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Metallic and Chemical Composition of Wild Plant (*Rheum Ribes*): A Natural Superfood with Hidden Elements

Chnar Z. Mahmud [1] *, and Nabil A. Fakhre [1, 2]

[1] Department of Chemistry, College of Education, Salahaddin University-Erbil, Erbil, Iraq.

[2] Tishk International University, Erbil, Iraq.

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*Email address:
chnar.mahmud@su.edu.krd
*Corresponding Author

Abstract: The natural product *Rheum ribes,* also known as Rivas in the Kurdistan area, is considered one of the most nutritious wild rhubarbs and is valued both as a dietary and medicinal resource. It is commonly referred to as *R. ribes* in several scientific databases, due to the presence of anthracene derivatives-compounds known for their laxative and anti-inflammatory properties R. *ribes is recognised* as a therapeutic herb traditionally used in medicine. This study highlights its potential as a natural superfood rich in bioactive constituents and trace elements beneficial to human health. The metal content analysis identified essential elements including Iron, Zinc, manganese, potassium and copper with concentrations of 145.755, 34.437, 3364.568, 28577.680 and 29.038 µg/mL, respectively, and limit of detection (LOD) at 0.0013, 0.0006, 0.0005, 0.0022 and 0.0048 µg/mL, respectively. Trace metals were quantified using a sophisticated analytical method, Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES) technique. Average chemical composition of *R. ribes*: total ash (9.542%), moisture content (7.655%), nitrogen content (4.26%), crude protein (26.62%), and pH (7.4).

Keywords: *Rheum Ribes; Metal Analysis; Crude Protein; Total Ash; Moisture Content; Bioactive Constituents; Therapeutic Effects*

1. Introduction

R. ribes is a perennial herb from the Polygonaceae family, commonly referred to as wild rhubarb or Rivas in the Kurdistan region. R. ribes in Kurdish is called revas in the Badini dialect and rewas in the Sorani dialect. It is a hardy perennial herb that grows 40–150 centimetres tall. *R. ribes* is a species of wild rhubarb in the Polygonaceae family. It can also tolerate highly clayish, acidic, or basic soils. It can withstand extremely low temperatures up to -20°C [1, 2, 3]. (Figure 1) display *R. ribes* natural superfood.

Figure 1: *R. ribes* (Rivas) in the mountainous areas of South Kurdistan-Iraq.

More than 60 species of Rheum are found in the genus and are utilized in traditional treatments and food products all over the world [4]. About 103 species of the plant family Rheum are found in moderate and subtropical regions, especially in Western Asian nations, including Iran, Iraq, Lebanon, Azerbaijan, Turkey, and Pakistan [1].

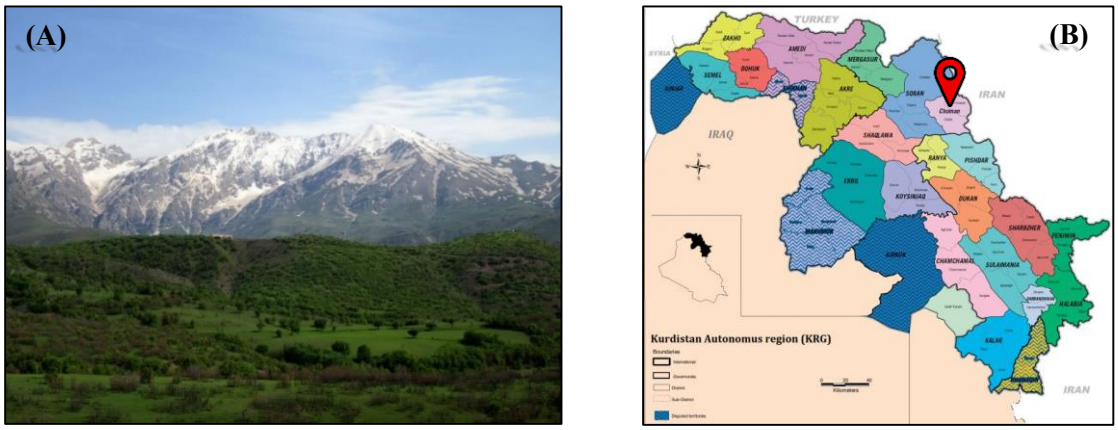

Figure 2: (A) Choman Mountain, Kurdistan region, Erbil–Iraq. (B) Map of Kurdistan Region-Iraq.

