

Research article

Micro mineral composition of *Ranunculus sphaerospermus* and its evaluation as an alternative forage source

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Abstract: *Ranunculus sphaerospermus* Boiss. & Blanche, a species naturally occurring in the wetlands of Bahçesaray, Van province, has been traditionally used as roughage for winter cattle feeding at farms in the region. This study aims to assess its suitability as an alternative forage resource by analyzing its micromineral composition. The elements evaluated included Sr, Al, Cd, Cr, Ba, Hg, Pb, Cu, Rb, Li, Be, V, Mn, Fe, Ni, As, Co, Ag, Zn, Ga, Se, Sb, Cs, Tl, and U. Among these, the concentrations of Mn, Se, and Hg were above the recommended levels for ruminant rations but remained within permissible safety limits. Compared to commonly used forage plants, *Ranunculus sphaerospermus* demonstrates potential as a viable alternative roughage source for ruminant nutrition due to its overall favorable mineral profile.

Keywords: *Ranunculus sphaerospermus*, nutrients, mineral substances

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Introduction

Ranunculus sphaerospermus Boiss. & Blanche (Buttercup) plant, used as animal feed in winter months in some villages of Van, grows in various wetlands such as streams, lakes, marshes and ponds. (Lumbreras et al. 2011; Budağ & Fırat, 2015,). The performance of these plant communities is influenced by physico-chemical factors, including water alkalinity and mineralization levels. Species like *Ranunculus. trichophyllus*, *Ranunculus circinatus*, *Ranunculus sphaerospermus*, and *Ranunculus rionii* are commonly found in base-rich wetlands (Hejny & Husák, 1978; Holmes, 1979; Melendo et al. 2003; Lumbreras et al. 2009; Hamdan et al. 2010).

Plants of the genus *Ranunculus* typically grow in humid areas, though some species thrive in calm regions of freshwater sources. While many species

are poisonous, a few are used for feed or medicinal purposes (Tanker, 2014). This genus is represented by approximately 3,410 species worldwide (The Plant List, 2015) and includes 101 taxa within the flora of Turkey (Tanker, 2014). Additionally, some species are dried and utilized as feed, food, or alternative roughage (Çağan & Arslan, 2020).

Research on plants with alternative feed potential focuses on factors such as their adaptability to soil and climatic conditions, nutrient content, and suitability for animal feed (Mitchell, 1966; Barry et al. 1971; Doust, 1981; Clark et al. 2000; Banuelos et al. 2002). Across the globe, locally used forage crops continue to be studied (Rao & Shahid, 2011; Oktay & Temel, 2015; Kuşvuran et al. 2019).

In Turkey, the use of alternative forage crops remains limited. Although many plant species in the

Turkish flora have cultivation potential, studies on their forage value and increasing their production are insufficient. It has been emphasized that further research is needed in this area to help address the country's forage deficit (Avcioğlu et al. 2000; Uzun et al. 2008; Bingöl et al. 2010; Kara & Yüksel, 2014).

The aim of this study is to determine the forage potential of *Ranunculus sphaerospermus* Boiss. & Blanche, a species that naturally grows in Van Province and is used as animal feed during the winter months. The study specifically evaluates this species in terms of its micro-mineral content. Additionally, the research aims to contribute to the potential utilization of alternative forage plants within the flora of Türkiye.

Materials and Methods

The material for the study (Figure 1) consists of samples of *Ranunculus sphaerospermus* Boiss. & Blanche. The identification of samples collected from the Bahçesaray surrounding wetlands in the Bahçesaray district of Van province was conducted based on the “*Flora of Turkey and the East Aegean Islands*” (Davis, 1965). As shown in Figure 1, the samples gathered during field studies were added to the Mehmet Fırat 31817 collection at the Van Yüzüncü Yıl University Faculty of Education Herbarium (VHLV).

In January, analysis samples were obtained from both vegetative (roots, stems, leaves) and generative (flowers and seeds-achenes) parts of the plants collected from five different locations within the same delta and stream. These were combined to form a representative aggregate sample, from which laboratory analysis samples were taken (Bulgurlu & Ergül, 1978).

The analyses included calcium (Ca), potassium (K), sodium (Na), magnesium (Mg), strontium (Sr), aluminum (Al), cadmium (Cd), chromium (Cr), barium (Ba), mercury (Hg), lead (Pb), copper (Cu), rubidium (Rb), lithium (Li), beryllium (Be), vanadium (V), manganese (Mn), iron (Fe), nickel (Ni), arsenic (As), cobalt (Co), silver (Ag), zinc (Zn), gallium (Ga), selenium (Se), antimony (Sb), cesium (Cs), thallium (Tl), and uranium (U), conducted according to AOAC (1990).

The analysis utilized an Agilent 7700x ICP-MS device with the following specifications: 1300 W RF power, 15 L/min plasma gas flow, 1 L/min auxiliary gas flow, 1 L/min carrier gas flow, a micro mist nebulizer, x-lens ion lens, and an analyzer pressure range of 1×10^{-4} to 2×10^{-3} Pa.

