism for cellular functions, are involved in the structure of vitamins and hormones and play roles in enzymatic reactions. However, these essential heavy metals become toxic at concentrations exceeding 1-10 ppm (parts per million) (Saad *et al*., 2014; Özbolat and Tuli, 2016). Non-essential heavy metals exert toxic effects irrespective of their concentration in biological systems. In particular, Cd and Hg can be toxic even at very low concentrations of 0.001-0.1 ppm (Özbolat and Tuli, 2016). The provisionally tolerable daily intake of some heavy metals (mg/kg body weight) are presented in Table 1.

Plants have been used by people due to their nutritional and medicinal properties for a long time. The seeds and aerial parts of certain *Lathyrus* L. species are used as sources of human food and animal feed in various regions of the world (Yildirim *et al*., 2023). In addition to the

[1]Department of Plant and Animal Production, Burdur Food Agriculture and Livestock Vocational School, Burdur Mehmet Akif Ersoy University, Burdur, 15030, Türkiye.

Corresponding Author: Bekir Yildirim, Department of Plant and Animal Production, Burdur Food Agriculture and Livestock Vocational School, Burdur Mehmet Akif Ersoy University, Burdur, 15030, Türkiye. Email: byildirim@mehmetakif.edu.tr
ORCID: https://orcid.org/0000-0003-2378-7697

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nutritiousness of Legumes (Fabaceae), which are considered a good source of food, especially in developing countries, it has been reported that some species of the genus *Lathyrus* belonging to this family may be a source of phenolic compounds with strong antioxidant effects (Pastor-Cavada *et al.*, 2009). However, understanding the elemental composition, particularly the levels of toxic heavy metals, of plants with potential bioactivities and nutritional value is important for human health. In Arslan *et al.* (2018) and Yildirim *et al.* (2020), the metal compositions of some *Lathyrus* species have been investigated.

This study aimed to determine the chemical compositions and selected heavy metal concentrations in the aerial parts of *Lathyrus annuus* L., *L. hierosolymitanus* Boiss., *L. hirsutus* L., *L. undulatus* Boiss. and *L. tuberosus* L. and to assess whether there are significant differences among these species. The same analyses were also applied to the tubers of *L. tuberosus* and comparisons were executed.

MATERIALS AND METHODS

This study was conducted from 2023 to 2024 at Food Agriculture and Livestock Vocational School, Burdur Mehmet Akif Ersoy University.

Plant samples and identification

Samples were collected from the natural distribution areas listed below during the flowering periods.

Table 1: Provisionally tolerable daily intake (TDI) of some heavy metals.

Heavy metal	Tolerable daily intake	Reference
Fe	0.8 mg/kg bw*	Asomugha *et al.*, 2016
Zn	0.43 mg/kg bw	Asomugha *et al.*, 2016
Mn	11 mg/day	Deka *et al.*, 2023
Cr	0.003 mg/kg bw	Deka *et al.*, 2023
Ni	0.0028 mg/kg bw	Asomugha *et al.*, 2016
Cd	0.001 mg/kg bw	WHO, 2007

*Body weight (commonly assumed to be a 60 kg adult).