The Kurdistan Region in northern Iraq is renowned for its diverse landscapes, rich biodiversity, and traditional use of wild plants. Choman District in the northeast Erbil Governorate, bordering Iran, is characterised by mountains, one of which is the conspicuous Halgurd Mountain. Such a unique habitat enhances the presence of many native species, one of which is *R. ribes*, which is traditionally valued for its nutritional and medicinal properties. Figures 2A and 2B indicate the geographic locations of interest in this study. Figure 2A is a picture or map of Choman Mountain in the Kurdistan Region of Iraq, roughly around Erbil. Figure 2B is a broader map of the Kurdistan Region of Iraq, marking its border and salient features. Together, these figures offer spatial context to the study location and its ecological or geographical significance [5].

Throughout the course of human history, ethnobotanic applications of phytotherapeutic agents have been extensively utilized globally to manage different pathological conditions and have also contributed significantly to the establishment of international trading networks. Traditional knowledge regarding the use of medicinal plants for pharmacological ends has largely been acquired through empirical, trial-and-error practice, and most of it has been transferred across generations by word of mouth. Iraqi Kurdistan Region's unique geographical, geology, and ecological characteristics bestow upon it one of Iraq's most heterogeneous and biotic diversities abundant [2].

All physiological processes that the human body requires to sustain itself mentally as well as physically are linked to micronutrients. Humans can only obtain micronutrients from their food since they are unable to generate these molecules on their own [6]. Changes in nutrition and environment have a significant impact on human evolution [7]. Natural products have been widely utilized historically to heal a wide range of diseases and medical conditions since ancient times and in tradition [8]. Natural

products play a vital role in the drug discovery process, particularly in the context of cancer research [9]. As illustrated in Figure 3.

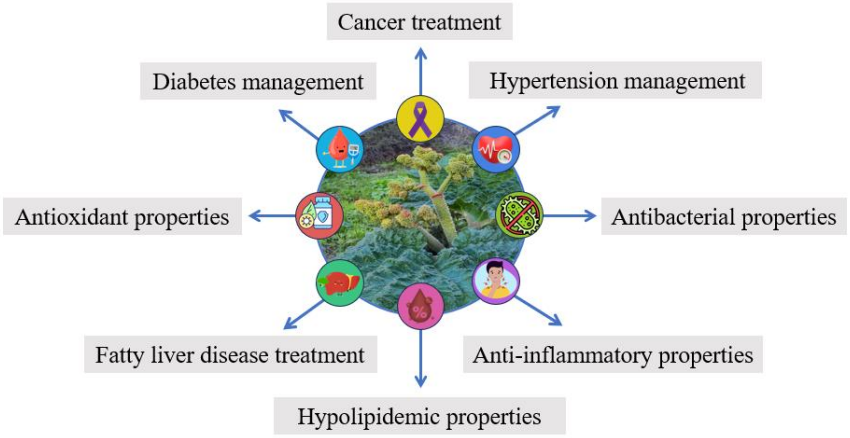

Figure 3: The pharmacological and therapeutic effects of *R. ribes*.