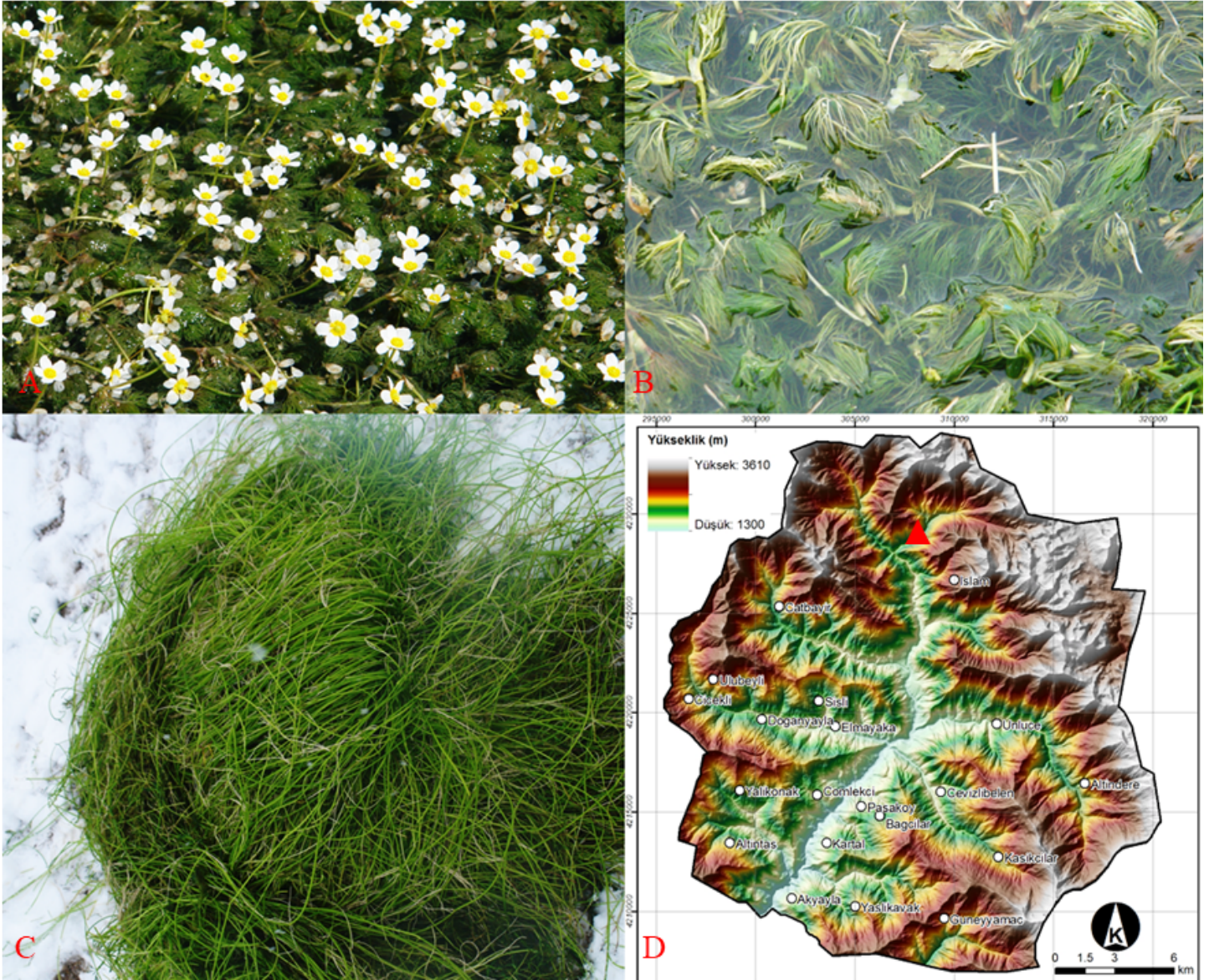


Figure 1. (A, B, C): *Ranunculus sphaerospermus* in the Bahçesaray for analysis (▲); (D): Map of Bahçesaray

Results

The strontium (Sr) content of the plant was determined to be 28.91 ppm (Table 2). A study involving Sr supplementation up to 2000 ppm in cattle reported no toxic effects (Hansard, 1964). For goats, the recommended Sr level in rations is 40.00 ppm (Li et al. 2009). It has been reported that Sr levels in plants can range from 25.40 to 274 ppm (Sasmaz et al. 2021).

In a limited number of studies on Sr levels in roughages, Goldman et al. (1965) reported an Sr content of 36.00 ppm in alfalfa, while Veresoglou et al. (1995) found 57.00–62.50 ppm in English ryegrass

and 0.63–1.74 ppm in white clover. Walsh (1944) recorded Sr levels of 20.40 ppm in oat straw, 13.10 ppm in wheat straw, and 10.90 ppm in barley straw. Schmidt (1965) reported Sr levels of 20.00–22.00 ppm in corn leaves, and Smith (1971) noted a range of 13.00–53.20 ppm in wheat yield. Beresford et al. (2000) found Sr levels of 0.15–0.31 ppm in meadow grass, and Sasmaz et al. (2021) reported 23.70–288.00 ppm in nine wetland plants, including hemp-agrimony (*Eupatorium cannabinum* 67.80 ppm) and reed (*Phragmites australis* 119.20 ppm).

Table 1. Physicochemical parameters of 4 stingless bee honey species. Values expressed as mean \pm standard deviation, and * = significant differences ($P < 0.05$).

Element	
Strontium (Sr) ppm	28.91 \pm 0.32
Aluminum (Al) ppm	236.07 \pm 3.30
Cadmium (Cd) ppm	0.21 \pm 0.01
Chromium (Cr) ppm	1.30 \pm 0.09
Barium (Ba) ppm	7.57 \pm 0.01
Mercury (Hg) ppm	0.38 \pm 0.02
Lead (Pb) ppm	0.10 \pm 0.01
Copper (Cu) ppm	2.08 \pm 0.02
Beryllium (Be) ppm	0.80 \pm 0.01
Vanadium (V) ppm	2.02 \pm 0.01
Manganese Mn) ppm	52.10 \pm 0.10
Iron (Fe) ppm	234.97 \pm 4.23
Nickel (Ni) ppm	2.89 \pm 0.29
Arsenic (As) ppm	1.54 \pm 0.13
Cobalt (Co) ppm	2.28 \pm 0.04
Silver (Ag) ppm	0.02 \pm 0.00
Cadmium (Cd) ppm	0.21 \pm 0.01
Lithium (Li) ppm	1.77 \pm 0.01
Antimony (Sb) ppm	190.13 \pm 3.23
Cesium (Cs) ppm	0.22 \pm 0.01
Thallium (Tl) ppm	0.06 \pm 0.00
Uranium (U) ppm	0.26 \pm 0.02
Selenium (Se) ppm	0.71 \pm 0.01
Gallium (Ga) ppm	1.36 \pm 0.02
Zinc (Zn) ppm	14.99 \pm 0.02
Rubidium (Rb) ppm	83.19 \pm 0.50

The Sr content of the plant is lower than the recommended level for goats and aligns more closely with the lower end of the range reported for plants in the literature. Therefore, it is considered that using the plant as an alternative roughage source would not pose any harm.

The aluminum (Al) content of the plant was found to be 236.07 ppm (Table 2). Allen (1984) and Eppe et al. (2023) report that up to 1000 ppm of Al in ruminant rations does not have any adverse effects on animals. Hmeer (2020) states that Al levels in various plants range between 2.32 and 499.70 ppm, while Allen (1979) indicates that Al levels in roughages range from 2000 to 8000 ppm.

Pekol (2018) reported an Al content of 421.00 ppm in the green parts of wheat plants, noting that this value varies depending on several factors. Büyükkeskin (2008) found a normal Al content of

782.00 ppm in broad beans (*Vicia faba*), which decreased to 477.00 ppm when inhibitory receptor agents were applied. In the same study, Al levels were reported as 94.30–104.10 ppm in soybeans and 88.00 ppm in the green parts of early-stage corn plants. Suthipradit et al. (1990) reported Al levels of 8.00–47.00 ppm for soybeans and 7.00–47.00 ppm for mung beans. McLean & Gilbert (1928) documented Al levels of 460.00 ppm for rye (*Secale cereale*), 1180.00 ppm for oats, 190.00 ppm for alfalfa (*Medicago sativa*), and 660.00 ppm for wheat (*Triticum sp.*).

