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Evaluation of mineral, proximate compositions and anti-oxidant activities of some wild edible vegetables of District Kurram Khyber Pakhtunkhwa, Pakistan

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Abstract

This study was conducted to determine the mineral contents and some nutritional properties of five local wild vegetable; *Allium griffithianum* Boiss, *Buglossoides arvensis* (L.) I. M.Johnst., *Caralluma tuberculata* (N. E. Br.), *Chaerophyllum reflexum* Aitch and *Stellaria media* (L.) Vill., from district Kurram KP, Pakistan which are associated with folk knowledge in the region. This research is the first scientific report on the nutritional composition of the above mentioned species. Among five wild species, the *Buglossoides arvensis* had the highest carbohydrate content (71.99 ± 0.5%) and high lipid contents (4.8 ± 0.2%). Besides that, it has the highest total energy (349.024 kcal/100 g). *Stellaria media* was found to have the maximum ash (22.77 ± 0.1%) and lipids (4.87 ± 0.06%). *Chaerophyllum reflexum* showed the highest protein content (10.5 ±0.4%) and high total energy (332.68 ± 0.3 kcal/100 g). Mineral analysis showed that the local wild vegetables contained considerable amount of minerals; Calcium (2.20 ± 0.5-1506 ± 0.06 μg/g), Potassium (3.018 ± 1.7-1272.06 ± 0.005 μg/g), Phosphorous (2.98 ± 0.01-180.01 ± 0.1 μg/g), Chromium (0.98 ± 0.6-42.9 ± 0.5 μg/g), Cobalt (0.18 ±0.005-7.7 ± 0.2 μg/g), Sodium (0.78 ± 0.5-205.53 ± 0.4 μg/g) and Copper (5.5 ± 0.4-35.06 ± 0.7 μg/g). These data suggest that wild plants from district Kurram could be useful for nutrition or other applications. For instance, *Caralluma tuberculata* contains the highest number of mineral elements, which has been traditionally used as an anti-diabetic, blood purifier and for weight loss.

Keywords

Proximate analysis, Antioxidants, Wild vegetables, District- Kurram.

Introduction

All living creatures need food for survival. Increasing global population, the people have been suffering from malnutrition due to the unavailability of a balanced diet (1). So, wild vegetables could be a cheap and good source of energy for human beings, particularly in rural areas of the world (2, 3). Balanced food mainly depends upon the relative concentration of food supplements i.e. minerals, carbohydrates and vitamins in the diet (4). The vegetable food consumers are more than 85 - 90 % based mainly on staple food. Still, it is a fact that in many parts of the world, wild edible plants have been a greater contribution to the daily intake of food (5-8). It is estimated that more than 350000 various plant species have been identified, but a limited number, around 80000 are considered as safe for human consumption (2, 9).

Vegetables are the edible part of plants that are used up entirely or in part either cooked as a central part of a dish or salad. Vegetables are a good and primary source of food supplements such as carbohydrates, oil, vitamins and minerals, which may or may not be obtainable in other food sources. Vegetables constitute a more significant part of daily food intake and play a prominent role in a balanced diet, which keeps a person healthy (10).

Wild edible plants can grow in natural or semi-natural environments and can exist without dependence, and autonomously of direct human action (11-13). Around the world, one billion people harvest land crops and around 3000 species of wild plants, which have been used as wild vegetables across the globe (14-16). According to modern nutritional studies, the utilization of wild or cultivated leafy vegetables provide many health benefits, because they contain different valuable contents which are essential to human diet and in the treatment of disease as they contain vitamins, significant amount of fibers, amino acids, fatty acids and minerals (15). Wild vegetables contain different valuable contents, which is essential to the human diet and in the treatment of disease as they contain vitamins, a crucial amount of fibers, amino acids and minerals (17, 18).

According to the World Health Organization (19), minerals and essential protein deficiencies are linked to a wide variety of diseases (i.e. cardiovascular disease, diabetes, hypertensive pregnancy disorders and anaemia, premature delivery of a child, delayed sexual maturation and behavioural changes); particularly in developing and under-developed countries (15). In low-income countries, diets are dominated by a single staple food item and shared with small amounts of other food products, resulting in a monotonous diet and high risk of inadequate intake of macronutrients and micronutrients (15, 20).