The active ingredients in most drugs are derived from natural sources [10]. Additionally, a variety of bioactive compounds with multiple therapeutic characteristics are found in *R. ribes.* It is consumed both raw and cooked, and it is widely employed in "folk treatment" for treating many kinds of illnesses [4]. *R. ribes* comprises bioactive molecules that have been found to enhance lipid profiles, regulate blood glucose levels, negate oxidative stress by free radical scavenging, and uphold gastrointestinal well-being, among other advantages. Members of the Polygonaceae family are rich sources of bioactive molecules underlying a wide range of pharmacological properties [19]. *R. ribes* have been shown to have a broad variety of bioactive compounds, including vitamins, flavonoids, stilbenes, anthocyanins, anthrones, and acyl glucosides [4]. The antioxidant activity is attributed to the complex polyphenolic compounds present in the phytochemical constituents, such as compounds like alkaloids, saponins, tannins, and flavonoids. Diabetes mellitus, a common global pandemic, is a disease that impacts millions of individuals. The primary treatment approach currently is to counteract postprandial hyperglycemia by inhibiting intestinal carbohydrate-hydrolysing enzymes. Different bioactive phytochemicals present in plant extracts have been reported to exhibit potential activity in the management of diabetes [20]. Bacterial infections can be alleviated, managed, and cured by a group of antimicrobial drugs known as antibiotics. These substances, whether natural, semi-synthetic, or purely synthetic, function either by killing bacteria or inhibiting their multiplication [21]. Minerals are components of all living organisms, including plants and animals, and can also be suspended in the surrounding air. Minerals serve three primary functions in the human body: First, they provide structural support for the growth and formation of bones and teeth. Secondly, they assist in maintaining proper heart rhythm, muscle contraction, nervous system function, and acid-base balance. Thirdly, they help regulate metabolic processes in cells by participating in hormones and enzymes that control cellular activity [11]. Multiple investigations indicate that wild plants are great sources of macro and micronutrients and play a significant role in supporting human nutrient needs [12]. Regarding nutrition, minerals are inorganic substances that humans and other living things need as vital nutrients to perform life-sustaining tasks. However, the lists of primary nutrition minerals commonly do not include the four fundamental structural constituents of the human body by weight: carbon, nitrogen, oxygen, and hydrogen. In combination, the four elements make up around 96% of the weight of the human body, with the remaining portion consisting of major minerals (also known as macro elements or macro minerals) and minor minerals (also known as trace elements or microelements) [13]. Fruits and vegetables are common sources of various bioactive compounds. The phrase "plant bioactive compound" generally does not refer to plant nutrients. While these compounds are not essential for the plant's daily operations, such as growth, they are often produced as secondary

metabolites. These bioactive compounds are crucial for competition, defense, attraction, and signaling [14]. A widely utilized technique in emission spectroscopy is Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES) technique was also determined to be an appropriate analytical technique for mineral determination in samples dissolved at 180°C using concentrated nitric and hydrochloric acids, as utilized in another study [15]. An argon ICP operates at 5500–6500 K, efficiently ionizing most elements from aqueous aerosols. The most crucial components include a spray chamber to prevent large droplets and a pneumatic nebulizer to create aerosols, allowing for high atomization and sensitivity [16, 17]. This study aims to identify and analyze essential metals and chemical constituents in *Rheum ribes*, a wild plant from Choman, South Kurdistan-Iraq, with the goal of understanding its nutritional and therapeutic potential, thereby contributing to public health knowledge and the development of functional foods or herbal medicines.

2 Methodology

2.1 Chemicals and reagents

Table 1 utilized reagents.	
Type	**Reagents with Brand**
Standard solution for the analysis	Multi Elements ICP 24E ROTI Star (ROTH, Germany; Lot Number: 796301)
Standard solutions for calibration and quality control	Multi Elements Standard QC 22E (NIST SRM)
For sample preparation, Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES)	Nitric acid (HNO_3) 69% BIOCHEM (France)
	Hydrogen peroxide (H_2O_2) 30%, BIOCHEM (France)
For sample digestion	Sulfuric acid 95-97% for analysis, ExpertQ®, ISO
	Hydrogen peroxide, solution 50% w/w, EssentQ®
For the Kjeldahl method	Sodium hydroxide, Pellets, Pharmpur®, Ph Eur, BP, NF
	Boric Acid, ExpertQ®, ISO, Reag. Ph Eur

All chemicals were of high purity to ensure accuracy and reliability in the results.