The Al content of the plant is comparable to that of some feed sources, higher than some, and lower than others. However, since the Al level in the plant is below the upper limit deemed safe for ruminant rations, it suggests that the plant could serve as a suitable alternative roughage in terms of Al content.

The maximum allowable cadmium (Cd) level in feeds is set at 1.00 ppm Official Gazette (2014). Bağdatlı (2019) reported that the Cd content in alfalfa varies between 0.005 and 167.161 ppm, depending on the Cd concentration in the soil. Similarly, Guala et al. (2010) found a range of 1.00–174.70 ppm in alfalfa. Duman et al. (2007) reported Cd levels of 0.11–0.36 ppm in lakeshore bulrush (*Schoenoplectus lacustris*) and 0.15–0.42 ppm in reed (*Phragmites australis*). Ört (2019) recorded a Cd level of 0.05 ppm in vetch (*Vicia sp.*) silage.

The Cd levels reported in other plants by various researchers include: canola (*Brassica napus*) 0.30–0.92 ppm (Marchiol et al., 2004), Italian rye-grass (*Lolium multiflorum*) 0.12 ppm (Certification Report, 2010); 0.11 ppm (Madejón et al., 2003), corn (*Zea mays*) 0.40–0.80 ppm, Bermuda grass (*Cynodon dactylon*) 0.24 ppm (Madejón et al., 2002), and Johnson grass (*Sorghum halepense*) 0.03 ppm (Madejón et al., 2002).

Although the Cd content of the plant is similar to some roughages and different from others, it remains below the upper limit of 1.00 ppm established for feeds. Therefore, it is considered safe to use the plant as an alternative roughage in terms of Cd content.

The chromium (Cr) content of the plant was found to be 1.30 ppm (Table 2). According to NRC (2001), the toxic Cr level for animals is 3000 ppm. Pirhofer-Walzl et al. (2011) reported Cr levels of

0.20–0.30 ppm in forage grasses, while four different forage legumes (white clover, alfalfa, stone clover, and gazal horn) had an average Cr content of 0.20 ppm. Ugulu et al. (2019) reported a Cr content of 0.019 ppm in fenugreek (*T. foenum-graecum*). Spears et al. (2017) documented Cr levels in various forages, including alfalfa (0.20–0.89 ppm), corn silage (0.11–0.44 ppm), meadow grass (0.01–0.32 ppm), orchardgrass (*Dactylis glomerata*) (0.09–0.11 ppm), reed ball (0.11–0.25 ppm), and white clover (0.33–0.39 ppm).

Lashkari et al. (2018) reported Cr levels of 0.27 ppm in dense meadow grass, 0.80 ppm in mixed grass-legume meadow grass, and 2.20 ppm in barley yield. Beyzi et al. (2023) reported significantly higher Cr levels in reed (*Phragmites australis*), ranging from 27.19 to 29.62 ppm.

Although the Cr content of the plant is higher than that of many forage plants, it remains well below the toxic threshold for cattle. Therefore, using the plant as alternative roughage poses no risk in terms of Cr content.

The barium (Ba) content of the plant was determined to be 7.57 ppm (Table 2). While no specific data is available on the Ba requirement in ruminant rations, it has been reported that when the Ba level in the diet exceeds 30.00 ppm, cellulose digestion in the rumen decreases (Martinez & Church, 1970). The Ba content in plants varies depending on numerous factors, particularly soil characteristics and plant type (Kujawska & Pawłowska, 2019).

Chamberlain & Miller (1982) reported Ba levels in various forages ranging between 4.02 and 24.47 ppm, including 18.30–19.70 ppm in Bermuda grass, 4.40–22.50 ppm in alfalfa, 22.80–27.60 ppm in purple threeawn (*Ar. purpurea*), 4.50–5.70 ppm in Texas winter ryegrass (*Secale leucotricha*), and 22.80–27.60 ppm in supplemental Aegilops (*Aegilops purpurea*). Kabata-Pendias, (2000) noted that Ba levels in most plants range from 4.00 to 50.00 ppm.

The Ba content of the plant is close to the lower limit of the ranges reported in the literature. Based on these findings, the Ba level in the plant is not

excessively high, and its use as an alternative roughage is not expected to pose any harm.

The mercury (Hg) content of the plant was found to be 0.38 ppm (Table 2). According to the Ministry of Food, Agriculture, and Livestock, the acceptable upper limit for Hg in feedstuffs is 0.10 ppm (Official Gazette, 2013). As a heavy metal, Hg levels in plants vary depending on soil and plant characteristics (Ziarati et al., 2020). Bampidis et al. (2013) reported the toxic Hg level for cattle as 8.00 ppm. Amonoo-Neizer et al. (1996) noted Hg levels in Burgu millet (*Echinochloa stagnina*) ranging from 0.10 to 6.90 ppm.

Edwards & Pumphrey (1982) found that Hg levels in meadow grass from a region contaminated with Hg were 1.90 ppm during the cold season and 6.50 ppm during the hot season, while under normal conditions, Hg levels in meadow grass were reported to be 0.20 ppm. Chekmarev et al. (2021) reported the Hg content in various plants as follows: corn straw (0.0099–0.0109 ppm), soybean straw (0.0088–0.0093 ppm), sunflower leaves (0.0097–0.0110 ppm), sainfoin (*Onobrychis*) straw (0.0024–0.0032 ppm), white clover (*Trifolium repens*) straw (0.0032–0.0036 ppm), and steppe meadow straw (0.0126–0.0136 ppm).

The higher Hg content in the plant compared to some other feeds is likely due to soil characteristics, as there are no apparent sources of Hg pollution (e.g. mines, roadsides, or industrial facilities) in the region. Given that the plant's Hg level is approximately four times the acceptable limit, it is recommended not to include it in rations at a proportion exceeding 20–25%. While using the plant as an alternative roughage for cattle at this ratio is unlikely to cause harm, the Hg levels in other feed or feed ingredients included in the ration must be carefully considered to avoid exceeding safe limits.

The lead (Pb) content of the plant was found to be 0.10 ppm (Table 2). Limited data exists in the literature regarding Pb levels in forages. According to the Official Gazette (2014), the acceptable upper limit for Pb in feedstuffs is 10 ppm. Reis et al. (2010) reported that Pb levels of 200–400 ppm per kilogram of body weight in cattle have toxic effects.