L. annuus: Around the ancient city of Stratonikeia, Yatağan, Muğla, Türkiye.

L. hierosolymitanus: Around the Çamlıca neighborhood, Alanya, Antalya, Türkiye.

L. hirsutus: Eğirdir-Aksu road, 5-6 km from Eğirdir, roadside, Eğirdir, Isparta, Türkiye.

L. undulatus: Kuyucak-Yerkesik road, under the *Pinus brutia* trees, Menteşe, Muğla, Türkiye.

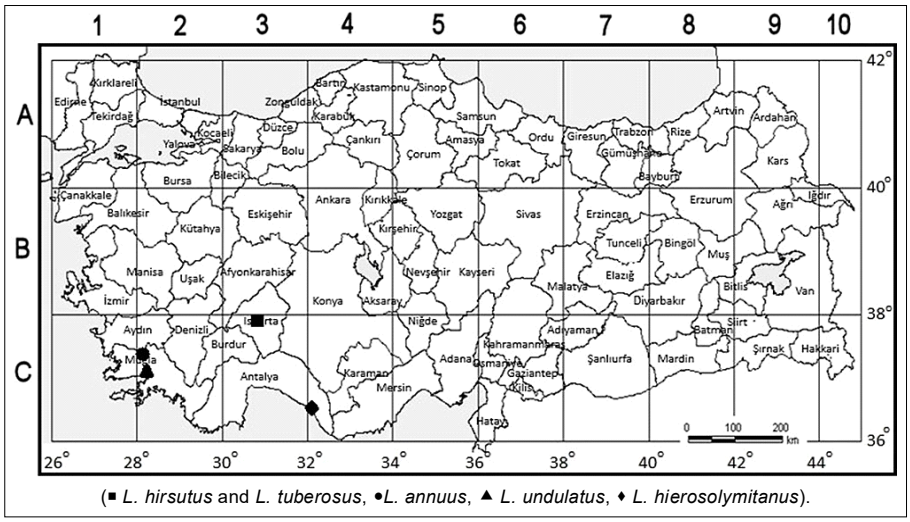
L. tuberosus: Around the entrance of Pazarköy village, Eğirdir, Isparta, Türkiye.

The collection locations are shown in Fig 1. Species identification was carried out using Flora of Türkiye (Davis, 1970).

The entire aerial parts of the samples and the tubers of *L. tuberosus* were separated and dried in a room without sunlight for ten days, then pulverised using a grinder. The abbreviation “tb.” is used to refer to tuber(s) from this point onward throughout the manuscript.

Chemical analyses

Chemical analyses were performed by Suleyman Demirel University, Innovative Technologies Application and Research Center (YETEM). The concentrations of a total of 11 elements, K, Ca, Mg, Na, Fe, Zn, Mn, Cr, Ni, Cd and Cu, were analyzed in the plant samples. The last seven of these elements are classified as heavy metals. Sample preparation was carried out by the wet digestion method by adding 8 ml of HNO_3 + 2 ml of H_2O_2 to 0.5 g of sample using the Milestone brand Ethos One (Fig 2) model microwave sample preparation unit according to EPA method 3015A. The final volume was completed to 20 ml with distilled water. ICP-OES tests were made in accordance with the EPA method 6010 using the Perkin Elmer Optima 5300 DV (Fig 2) under the following conditions. Plasma Gas Flow: 15 L/min, Auxiliary Gas Flow: 0.2 L/min, Nebulizer Gas Flow: 0.6 L/min, Power: 1450 Watts, Torch Cassette Position: -3, Pump Speed: 1.5 mL/minute, Purge: Normal, Resolution: Normal, Integration Time: 10 secondsmin/20 secondsmax, Read Delay: sixty seconds, Replicates: three.



(■ *L. hirsutus* and *L. tuberosus*, •*L. annuus*, ▲ *L. undulatus*, ♦ *L. hierosolymitanus*).

Fig 1: Collection locations of the *Lathyrus* species.

RESULTS AND DISCUSSION

Some *Lathyrus* species have the potential for human consumption due to various attributes such as edibility and medicinal effects. Therefore, in this study, the chemical compositions and certain heavy metal contents of the aerial parts of *L. annuus, L. hierosolymitanus, L. hirsutus, L. undulatus, L. tuberosus* and tubers of *L. tuberosus* (Fig 3) were investigated.

The findings are showcased comparatively in Table 2. All elements in Table 2 are heavy metals except Na, Mg, K and Ca. Among the investigated metals, the highest amounts of Ca, Mn, Cd, Zn and Cu were found in *L. hirsutus*; Na and Fe in *L. hierosolymitanus*; Mg in *L. annuus*; and K in *L. tuberosus*. The lowest amounts of K, Na, Fe and Mg were found in *L. hirsutus*; Ca in *L. tuberosus* (tb.); Zn in *L. tuberosus*; Mn in *L. tuberosus* and tubers of *L. tuberosus*. In all samples, the amounts of Cr and Ni were below 0.008 and 0.006 ppm, respectively. Cd and Cu levels were below 0.005 and 0.003 ppm, respectively, in all species except *L. hirsutus*. Zn was not detected in *L. undulatus*.