In the developing world to take a balanced diet in daily routine is not easy; due to low-income, high population and living expenses. To overcome minerals and protein deficiencies in our bodies, we could easily include wild vegetables in daily consumption. The wild plants could naturally and easily be available, which are rich in carbohydrates, protein, enzymes, fibers, pigment, minerals and vitamins such as A, C, B1, B2, B6, niacin, iodine, potassium, iron, magnesium, manganese and calcium (21, 22). This is why wild plants are now consumed around the world for their nutritional value. These plants could offer an alternative ingredient compared to other foodstuffs (23). The nutritional components of wild vegetables depend upon the plant families, geography, season and climatic situation of habitat (24, 25). The aim of this research was to investigate the mineral contents and some nutritional properties of 5 uninvestigated wild plants (*Stellaria media*, *Buglossoides arvensis*, *Caralluma tuberculata*, *Chaerophyllum reflexum* and *Allium griffithianum*), which have been consumed as vegetables by local communities of the district of Kurram.

Materials and Methods

The determination of different nutritional parameters such as moisture content, ash%, crude fibers%, crude protein%, crude lipid%, carbohydrates%, and total energy was carried out using the methods of Association of Official Analytical Chemists method (AOAC) (26). While minerals analysis was carried out using Atomic Absorption Spectrometry (AAS): calcium, magnesium, iron, manganese, zinc, chromium, cobalt and copper were analyzed using AAS, while Sodium and potassium were analyzed by flame photometry and phosphorous was analyzed by colorimetric method.

Selected geographical region of the study

The district of Kurram is 115 km long and has a total area of 3380 sq. km, which has a unique geographical location attached by the province of Paktia, Nangarhar in the West and North, Orakzai, Khyber districts in the East and Hangu district in the South East. It is attached to North Waziristan in the south (27). The climate of the Kurram valley remains pleasant for most of the summer, but in winter, the minimum temperature drops below freezing 0 (28, 29). These climate and weather uniqueness of Kurram profoundly affects the nutritional components of the wild vegetables. The river Kurram highly promotes bountiful growth of vegetation (30, 31). However, in this district of Khyber Pakhtunkhwa, Pakistan, due to its complex geographical nature, the area had not been widely explored for the medicinal and nutritional benefits.

Plant collection

Five wild green vegetables *Allium griffithianum* Boiss*, Buglossoides arvensis* (L.) I. M.Johnst.*, Caralluma tuberculata* (N. E. Br.), *Chaerophyllum reflexum* Aitch and *Stellaria media* (L.) Vill., were collected during the period of their consumption from different areas of Kurram. The plants were identified and submitted in the Department of Botany, GPGC (Government Postgraduate College) Parachinar district, Kurram.

Local consumers helped a lot in the collection of these wild vegetables investigated during fieldwork. During the specimen collection, vegetative parts were gathered for the exploration. We put all these plants in zippered bags immediately after its collection. Every specimen was recorded and labelled with a comprehensive note in a specimen collection notebook to maintain proper information labelled during the collection and processing period. Some of the selected wild vegetables for mineral and proximate compositions are shown in Fig. 1.

Sampling and Proximate analysis

Plants were washed three to four times with tap water to clean the adhering materials and placed in the shade for drying. After drying, the samples were ground into powders then packed in plastic bags. Proximate analysis was carried out according to the Association of Official Analytical Chemist method (26).

Moisture content

Moisture content was determined according to the standard procedure (32). Two g of each dried sample was placed in an electric oven (Precision Thelco no 17) at 105 ^{0}C for 4-6 hrs. Then transferred the samples into a desiccator and cooled it down for 20 min. Again, weighed and calculated the % moisture by the following formula,

$$Moisture\ (\%) = \frac{Wm_1 - Wm_2}{\text{Weight of sample}} \times 100, \quad \text{(Eqn. 1)}$$

where Wm_1 is the initial weight of the sample taken for moisture measurement and Wm_2 is the weight of the sample after drying it in the oven.