Ağırağaç & Çelebi (2021) reported that the Pb content in caramba forage ranges between 0.008 and 0.663 ppm, while Ört (2019) noted an average Pb content of 0.07 ppm in vetch (*Vicia sativa*) silage. Bağdatlı (2019) stated that Pb levels in alfalfa (*Medicago sativa*) can range from 0.059 to 71.334 ppm, depending on soil characteristics. Similarly, Guala et al. (2010) reported Pb levels in alfalfa between 0.057 and 41.950 ppm.

Duman et al. (2007) reported Pb levels of 4.94–50.47 ppm in great bulrush (*Schoenoplectus lacustris*) and 7.46–11.31 ppm in common reed (*Phragmites australis*), both of which have potential as alternative roughages. Recorded Pb levels of 4.18 ppm in wheat (*Triticum* sp.) yield.

Although the Pb content of the plant is higher than that of some feeds and lower than others, it is well below the acceptable upper limit for feedstuffs. Therefore, the plant can be considered a safe and viable alternative roughage for animal feeding.

The copper (Cu) content of the plant was determined to be 2.08 ppm (Table 2). According to NRC (2001), the Cu content of various forages includes 3.02 ppm in corn, 4.39 ppm in alfalfa, 7.00 ppm in barley, 50.76 ppm in wheat, and 10.70 ppm in oats. The recommended Cu level in cattle rations, as specified by NRC (2001), is 30.00 ppm.

Saharan et al. (2022) reported a Cu content of 1.24 ppm in rice (*Oryza sativa*) straw, while many researchers have reported Cu levels in cultivated and alternative roughages ranging from 3.00 to 48.80 ppm, with the lowest in Italian ryegrass (*Lolium multiflorum*) and the highest in quinoa (*Chenopodium kinoa*) (Ogebea & McDowell, 1998; Orden et al. 1999; Bailey, 2001; Pirhofer-Walzl et al. 2011; Mirzaei, 2012; Ozyazici & Acikbas, 2019; Tan, 2020; Uslu et al. 2021; Saharan et al. 2022). Similarly, Bhandari et al. (2016) reported Cu levels in 21 roughages ranging from 21.5 to 34.45 ppm, with the lowest in Maize bhardo (ground maize) and the highest in Chicory green (*Taraxacum officinale*).

The Cu content of the plant is lower than the levels specified by NRC (2001) for feeds and cattle rations. Based on the literature, the plant has potential as an alternative feed source with low Cu content.

The rubidium (Rb) content of the plant was determined to be 83.19 ppm (Table 2). While there is limited information in the literature regarding the Rb requirements for ruminants, the Rb requirement for goats is reported as 0.28 ppm (2007). In a study by Cabrita et al. (2016), no adverse effects on productivity or health were observed despite an Rb level of 41.00 ppm detected in goat liver. Underwood (1977) reported the toxic Rb level for animals as 8,000–12,000 ppm. The same source noted Rb levels of 160–225 ppm in soybean and 130 ppm in compact brome and sorghum silage.

Helder (1958) documented the Rb content in barley (*Hordeum vulgare*) crops as 0.82–1.59 ppm. Shtangeeva et al. (2021) reported Rb levels in sorghum (*Sorghum bicolor*) and dandelion (*Taraxacum officinale*) leaves as 7.60–9.60 ppm and 11.00–20.00 ppm, respectively. However, when additional Rb was added to plant nutrition, these levels increased to 429–1169 ppm. Tyler (1977) and Nyholm & Tyler (2000) reported Rb levels in sedge (*Carex pilulifera*) growing in humid meadows as 0.03–1.56 ppm. In another study, the same plant had Rb levels of 0.08–0.76 ppm, which increased to 2.09–2.42 ppm in samples taken from a different region with varying soil structures Drobner & Tyler (2000).

As shown in the literature, the Rb content of the plant is higher than that of plants grown in normal soils. Although the Rb level of the plant is higher than that of many other plants, it remains well below the toxic threshold. Therefore, it is considered that the plant would not pose any harm if used as an alternative roughage.

The lithium (Li) content of the plant was determined to be 1.77 ppm (Table 2). In a study with goats, rations containing 3.30 ppm and 24.00 ppm Li resulted in daily live weight gains of 77 g and 107 g, respectively (Underwood, 1977). According to Schrauzer, (2002), the Li requirement for ruminants is 5.00 ppm. Kastori et al. (2022) provided detailed information on Li levels in various plants, reporting that the Li content in the leaves of plants from the *Solanaceae* family can reach up to 1,000.00 ppm. The same researchers noted that under normal conditions, the highest Li levels are found in plants from the *Rosaceae* family (2.90 ppm), the

Ranunculaceae family (2.00 ppm), and the *Solanaceae* family (1.90 ppm). Conversely, the lowest Li levels were reported in plants from the *Urticaceae* family (0.24 ppm), the *Poaceae* family (0.24 ppm), and the *Polygonaceae* family (0.10 ppm). Franzaring et al. (2016) reported that among thirteen forage plants, the highest Li content was found in buttercup (*Ranunculus sardous*) at 2.16 ppm, while the lowest was in centaury (*Hypericum perforatum*) at 0.05 ppm. Borovik-Romanova (1965) determined Li levels in plants from 138 different families growing in eight different soil types to range between 0.15 and 0.30 ppm.

The Li content of the plant is comparable to that of plants with relatively high Li levels. However, based on the literature, this amount is not expected to negatively impact ruminants. Therefore, the plant can be considered safe for use as alternative roughage in terms of its Li content.

The beryllium (Be) content of the plant was determined to be 0.80 ppm (Table 2). Be is a potentially hazardous heavy metal pollutant that can significantly impact agricultural ecosystems by affecting productivity and sustainability. It is widely used in various industries, including the production of nuclear weapons and reactors, aircraft and spacecraft structures, tools, and X-ray machines. Its release into the environment raises concerns about ecosystem productivity and sustainability.

Although no data is available on the Be content of forages in the literature, Be levels have been reported for other plants. In cannabis plant (*Cannabis* sp.), Be levels ranged from 0.001 to 0.002 ppm. In cowpea (*Vigna unguiculata*), a legume, Be levels were found to be 0.50–5.00 ppm under normal conditions and 4.00–70.00 ppm in the dry matter of plants subjected to Be-containing fertilization (Barber et., 2022). Additionally, Be levels in cabbage (*B. oleracea*) plants ranged from 0.03 to 51.00 ppm depending on soil composition, while levels were 0.00–5.00 ppm in beans (*Phaseolus vulgaris*) and as high as 150.00 ppm in soybeans (*Glycine max*). Budağ & Fırat (2015) reported a Be content of 0.83 ppm in *Ranunculus trichophyllus*.