2.2 Sampling

The sample of *R. ribes* was collected in May, during the spring season, from the mountainous regions of Choman in South Kurdistan-Iraq. The *R. ribes* was cleaned to remove any contaminants, purified using deionized water, and then allowed to dry at room temperature. Once dried, it was powdered by a blender machine, placed in plastic containers, and stored until further analysis.

3 Experimental

3.1 Sample Preparation (ICP-OES)

0.5 g of the *R. ribes* sample was digested with 6.0 mL of concentrated nitric acid (HNO_3) and 0.5 mL of Hydrogen peroxide (H_2O_2). The microwave digestion program was set for 20 minutes at 180°C. After cooling, the solution was filtered by a Whatman No. 42 filter and diluted for ICP-OES analysis [22]. The ICP-OES procedure diagram is displayed in Figure 4.

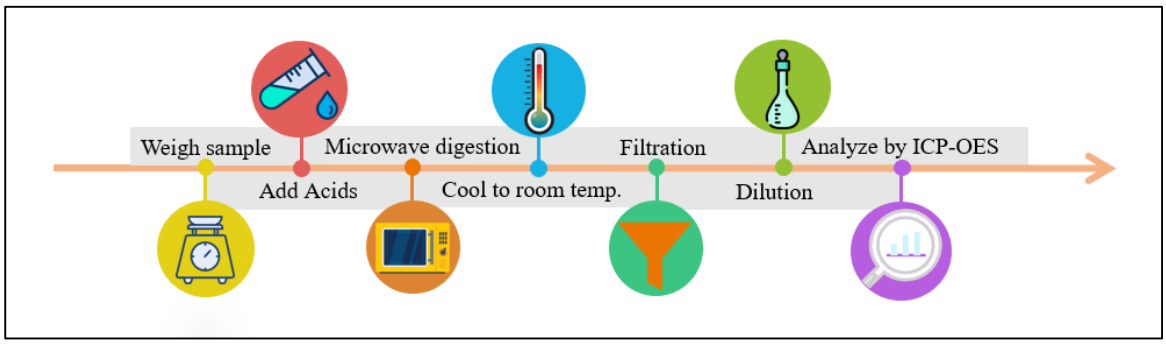

Figure 4: Sample preparation for ICP-OES analysis.

3.2 Ash content

An empty crucible (W1) and 5.0 g of the R. ribes sample (W2) were weighed, and then put in a muffle furnace set at 550°C for 5 hours. The crucible was removed after fully ignition and forming a white-gray residual, cooled down in the desiccator, and (W3) was taken [18]. (Figure 5A) Panel illustrate *R. ribes* sample before thermal incineration. And (Figure 5B) Panel shows *R. ribes* sample after thermal incineration.

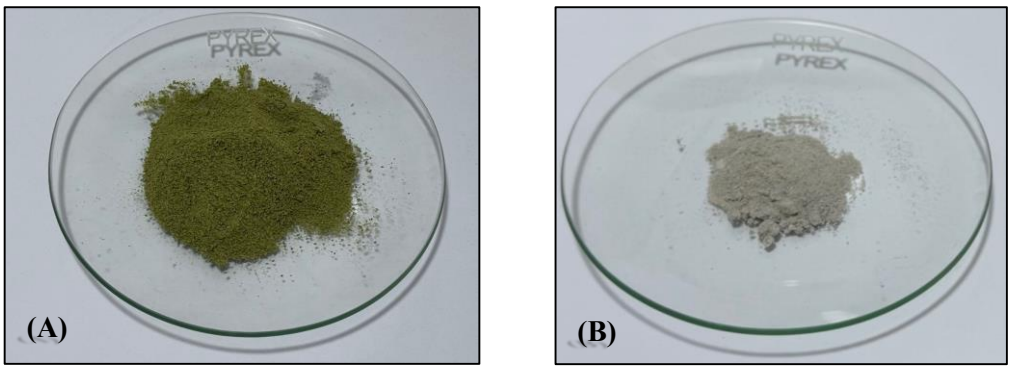

Figure 5: (A) Indicates *R. ribes* before heating. (B) shows the morphological and chemical transformations of *R. ribes* After thermal incineration treatment.