Although metals with a density of greater than 5 g/cm^3 are generally classified to as heavy metals, in medicine, metals with toxic properties are defined as heavy metals, regardless of their density. While Fe, Cu, Zn and Ni are essential at certain levels in living organisms, they exert toxic effects when their concentrations exceed 1-10 ppm. Some non-essential heavy metals, particularly Cd, Hg and Pb, can be toxic even at trace levels ranging from 0.001-0.1 ppm (Özbolat and Tuli, 2016). Heavy metals such as Cr, Cu, Fe, Cd, Zn, Ni, As and Pb can cause organ dysfunction even at low concentrations within the human body (Ohiagu *et al.*, 2022). The accumulation of heavy metals in plants and their subsequent entry into the food chain pose a serious threat not only to the environment but also to human health (Khan *et al.*, 2017; Ansari *et al.*, 2024).

The species whose chemical composition and some heavy metal contents were examined in the current study are utilised by humans for various purposes, including medicinal properties and nutritional values.

The seeds of *L. annuus* and the raw form of *L. tuberosus* are used as food. The tubers of *L. tuberosus* are consumed raw, roasted, or boiled for both nutritional and medicinal purposes, particularly in the treatment of diarrhoea. *L. hierosolymitanus* is used medicinally for the treatment of obesity and overweight. *L. undulatus* serves

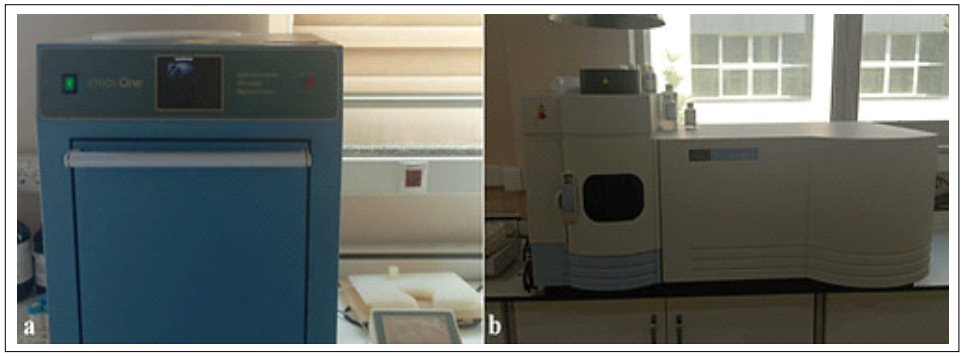


Fig 2: Analytical instruments (a. Milestone Ethos One digestion system, b. Perkin Elmer Optima 5300DV).

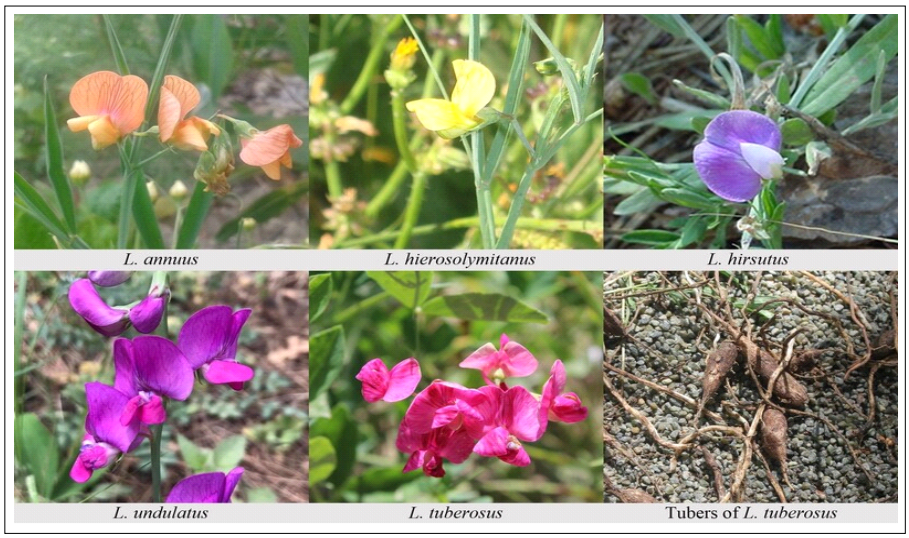


Fig 3: Investigated *Lathyrus* species.