Due to the lack of specific data or established limits for Be content in roughage and rations, it is not possible to comprehensively evaluate the plant in terms of its Be levels. Further studies are needed to determine the acceptable levels of Be in forages and its potential impact on animal health and productivity.

The vanadium (V) content of the plant was found to be 2.02 ppm (Table 2). Studies on vanadium indicate that dietary levels of 6–10 ppm do not cause problems in ruminants (Tripathi et al., 2019). Gummow et al. (2006) reported that the normal V content in plants is around 1.00 ppm, while in some ruminant diets, V levels can range from 0.57 to 2.00 ppm. Although no specific standard for ruminants is established in the literature, the maximum tolerance level for V in ruminants is reported to be 50.00 ppm (Guapta et al., 2020). However, V levels of 7.00 ppm in the diet have been shown to impair rumen development in calves Gummow et al. (2006)

Additionally, Gummow et al. (2006) noted that pastures highly contaminated with V contained levels ranging from 64.00 to 127.00 ppm. Antal et al. (2009) measured V levels in thirty different medicinal and aromatic plants, with amounts ranging from 31.00 ppm in field horsetail (*Equisetum arvense*) to 76–300.00 ppm in wild thyme (*Thymus pulegioides*). Velch & Cary (1975) found V levels in wheat 0.046 ppm, barley 0.028 ppm, oats (*Avena sativa*) 0.055 ppm, and peas (*Pisum sativum*) 0.054 ppm. Yao et al. (2022) reported V levels in the stems and leaves of two marsh plants (*Phragmites australis* and *Cyperus malaccensis*) ranging from 0.01 to 15.23 ppm. Budağ & Fırat (2015) reported a V content of 2.17 ppm in *Ranunculus trichophyllus*.

While the V content of the plant is higher than the levels reported by Velch & Cary (1975) for some roughage plants, it remains well within the tolerance limits for ruminants. Therefore, the plant can be considered suitable alternative roughage for ruminants in terms of its V content.

The manganese (Mn) content of the plant was found to be 52.10 ppm (Table 2). According to NRC (2001), Mn levels in various feedstuffs are as follows: oats 51.00 ppm, corn 17.10 ppm, alfalfa 22.70 ppm,

and barley 38.00 ppm). The recommended Mn levels for cattle rations are between 17.80 and 24.20 ppm. Hidiroglou (1979) reported that Mn levels in ruminant rations can range from 5 ppm to 1000 ppm. For optimal performance in ruminants, the Mn content should be around 40.00 ppm, while levels of 100 ppm or higher can decrease cellulose digestion in the rumen.

The Mn content of the plant is higher than the Mn levels found in feeds reported by NRC (2001) and exceeds the required levels for cattle rations. However, it is lower than the Mn content of many forage plants, which ranges from 56.70 ppm in triticale to 278.00 ppm in clover (Ogebea & McDowell, 1998; Orden et al., 1999; Bailey, 2001; Pirhofer-Walzl et al., 2011; Mirzaei, 2012; Ozyazici & Acikbas, 2019; Tan, 2020; Uslu et al., 2021; Saharan et al., 2022). Similarly, Weiss & Socha (2005) reported Mn levels of 78.00 ppm in alfalfa and 80.00 ppm in meadow grass.

The plant's Mn content is lower than most roughages but higher than Mn levels reported in Bailey (2001) for alfalfa 30.30 ppm) and corn straw 23.50 ppm), Saharan et al. (2022) for pearl millet (*Pennisetum glaucum* 28.10 ppm), wheat straw 29.69 ppm), and rice straw 33.87 ppm). Although the Mn content of the plant is lower than many forage plants, its level exceeds the amount required for cattle rations, suggesting that the plant can serve as a viable alternative roughage.

The iron (Fe) content of the plant was found to be 234.97 ppm (Table 2). According to NRC (2001), the recommended Fe level in cattle rations is 50 ppm. NRC (2001) provides the Fe contents of several plants as follows: corn 98.20 ppm), alfalfa 159.00 ppm), barley 86.00 ppm), wheat 71.60 ppm), and oats 107.00 ppm). Pirhofer-Walzl et al. (2011) reported Fe levels in gramineous forage crops ranging from 50.90 to 82.30 ppm and in legume forage crops from 73.80 to 77.30 ppm.

Bailey (2001) reported Fe levels of 132.00 ppm in meadow grass (*Festuca pratensis*), 198.00 ppm in alfalfa, and 131.00 ppm in cornmeal. Mirzaei (2012) reported Fe contents of 117.50 ppm in sorghum, 170.00 ppm in pearl millet, and 103.00 ppm in millet. Uslu et al. (2021) recorded an average Fe content of

92.35 ppm across nine varieties of triticale (*Triticale*). Tan (2020) and Saharan et al. (2022) both reported Fe levels of 112.60 ppm in Shojne (*Moringa oleifera*), 133.45 ppm in millet (*P. miliaceum*), 147.55 ppm in wheat straw, and 185.79 ppm in rice straw. Özkan et al. (2020) reported Fe levels in eleven out of thirteen roughages from legume and other families ranging between 285.00 ppm (*Trifolium repens*) and 1187.50 ppm narrow-leaved plantain (*Plantago lanceolata*). Similarly, Mirzaei (2012) recorded Fe levels in gramineous forages ranging from 265.00 ppm (oat, *Avena* sp.) to 912.00 ppm (couch grass, *Elymus repens*), while Ogebea & McDowell (1998) reported Fe levels of 650.00 ppm in grass forage plants and 895.00 ppm in legume forage plants.

Although the Fe content of the plant is lower than that of many other forage plants, it exceeds the amount recommended for cattle rations. Therefore, the plant can be considered a viable alternative roughage if the ration is balanced appropriately in terms of Fe.

The Ni amount of the plant was determined as 2.89 ppm (Table 2). It is reported that adding up to 5.00 ppm of Ni to the ration in cattle gives positive results (Spears et al. 1979; Spears et al. 1986), and causes various problems in ruminants at 10.00 ppm (Rizvi, 2020). The average Ni amount detected in clover (*Medicago sativa*) in the Diyarbakır region is given as 12.81 ppm (Demir & Düz, 2008). In the literature where the Ni contents of various roughages are given, the range of change is quite high. As a matter of fact, while the Ni amount in alfalfa is given in the range of 0.059-71.334 ppm (Bağdatlı, 2019). Duman et al. (2007) give the Ni amounts in lakeshore bulrush (*Schoenoplectus lacustris*) and reed (*Phragmites australis*) as 11.18-15.12 ppm and 8.60-16.55 ppm, respectively. While the amount of Ni in wheat is given as 4.21 ppm (Karataş et al., 2007), Ört (2019) gives it as 0.36 ppm in vetch silage, 0.55-1.17 ppm in canola (Marchiol et al., 2004), 9.95 ppm in *Cynodon dactylon*, 0.96 ppm in *Sorghum halepense* Madejón et al., (2002), 15.20 ppm in Italian

ryegrass (Certification Report, 2010). Although the amount of Ni in the plant is lower than the amount determined for cattle, it is higher than the amount of Ni in some feeds and lower than some. Therefore, it is thought that the plant can be alternative roughage that will not cause problems in the ration in terms of Ni.