3.3 Moisture content

For determining moisture content, 2.0 g of wet *R. ribes* sample was weighed, and put in the oven for about 1.5 hours at 110°C, and allowed to cool down in a desiccator. Then the sample was weighted again to calculate moisture percentage content "from the difference in weight as" mentioned in the result section [19].

3.4 Crude protein

1.0 g of the *R. ribes* sample was digested in 10 mL of each concentrated sulfuric acid and hydrogen peroxide in a digestion instrument, which was set at 420°C for 2 hours. Then, by using the Kjeldahl method, the digested sample solution was treated with boric acid and sodium hydroxide, and then the solution was titrated with hydrochloric acid (0.2 M) [19]. After that, the percentage of nitrogen was calculated by a specific mathematical equation to find out crude protein content. Figure 6A shows the Kjeldahl technique, which is used to determine nitrogen content, and Figure 6B displays digital titration, which is used to determine the exact amount of nitrogen in the sample.

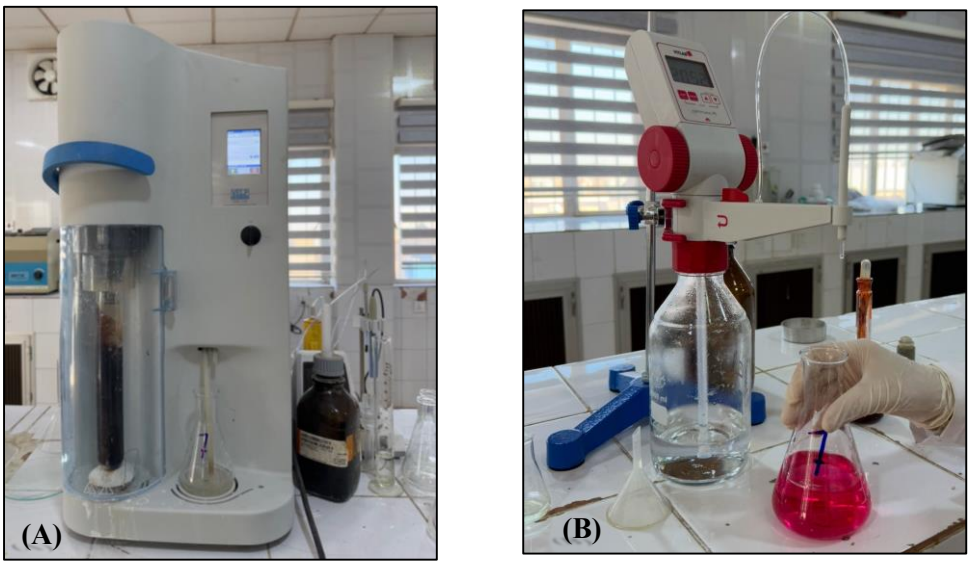

Figure 6: (A) Kjeldahl technique. (B) Titration to determine N%.

4 Results and Discussion

4.1 Metal analysis

Soil is rich in essential elements that plants absorb through their root systems. Once these elements enter the soil, they are taken up by the plants and used for their growth and development. Plant absorption of soil nutrients is specific and varies among different plants. This variation is influenced by both the unique characteristics of each plant and changing environmental conditions [23].

(Table 2) shows the mineral content of *R. ribes*, which was determined using the ICP-OES technique."