Crude fiber

Acid and alkali digestion was used to determine the crude fiber (34). In acid digestion, 200 ml of hydrochloride (HCl) with 2 g of each sample boiled on a water/steam bath at 90-95 ^{0}C for half an hr. The filtrate was collected and washed with hot water until it became acid-free, using a muslin cloth. The acid-free filtrate was treated with 200 ml of 2.5% Sodium hydroxide (NaOH) (Alkali digestion) and again kept on steam/water bath at the same temperature for half an hr and filtered the residue; washed thoroughly with hot water. The washed residue was transferred to the crucible to remove moisture in an oven at 105 ^{0}C for 4 hrs and then

Fig. 1. Selected wild vegetables **(a)** *Allium griffithianum* Boiss **(b)** *Buglossoides arvensis* (L.) I. M.Johnst **(c)** *Caralluma tuberculata* N.E Br **(d)** *Chaerophyllum reflexum* Aitch **(e)** *Stellaria media* (L.) Vill **(f)** experimental laboratory.

Ash content

The ash content of wild plants was determined according to the AOAC method (33). We placed 2 g of each dry sample in an electric furnace (i.e. VULCAN 3-130) at 550 ^{0}C for 6 hrs. The following formula calculated the ash %.

$$Ash\ (\%) = \frac{Wa_1 - Wa_2}{\text{Weight of sample}} \times 100, \quad \text{(Eqn. 2)}$$

where Wa_1 is the initial weight of the sample take for ash measurement and Wa_2 is the weight of the sample after combustion.

cooled in a desiccator. After cooling, it was weighed (Wf_1) and then placed in a furnace for four hours at 550-600 ^{0}C until it became grey-whitish ash. Desiccator was used to cool and weight again. Following formula was used to calculate the crude fiber.

$$Crude\ fiber\ (\%) = \frac{Wf_1 - Wf_2}{Wf} \times 100 \quad \text{(Eqn. 3)}$$

where, Wf is the weight of the sample taken for calculating crude fiber percentage, Wf_1 is the weight of oven-dried residue and Wf_2 is the weight of residue after the furnace.

Crude protein

We followed the Kjeldahl method to discover crude protein in terms of nitrogen content (34, 35). One g of the sample, digestion mixture Potassium sulphate: Copper sulphate (K_2SO_4: $CuSO_4$) 7:1 of 8 g and concentrated Sulfuric acid (H_2SO_4 12 ml) was digested on digester heater under a fume hood until the sample became clear and transparent (blue-green color). Distilled water was added to digest to make 100 ml volume.

The Kjeldahl apparatus was used to carry out the distillation of the digested sample. In this process, 10 ml of the digest sample, along with 10 ml of Sodium hydroxide (NaOH 40%), were appended. NH_3 produced in receiving flask, collected as Ammonium hydroxide (NH_4OH). Four percent (4%) Boric acid (H_3BO_3) solution of 20 ml and a meagre amount of methyl red indicator was added to the flask. As a result, the pink color of NH_4OH changed into yellowish during distillation (17).

When the pink color becomes visible, the distillate was titrated against standard zero of N (normality) HCl. A blank was also run. Crude protein was calculated as follows

$$Crude\ lipids\ (\%) = 6.25\frac{(S-N)\times N\times 0.04\times D}{Weight\ of\ sample\ \times V}\times 100, \quad \text{(Eqn. 4)}$$

where value 6.25 represents protein conversion factor for plants; S is the sample titration reading; N is the normality of HCI; D is the dilution of the sample after digestion; V is the volume distillation.

Crude lipids

For the determination of crude lipids, we used Soxhlet's apparatus. Moisture free biomass (1 g) was placed in a cellulose cartridge in ' 'Soxhlet's apparatus. 100 ml of petroleum ether were used to carry on the extraction for 6 hrs at the 40-60 ^{0}C (2, 36). The solvents were evaporated on a rotary evaporator to remove the solvent and obtain the crude lipids. The flask was kept to cool, to weight it correctly. The contrast among in weights was expressed as % crude lipids using following formula.

$$Crude\ lipids\ (\%) = \frac{Wl_1 - Wl_2}{\text{weight of sample}}\times 100, \quad \text{(Eqn. 5)}$$

where Wl_1= weight of flask with fat and Wl_2= weight of empty flask.

Carbohydrate content (%)

The total available carbohydrates of each sample were calculated as nitrogen-free extraction. The carbohydrates contents were found out by deducting the total amount of moisture, ash, fat, protein and fiber from 100 (AOAC) (2, 33, 34).