The amount of As in the plant was determined as 1.54 ppm (Table 2). The Ministry of Food, Agriculture and Livestock gives the acceptable upper limit of arsenic (As) in feedstuffs as 2 ppm (Official Gazette, 2014). The amount of As in plants varies according to the amount of As in the soil and the As accumulation characteristics of the plant (Ziarati et al., 2020). Ört (2019) gives the amount of arsenic in vetch silage as 0.04 ppm. In the literature, the amount of As in sunflower (*Helianthus annuus*) is 9.60-11.20 ppm, in Italian ryegrass (*Lolium multiflorum*) 0.104-0.132 ppm (Madejón et al., 2003), 0.68 ppm in dog's tooth wheat, 0.20 ppm in bloodroot (Madejón et al., 2002), 0.042 ppm in Italian ryegrass (Certification Report, 2010), 0.60 ppm in alfalfa, 0.20 ppm in gazal horn, 0.20 ppm in awnless brome, 0.40 ppm in corn grain, 0.40 ppm in millet 0.20 ppm in wheat yield, 0.20 ppm in sorghum yield, 0.10 ppm in sorghum yield (Furr et al., 1978), 0.32 ppm in corn, 0.71 ppm in sorghum (Zolnowski, 2010), 0.115 ppm in sweet yellow clover (*Melilotus officinalis*), 0.145 ppm in sqeet white clover (*Melilotus alba*), 0.175 ppm in wheat yield, 0.125 ppm in clustered wheat grass (*Agropyron desertorum*), 0.100 ppm in common reed (*Phragmites australis*) (Belozubova & Zubkova, 2021), and 0.60-0.90 ppm in rice yield (Rahman et al., 2028). Jahan et al. (2014) They reported that the amount of As in meadow grass in As-contaminated soils varied between 0.13 and 1.55 ppm, and in common water hyacinth (*Eichhornia crassipes*) between 0.162 and 0.555

ppm. Amonoo-Neizer et al. (1996) reported the amount of As in Niger grass as 1.10-27.60 ppm. The amount of As determined in the plant is higher than many forage plants. However, the amount of As in the plant is close to the amount of As given by Budağ & Fırat (2015) for *Ranunculus trichophyllus* (1.33 ppm). Although the amount of As in the plant is high, since it is below the upper limit required for rations, the plant can be accepted as an alternative roughage that can be easily used in cattle.

The amount of Co in plant was determined as 2.28 ppm (Table 2). The amount of Co that should be present in the ration is given as 0.17-0.29 ppm (Stemme et al., 2004). According to NRC (2001) data, the toxic level for Co in the ration was determined as 10 ppm. While FEEDAP (2009) gave the amount of Co in alfalfa as 1.73 ppm, Castagnino et al. (2017) gave the amount of Co in alfalfa as 1.27-1.48 ppm. The same researchers reported the Co content in orchardgrass (*Dactylis glomerata*) as 1.31-1.64 ppm, Zain et al. (2023) reported the Co content in five different alternative forage plants (*Calliandra calothyrsus*, *Arachis pintoii*, *Sesbania grandiflora*, *Calopogonim mucunoides* and *Arachis hypogea*) as 0.13-0.51 ppm, and four different grass forages (meadow grass, corn straw, corn cob, rice straw) as 0.33-0.52 ppm. Ogebea & McDowell (1998) reported the Co content for grasses as 0.01 ppm. Bhanderi et al. (2013) gave the Co amounts of seventeen different roughages between 0.23 ppm in fennel (*Foeniculum vulgare* subsp. *vulgare* var. *dulce*) (as an alternative feed, at least) and 0.66 ppm in chicory plants (*Cichorium intybus* as an alternative feed plant). When looking at the literature data, the Co amount of the plant is higher than the Co amount of many roughages. Since the toxic Co level for cattle is given as 40.00 ppm Wrzecińska et al. (2021), the plant can be

considered as alternative roughage that can be easily used in cattle rations.

The Ag amount of the plant was found to be 0.02 ppm (Table 2.). There is no sufficient literature on the amount of Ag in roughages. Stegemeier et al. (2015) reported that approximately 99% of the total Ag accumulated in alfalfa plants in fertilization with Ag-containing fertilizers accumulated in the roots, while 1% accumulated in the stems and leaves of the plant. Accordingly, it is reported that the amount of Ag in the plant, which is less than 1.00 ppm under normal conditions, increased to 1489-2884 ppm in the roots and approximately 14.89-28.84 ppm in the aboveground organs, depending on the fertilizer dose. In a similar study, Gardea-Torresdey et al. (2003) reported the amount of Ag accumulated in alfalfa roots as 320.00 ppm and 3.20 ppm in the aboveground parts. The average amount of Ag in terrestrial plants is given as 0.06 ppm (Horovitz, 1974). Arık & Yıldız (2010) reported that the amount of Ag determined in meadow grass varies between 0.01-0.09 ppm depending on the distance to the mineral deposits. Jones & Bailey (1974) reported that Ag compounds are relatively harmless to higher organisms. According to the researchers, free Ag ions that inhibit enzymes in the organism are rapidly inactivated and detoxified in the intestines of mammals. In a study conducted by the researchers on rabbits, which are herbivorous animals, 4.00 and 5.00 ppm Ag added to the ration was inactivated and neutralized by microorganisms in the cecum of the rabbits. Based on the assumption that the amount of Ag found in the plant (0.02 ppm) will not have a negative effect on ruminant rations, it was thought that there would be no harm in using the plant as alternative roughage.