medicinal purposes, specifically in reducing stomach cancer risk and is also used as fodder. Finally, *L. hirsutus* is utilised for fodder and forage (Hossaert-Palauqui and Delbos, 1983; Baytop, 1984; Yıldırım *et al.*, 2001; Özkan, 2011; Kızılarslan and Özhatay, 2012; Jaradat *et al.*, 2017; Smýkal,and Erdõs, 2020; Sakinoğlu-Oruç *et al.*, 2021; Ramya *et al.*, 2022).

The chemical compositions of several *Lathyrus* species have been characterised in previous studies. The seeds of *L. tefennicus* H.Genç & A.Şahin and *L. egirdiricus* H.Genç & A.Şahin, as well as the aerial parts of *L. phaselitanus* Hub.-Mor. & Davis, *L. lycicus* Boiss. and *L. belinensis* N.Maxted & D.J.Goyder, were examined in these studies. A summary of the literature data is presented in Table 3.

Soil and plants near highways are exposed to Cd and Mn contamination resulting from motor vehicle exhaust and tire wear (Lytle *et al.*, 1995; Öktüren-Asri *et al.*, 2007). Mn toxicity affects the nervous system (Mahurpawar, 2015), whereas Itai-itai disease, a form of Cd poisoning, manifests with various symptoms including osteomalacia, renal anaemia and tubular nephropathy (Ohiagu *et al.*, 2022). When the investigated species are compared with each other and with data from the literature, certain element concentrations stand out. Considering heavy metals known to cause toxicity, *L. hirsutus* exhibited relatively high levels of Mn and Cd, one of the most toxic heavy metals, according to literature data. Cd has not been detected in any of the species reported in the literature. The collection locality of the *L. hirsutus* is located near a road. It should be noted that the high Cd and Mn concentrations in this species may have been affected by environmental factors. Cd contents of the other species were lower than 0.005 ppm. Mn contents of the aerial parts and tb. of *L. tuberosus* were lower than 0.002 ppm. Mn levels of *L. hierosolymitanus* and *L. annuus* were 17 and 21 mg/kg, respectively and compatible with the literature.

Nausea, vomiting and diarrhoea are symptoms of Cu poisoning and increased exposure leads to pathological changes in multiple tissues, particularly the liver and kidneys. Increased absorption from the intestine leads to the development of Wilson's disease (Özbolat and Tuli, 2016). The Cu content in *L. hirsutus* is several times higher than the values reported in the literature, whereas it is below 0.003 ppm in the other species.

Zn (II) oxide adversely affects the gastrointestinal system, causing symptoms such as nausea, vomiting and

Table 2: Chemical compositions of the investigated *Lathyrus* species.

Metal (mg/kg)	*L. annuus*	*L. hierosolymitanus*	*L. hirsutus*	*L. undulatus*	*L. tuberosus*	*L. tuberosus* (tb.)
Na	1314±0.024	3080±0.031	<0.010*	38±0.001	37±0.001	163±0.003
Mg	2127±0.030	2076±0.112	<0.004*	902±0.016	1051±0.018	533±0.008
K	11970±0.058	6814±0.064	90±0.001	14250±0.143	16610±0.114	2504±0.046
Ca	11600±0.174	8530±0.465	18700±0.276	4949±0.089	4252±0.099	1891±0.026
Fe	47±0.001	229±0.014	<0.007*	15±0.001	39±0.002	80±0.004
Zn	2±0.001	20±0.002	52±0.001	**	<0.005*	21±0.001
Mn	21±0.001	17±0.003	6238±0.088	9±0.001	<0.002*	<0.002*
Cr	<0.008*	<0.008*	<0.008*	<0.008*	<0.008*	<0.008*
Ni	<0.006*	<0.006*	<0.006*	<0.006*	<0.006*	<0.006*
Cd	<0.005*	<0.005*	1470±0.016	<0.005*	<0.005*	<0.005*
Cu	<0.003*	<0.003*	19±0.001	<0.003*	<0.003*	<0.003*

*: ppm (parts per million), **: not detected.