Table 2: R. ribes Minerals analysis, with (standard DF: 325.5)

No.	Minerals	Concentration (µg/mL)	SD	RSD%	LOD
1.	K	28577.680	112.28	0.39	0.0022
2.	Mg	3364.568	39.57	1.16	0.0005
3.	Na	637.207	0.465	0.07	0.0007
4.	Fe	145.755	0.270	0.19	0.0013
5.	Al	29.299	0.2933	1.00	0.0417
6.	Zn	34.437	0.117	0.34	0.0006
7.	Cu	29.038	0.328	1.13	0.0048
8.	V	25.304	0.009	0.04	0.0023
9.	Mn	22.324	0.133	0.60	0.0003
10.	Ag	2.336	0.0957	4.35	0.0036
11.	Ti	6.474	0.077	1.19	0.0007
12.	Sr	12.985	0.038	0.29	0.0001
13.	Ni	9.593	0.113	1.18	0.0004
15.	Ba	8.623	0.0215	0.25	0.0002
16.	Pb	2.170	0.001	0.04	0.0021
17.	Mo	0.293	0.002	0.79	0.0004
18.	As	0.488	0.001	0.67	0.0021
19.	Se	2.425	0.003	0.12	0.0055
20.	Li	0.109	0.004	3.81	0.0001
21.	Co	0.075	0.004	5.33	0.0006

22.	Cd	0.019	0.001	5.56	0.0005
23.	Cr	1.226	0.0197	1.57	0.0017

 # DF: Dilution Factor, SD: Standard Deviation, RSD%: Relative Standard Deviation Percentage, and LOD: Limit Of Detection.

According to the findings, results show some heavy metal concentrations in *R. ribes* such as; (Pb, Cd, and As). They follow the acceptable range specified by the United States Pharmacopoeia (USP) and the World Health Organisation (WHO), in which lead (Pb) <10 µg/g, cadmium (Cd) <3 µg/g, and arsenic (As) <1 µg/g [24].

Mayer and Vyklicky stated that these elements are important for neurochemical transmission, as well as for biological molecules such as cofactors for different enzymes and a wide range of metabolic activities. Fe, Zn, and Mn are possible antioxidants that help maintain the defences of the body [25]. The roles and the recommended dietary allowances (RDA) of the major minerals are also clearly established. Iron (8–18 mg/day) participates in haemoglobin formation, calcium (800–1300 mg/day) is critical for bones, teeth, and other tissues, and copper (1.6 mg/day) participates in iron metabolism and vascular health. Magnesium (200–400 mg/day) for enzyme action and nerve transmission, manganese (~2 mg/day) for antioxidant activity, potassium (3,500 mg/day) for muscle and nerve function, sodium (2,400 mg/day) for blood pressure and fluid balance, and zinc (8–11 mg/day) for enzyme action and cellular activity [26].

Ca, P, K, Na, and Mg are the five primary minerals, also known as macro elements, that constitute the human body. Additionally, trace elements such as (I, S, Zn, Fe, Cl, Co, Cu, Mn, Mo, and Se) play significant metabolic roles in the body. Approximately 1.5% of an adult's body weight, which amounts to 920 to 1,200 grams, is made up of calcium, with 99% of this calcium located in the bones and teeth. Phosphorus accounts for about 1% of a person's body weight. The remaining macro elements (K, Na, Mg, Cl, and S) comprise only about 0.85% of body weight. Micronutrients, which include vitamins and minerals, offer many health advantages. They contribute to tissue preservation, support the development and health of bones and teeth, and act as cofactors and coenzymes in various enzyme networks. Additionally, they assist in regulating and organizing most bodily functions, as well as other biochemical and physiological processes. Essentially, humans and other creatures require different levels of micronutrients throughout their lives to maintain various physiological processes and overall health [13].

4.2 Chemical composition

Table 3: Average chemical composition values of R. ribes

Parameters	**Values**
Total ash (%)	9.542
Moisture (%)	7.655
N (%)	4.26
Protein (%)	26.62
pH	7.4

According to Tuncturk et al., previous studies on *R. ribes* grown in Turkey reported the following values that total ash, moisture content, nitrogen content, crude protein percentage and pH of *R. ribes* which grow in Turkey, showing: 7.32%, 91.84%, 3.47%, 21.69%, 5.91, respectively. Some of our results were higher than those of the other researchers. Environmental and growing factors may cause these variations among various plant species [13].