The Cd amount of the plant was found to be 0.21 ppm (Table 2). The acceptable upper limit of Cd in plant feeds is given as 1.00 ppm (Official

Gazette, 2014). Bağdatlı (2019) reported that the Cd amount in alfalfa varies between 0.005-167.161 ppm depending on the Cd amount in the soil structure. Guala et al. (2010) reported this change range in the Cd amount of alfalfa as 1.00-174.70 ppm. Duman et al. (2007) reported the Cd amounts of lakeshore bulrush (*Schoenoplectus lacustris*) and common reed (*Phragmites australis*) as 0.11-0.36 ppm and 0.15-0.42 ppm, respectively. Ört (2019) gives the Cd amount in vetch silage as 0.05 ppm. The amounts reported by different researchers for the amount of Cd found in different plants are as follows: Canola 0.30-0.92 ppm (Marchiol, 2004), Italian ryegrass 0.12 (Certification Report, 2010), Italian ryegrass 0.11 ppm (Madejón et al., 2003), corn 0.40-0.80 ppm (Keller et al., 2023), Bermuda grass 0.24 ppm and Johnsongrass (*Sorghum halepense*) 0.03 ppm (Madejón, 2002). Although the Cd amount of the plant is similar to some roughages and different from others, it is seen that there is no harm in using the plant as an alternative roughage since it is below the upper limit that should be found in feeds.

The Zn amount of the plant was determined as 14.99 ppm (Table 2). NRC (2001) stated that corn 12.20 ppm, alfalfa 13.40 ppm, barley 38.00 ppm, wheat 39.30 ppm and oat 45.00 ppm. NRC (2001) gives the amount of Zn that should be included in the ration for cattle as 22.80-33.00 ppm. The amount of Zn in the plant is lower than the amounts determined by NRC (2001) for cattle rations. Ogebea & McDowell (1998) reported the average Zn amount of Gramineous forage plants they examined in their study as 8.10 ppm, Paterson & Engle (2005) reported the amount of Zn in legume forage plants as 2.24 ppm, Mirzaei (2012) reported the amount of Italian ryegrass as 11.00 ppm and sorghum straw as 12.00 ppm, and Saharan et al. (2022) reported the amount of rice straw as 11.14 ppm. The plant's Zn content is higher than these

amounts. Orden et al. (1999) gave the average Zn content as 31.51 ppm in four different gramineous forages and 31.53 ppm in four different legume forages, while Paterson & Engle (2005) gave the average Zn content as 18.20 ppm in gramineous forages; Bailey (2001) gave the average Zn content as 35.00 ppm in tall fescue (*Lolium arundinaceum*), 18.80 ppm in alfalfa, 17.70 ppm in corn meal, Ogebea & McDowell (1998) gave the average Zn content as 30.10 ppm in legume forages, Pirhofer-Walzl et al. (2011) gave the average Zn content as 17.70-28.70 ppm in gramineous forages and 26.00-26.30 ppm in legume forages. In addition, Mirzaei (2012) gives the highest and lowest Zn amounts of eleven different forages as 24.00-48.00 ppm (lowest millet yield, highest barley yield). Tan (2020) reports that quinoa, among alternative forages, contains 64.30 ppm, palm residues contain 68.50 ppm, and moringa (*Moringa oleifera*) contains 46.70 ppm Zn. Uslu et al. (2021) reported an average of 34.10 ppm for nine different triticale, 21.25 ppm for pearl millet and 19.23 ppm for wheat straw, and Ozyazici & Acikbas (2019) reported the Zn amounts of thirteen different forages from other families excluding gramineous forages in the range of 27.00-84.00 (the lowest is white clover, the highest is knotgrass species (*Polygonum* sp.)). Khan et al. (2008) reported that the Zn amounts of meadow grass mixtures with many forage types varied between 15.20-32.77 ppm. The Zn amount of the plant is lower than many forage plants and is also lower than the amount that should be in the daily rations of cattle. For this reason, the plant will not cause any harm in terms of Zn, which is one of the heavy metals, when used in the ration, but it can be considered as alternative forage if the ration is balanced in terms of Zn so as not to cause deficiency.

The Ga amount of the plant was determined as 1.36 ppm (Table 2). Data on the Ga amounts of roughages are limited. It is reported that 2.77 ppm level of Ga added to dairy cattle feeds does not have a negative effect on performance (milk amount and components) in dairy cattle (Oancea et al., 2023). Shtangeeva (2023) gives the Ga amount in *Agropyron repens* as 0.21-6.75 ppm. The same author states that the Ga amount can be up to 11.60 ppm in English ryegrass (*Lolium perenne*). Kopittke et al. (2009) gives the Ga amount in cowpea (*Vigna unguiculata*) as 1.70-50.00 ppm. Syu et al. (2017), Gough et al. (1979) reports that the Ga amount in plant species in the USA varies between 3.00-30.00 ppm. Budağ & Fırat (2015) gave a similar amount of Ga (1.36 ppm) for *Ranunculus trichophyllus*. Since the amount of Ga in the plant's structure is between the lower and upper limits of the Ga amounts of plants given in the literature and below the limit value given for cattle rations, it was thought that there would be no harm in using it as alternative roughage.

The Se amount of the plant was found to be 0.71 ppm (Table 2). NRC (2005) and Saha et al. (2016) give the Se amount range that the ration should contain for cattle as 0.10-0.50 ppm. Ammerman & Miller (1975) give the toxic amount of Se for cattle as 5.00 ppm. In a literature study on Se amounts in forages, Winter & Gupta (1979) reported that Se amounts of 228 different forage plants were in the range of 0.02-0.19 ppm. Ogebea & McDowell (1998) reported that wheat contained 0.21 ppm, legumes 0.32 ppm, Bailey (2001) reported that alfalfa contained 0.41 ppm, and corn yield contained 0.53 ppm Se. Reported that the Se amounts in a natural habitat with twenty-four different forage plants were in the range of 0.07-1.55 ppm (lowest *Iris lactea*, highest *Alyssum desertorum*) during the growth period. As can be

seen in the literature, although the Se amount of the plant is higher than many plants, it is not as high as that of accumulator plants (Topuz & Topal, 2021). In addition, since the Se content of the plant is not at a toxic level (5-40 ppm) for cattle (Ayşan & Baylan, 2010), it can be easily used in rations as alternative roughage.