Table 3: Chemical compositions of some *Lathyrus* species reported in the literature.

Metal (mg/kg)	*L. tefennicus*	*L. egirdiricus*	*L. phaselitanus*	*L. lycicus*	*L. belinensis*
	(Arslan *et al.*, 2018)			(Yildirim *et al.*, 2020)	
Na	6650	4967	520	1575	1792
Mg	1347	1026	1086	2242	3348
K	5306	5919	2877	4824	6847
Ca	760	1343	*	*	*
Fe	66	51	159	189	672
Zn	25	42	97	85	110
Mn	9.7	11	25	90	83
Cr	**	**	**	**	**
Ni	**	**	**	**	**
Cd	**	**	**	**	**
Cu	4.7	3.7	8	5	5

*: Not studied, **: Not detected.

abdominal pain. Excessive Zn accumulation within cells triggers cell death (Ohiagu *et al*., 2022). Zn concentrations in *L. hirsutus*, *L. hierosolymitanus* and the tb. of *L. tuberosus* were relatively consistent with values reported in the literature. Zn levels in *L. annuus* and *L. tuberosus* were considerably lower than those reported in the literature. Zn was not detected in *L. undulatus*.

High accumulation of Ni and its compounds may cause various types of cancer, lung fibrosis, as well as kidney and cardiovascular infections, whereas exposure to elevated levels of Cr is toxic and carcinogenic to humans (Mahurpawar, 2015; Ohiagu *et al*., 2022). Ni and Cr concentrations were found to be below 0.008 ppm in all examined species, whereas literature reports indicate that these metals were not determined in *L. tefennicus*, *L. egirdiricus*, *L. phaselitanus*, *L. lycicus* and *L. belinensis*.

The highest Fe concentrations were found in *L. hierosolymitanus* (229 mg/kg) and the tb. of *L. tuberosus* (80 mg/kg). The Fe contents in the other species were determined to be lower than those reported in the literature. The K contents of *L. tuberosus*, *L. undulatus*, *L. annuus* and *L. hierosolymitanus*, as well as the Ca contents of all species, were found to be higher than those reported in the literature. *L. hirsutus* contains very low levels of Na, Mg and Fe.

Element concentrations in plants are known to depend on the amount of the element in the soil, the plant's uptake efficiency, species-specific characteristics and the plant's ability to transport elements through its vascular tissues (Viers *et al*., 2013). Different element concentrations were observed even in different parts of the same plant, such as the aerial parts and tb. of *L. tuberosus*. These differences were more pronounced in the concentrations of Na, K and Zn. The significantly higher levels of Mn, Cd and Cu in *L. hirsutus* compared to the other species may be attributed to the environmental conditions of its habitat. Since the collection locality of this species is in close proximity to a road, exhaust emissions may have affected the environment.

The samples of the species examined in this study were collected from different localities and habitats. The soil composition and climatic conditions of these regions may also have affected their elemental profiles. Considering that the element concentrations may also vary according to the plant species, it can be thought that it is normal for the results obtained in the species studied in the current study to show differences from each other and the literature. However, further research is required to clarify whether the elevated heavy metal contents of the species are attributable to environmental conditions or inherent species-specific characteristics.

CONCLUSION

The findings of this study reveal significant variation in elemental composition among the examined *Lathyrus* species, with *L. hirsutus* notably accumulating higher levels of certain heavy metals, particularly Mn and Cd. Different parts of the same species, such as aerial parts and tubers, may exhibit variations in both content and biological effects. Chemical composition differences in plants may be attributed to species-specific characteristics or environmental factors.

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Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the views of their affiliated institutions. The authors are responsible for the accuracy and completeness of the information provided, but do not accept any liability for any direct or indirect losses resulting from the use of this content.

Informed consent

All animal procedures for experiments were approved by the Committee of Experimental Animal care and handling techniques were approved by the University of Animal Care Committee.

Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

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