A pH meter was used to measure the *R. ribes* pH levels [13]. The pH value of 7.4 indicates a neutral medium. The absorption of heavy metals by plants from soil is highly dependent on pH levels. Research shows that zinc (Zn) levels in plants significantly decrease when the soil pH is raised to 10. Conversely, it is generally understood that lower pH levels enhance the migration of metals in the soil to plants. Interestingly, while increasing soil alkalinity reduces zinc (Zn) levels, it promotes the absorption of copper (Cu) and magnesium (Mg) by plants [27]. Table 3 shows the results obtained from this study.

An essential initial step in proximal or particular mineral analysis is ashing [28]. Ash content describes the inorganic residue that remains in a food sample after organic materials have been burned or fully oxidized. Ash content is a crucial quality factor for some food components and is determined as part of proximate analysis for nutritional assessment [29]. The total ash was calculated by this equation;

(1) $$\%\, Ash = \frac{W3-W1}{W2} \times 100$$

$$\%\, Ash = \frac{0.4771\, g}{5\, g} \times 100 = 9.542\%$$

Food producers care about a food's moisture (or total solids) level for several reasons. Food quality, preservation, and ability to resist degradation are all significantly influenced by moisture. Moisture content must be calculated to establish the total carbohydrate amount, even if it is not included on the nutritional label [30]. A thorough moisture analysis of a food can be achieved by measuring its ability to absorb water as well as its water content [31].

This equation calculated the moisture content;

(2) $$\%\text{Moisture} = \frac{Wet\ weight - Dry\ weight}{Wet\ weight} \times 100$$

$$\%\text{Moisture} = \frac{2\, g - 1.8469\, g}{2\, g} \times 100 = 7.655\%$$

The nitrogen level of the sample was determined using the Kjeldahl technique. After evaluating the total nitrogen content, a mathematical method was used to find the crude protein concentrations. Plant composition, growth season phase, fertilization, and growing situations significantly impact the chemical constituents and nutritional content. Rich crude protein content in the plants is due to nitrogen accumulation [32].

(3) $$\text{N\%} = \frac{(Vol.\ titrated\ HCl \times Vol.\ Blank\ HCl)\ Conc.\ HCl \times \text{Molar mass of nitrogen}}{weigh\ of\ sample \times 1000} \times 100$$

$$\text{N\%} = \frac{(21.73\ mL \times 0.7ml)\ o.2M \times 14.01\text{g/mol}}{1g \times 1000} \times 100 = 4.26\%$$

Then the determined N% was Transformed protein% by utilizing the conversion factor 6.25 [3]. by this equation:

(4) $$\text{Protein \%} = \text{N\%} \times 6.25$$

$$\text{Protein \%} = 4.26 \times 6.25 = 26.62\%$$

4. Conclusion

The aim of this research was to analysis the metal and chemical composition of the *R. ribes* as a natural product and superfood, involving hidden minerals and bioactive components which can be used as traditional medicine for the treatment of many illnesses. ICP-AES was utilized to be a suitable, more accurate and sensitive technique for metallic analysis purposes. In addition, this study found that wild rhubarb *R. ribes* is the good source for nitrogen content and crude protein, which have many effective roles on human health, also the moisture content was tested, which shows the resistance of the plant product toward deterioration. *R. ribes* provides an excellent source of Fe, Zn, Mn, K, and Mg, all of which are crucial for the health of people and cattle. Maximum doses of these elements may help minimize individual risk factors for health issues, including cardiovascular disease. Growing circumstances, regional variances in soil quality, mineral absorption efficiency, and testing techniques could potentially cause these discrepancies. Herbal products may include heavy metals because they were irrigated with contaminated water and grew on contaminated soil. The *R. ribes* sample had a low concentration of lead (Pb), which is negligible from a health standpoint. There were some discrepancies between our results and those of other studies on the concentrations of minerals and heavy metals, as well as protein content. Differences in growth conditions, soil nutrient levels, mineral absorption capacity, and methods of testing can all explain these differences.

Author's Contribution

Created the idea, performed the investigation, analyzed the findings, and wrote the paper.

Conflict of Interest

This paper does not include any conflicts of interest.

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