The Sb amount of the plant was found as 190.13 ppm (Table 2). There is insufficient evidence regarding the Sb requirement in animals. NRC (1980) gives the maximum Sb limit values as 70-150 ppm in rabbits. Hammel et al. (2000) reported that the Sb amount of wheat, barley, rye, oat and corn plants varied between 200.00-600.00 ppm. While Murillo et al. (1999) gave the Sb amount of sorghum as 39.00 ppm, Feng et al. (2013) gave the Sb amount between 5.00 (*Pteris vittata-fern*) 4029.00 ppm (*Boehmeria nivea*), (*Gaudich-Nettle*) in 36 plants including meadow clover (160-2151 ppm). Qi et al. (2011) reported that in their studies, in which the average Sb amount of many plants from different families was 68.00 ppm, the lowest Sb amount was found in sorghum with 3.92 ppm and the highest Sb amount was found in red summer tamarisk (*T. ramosissima*) with 143.69 ppm. Baroni et al. (2000) reported in their study determining the Sb amounts in different organs of plants that the lowest Sb amount in sweet yarrow (*Achillea ageratum*) was 4.62 ppm in basal leaves (in the uncontaminated area) and the highest Sb amount was 1401.13 ppm also in basal leaves (in the mine waste area); the lowest Sb amount in narrow-leaved plantain (*Plantago lanceolata*) was 26.38 ppm in basal leaves (in the uncontaminated area) and the highest Sb amount was 590.47 ppm again in basal leaves (in the mine waste area) and the lowest Sb amount in bladder campion (*Silene vulgaris*) was 15.63 ppm in shoots (in the uncontaminated area) and the highest Sb amount was 1191.25 ppm again in shoots (in the mine waste area). He & Yang

(1999) reported that the amount of rice plant in areas close to mining areas increased to 1565.00 ppm. On the other hand, Hajiani et al. (2015) reported that the amount of Sb in continental climate plants was between 0.20-50.00 ppm. Since the amount of Sb in the plant was less than that of meadow clover, wheat, barley, oat and corn plants, it was thought that using the plant as alternative roughage would not pose an obstacle for animals.

The Cs amount of the plant was determined as 0.22 ppm (Table 2). Meena & Rajarajan (2003) reported that different soil types and different plant species affect the Cs amounts of plants and reported that the Cs amount of sorghum varied between 0.090-0.489 ppm and alfalfa between 0.051-0.158 ppm. Takeda et al. (2008) gave the Cs amount as 0.15 ppm in corn. Veresoglou et al. (1995) gave Cs as 1.410-3.506 ppm in English ryegrass, 1.932-5.019 ppm in white clover, Takeda et al. (2008) gave it as 0.014-0.21 ppm in ryegrass (*Lolium perenne*), Sunaga & Harada (2016) gave it as 0.20-1.40 ppm in rye, Kahn et al. (148) gave it as 0.20-0.60 ppm in alfalfa, 0.25-0.70 ppm in corn silage, 0.06-0.08 ppm in grass silage. Fowler & Christenson 1959), gave it as 1.27 ppm in alfalfa, and 0.79 ppm in meadow grass. Data on Cs in forage crops vary, and some plants are reported to contain high Cs. Considering the literature, the Cs content of the plant is not higher than many traditionally used roughage crops. Therefore, it was thought that the plant could be used as an alternative feed.

The Tl amount of the plant was found to be 0.06 ppm (Table 2). The recommended Tl amount in ruminant rations is given as 1.00 ppm Hapke et al. (1980). Hapke et al. (1980) reported that the Tl amount in corn silage varies between 0.44-0.90 ppm. Makridis & Amberger (1996) gave the Tl amount of bean plant as less than 1.00 ppm, while the Tl amount of rapeseed was 1.00 ppm. Kabata-Pendias & Mukherjee (2007)

gave the Tl amount in grasses forage crops as between 0.02-1.00 ppm. The same author reported the Tl amount of clover as 0.008-0.01 ppm, alfalfa as 0.020-0.025 ppm, and Zyka (1972) reported the Tl amount of clover as 0.008-0.010 ppm. Tremel et al. (1997) reported the Tl amount of corn stalks as 0.0032-0.0045 ppm, Vaněk et al. (2009) reported that some of the weeds contained an average of 36.00 ppm Tl, while in meadows the Tl amount in the total plant mass varied between 0.005-0.020 ppm, and in white clover this rate was 0.002-0.01 ppm, and Xiao et al. (2004) reported that the Tl amount in weeds varied between 0.0008-1.00 ppm. The Tl amount of the plant is lower than the amount recommended in ruminant rations. The plant has a Tl amount lower than some roughage and higher than some roughages. Since the plant has a Tl amount lower than the maximum value required in ruminant rations, it is thought that there will be no harm in using the plant as alternative roughage.

The U amount of the plant was determined as 0.26 ppm (Table 2). There is limited information in the literature about the U amounts of forage plants. Linsalata et al. (1989) gives the maximum U amount that should be in the ration for cattle as 1.50 ppm. In a study conducted by Gradašcevic et al. (2015), they reported that the U amount of meadow grasses in the region where there is no U pollution is between 0.10 ppm and 0.40 ppm (approximately 0.30-1.20 ppm in dry matter) based on fresh weight, while this rate can increase up to 2.60 ppm depending on the pollution of the soil in terms of U. Gulati et al. (1980) reported that the amount of U in wheat varied between 0.026-0.079 ppm. On the other hand, Takeda et al. (1946) reported the amount of U in corn as 0.0002 ppm. Samuel-Nakamura (2013) reported that the amount of U in meadows, the majority of which is composed

of grasses, varied between 0.50-7.70 ppm. The same study reported that the U content of plants consumed by ruminants was between 0.002-0.101 ppm in corn, 0.29-0.58 ppm in Indian ricegrass (*Achnatherum hymenoides*), 0.28-0.44 ppm in purple three-awn (*Aristida purpurea*), 0.06-0.25 ppm in blue grama grass (*Bouteloua gracilis*), 0.07-0.32 ppm in creeping muhly (*Muhlenbergia repens*), 0.20-1.08 ppm in western wheatgrass (*Pascopyrum smithii*), 0.11-0.14 ppm in James' galleta (*Pleuraphis jamesii*) and 0.10-0.26 ppm in thyme-leaf sandwort (*Arenaria serpyllifolia*). The plant's U content was found to be higher than some plants and lower than some plants. Since the plant's U content was lower than the maximum U content in ruminant rations, it was thought that there would be no harm in using the plant as alternative roughage.

In this study, the micromineral composition of *Ranunculus sphaerospermus* Boiss. & Blanche, a plant species naturally growing in the wetlands of Bahçesaray, Van, was analyzed to assess its potential as an alternative forage source. The findings revealed that although the levels of certain elements such as manganese (Mn), selenium (Se), and mercury (Hg) exceeded recommended values, they remained within permissible safety limits for ruminants. In terms of other mineral elements, the plant showed a favorable profile suitable for ruminant nutrition. Based on these results, *Ranunculus sphaerospermus* appears to be a safe and promising alternative forage plant, particularly for winter feeding. However, due to the relatively high concentrations of some elements, its inclusion rate in rations should be carefully balanced with other feed ingredients. Further research and wider cultivation of this species may help alleviate the forage deficit in Türkiye.

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Ethical Approval

No need to ethical approval for this study.

Conflict of interest

The authors have no conflict of interest.

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