All the samples were run in triplicate. And all of the above contents were expressed on a dry matter basis.

$$\text{Carbohydrates}(\%) = 100 - (\%\ \text{crude protein} + \%\ \text{crude fat} + \%\ \text{crude fiber} + \%\ \text{ash content}). \quad \text{(Eqn.6)}$$

Total energy content

The total energy value or calorific value of the selected plants in kcal/100 g was calculated by the following formula reference by (33, 37).

$$\text{Total energy (kcal per100g)} = (\%\ \text{protein} \times 4.1) + (\%\ \text{lipids} \times 9.3) + (\%\ \text{carbohydrates} \times 4.1) \quad \text{(Eqn.7)}$$

As one g of protein contributes 4.1 kcal of energy, 1 g of lipids contributes 9.3 kcal energy and 1 g of carbohydrates contributes 4.1 kcal energy.

Mineral analysis

Before analysis, plants were oven-dried at 65 °C for a period of 72 hrs. Clean and clear polythene bags were used to store the powdered plant material. One g of each dried sample was put in the flask and digested by 12 ml of concentrated Nitric acid and left overnight. To this solution, 5 ml of per-chloric acid was added, and heated on hot plates for 20 min until the solution seems to be transparent. The sample was cooled and then filtered through filter paper No. 42. On the completion of the filtration, the filtrate was then poured into a 100 ml volumetric flask. Five samples solution were investigated for Ca, Mg, Fe, Mn, Cu, Zn, Cr, Na, K and Co elements by Atomics Absorptions Spectrometers (Shimadzu AA-670). As the atom absorbed light energy of a specific wavelength, it entered from the ground state to the excited state. Quantitative determination of the amount of analyte can be made when the numbers of atoms in the light path are absorbed. Atomic absorption spectroscopy with an air acetylene flame, laminar flow burner and hollow cathode lamps are used to determine above mentioned elements using the absorption of optical radiation by free atoms in the gaseous state (37). The acid digest was sucked by means of a capillary tube into the flame. The device was set for analysis of each element by setting a light source of the desired element. The instrument calibration was performed, introducing standard and digest samples into the device (38). On-screen, the absorbance readings appeared were noted and used to calculate the concentration of each element as follows,

$$\mu\frac{g}{g} = \frac{Instrumental\ reading\ \times Dilution\ factor}{\text{weight of sample}}. \quad \text{(Eqn. 8)}$$

For phosphorus determination, the mixed reagent was made from "ammonium molybdate (6g/125 ml distilled water), sulfuric acid and antimony potassium tartrate (146/500 ml of 5N sulfuric acid)". For the color reagent, 370 mg of ascorbic acid was dissolved in the mixed reagent. Using a spectrophotometer (Spectronic SP3000 Japan) on the basis of molecular absorption spectrophotometry (26), a standard solution was prepared from phosphorous salt. Potassium dihydrogen phosphate (KH_2PO_4), 439 mg was solvated in distilled water. The mixture was equivalent to a concentration of 100 mg/l i.e. 100 ppm by a dilution formula $C_1V_1 = C_2V_2$ (C_1 = concentration of starting solution, V_1= volume of the starting solution, C_2 = final concentration of the solution and V_2 = final volume of the solution).

We diluted the solution to 2, 4, 6 and 8 ppm and transferred 1 ml of each to 25 ml of the volumetric flask. Then, color reagent of 4 ml was added to it and by adjusting the volume with distilled water up to the mark. It was lifted until the bluish color was developed. The absorption of these solutions against the blank was determined by the spectrophotometer. These are used in the preparation of the standard curve.

The amount of phosphorous was calculated by the following formula

$$Amount\ of\ P = \frac{Graphical\ reading\ \times Dilution\ factor}{\text{weight of sample}}$$

(Eqn. 9)

Antioxidant assays

Determination of % free radical scavenging activity (% FRSA)

The free radical scavenging activity (FRSA) of wild vegetables was assessed by monitoring their ability to extinguish stable 2, 2-diphenyl 1-picrylhydrazyl (DPPH) free radicals. A DPPH solution was prepared by dissolving 3.2 mg DPPH in 100 ml of 82% methanol. Then, 2800 µl of DPPH solution was added to glass vials followed by the addition of 200 µl of Crude methanolic extracts (CME) solution in methanol, leading to the final concentrations of 100 µg/ml, 50 µg/ml, 25 µg/ml 10 µg/ml, 5 µg/ml, 2 µg/ml and 1 µg/ml. The mixtures were shaken and kept in the dark at a controlled room temperature (25 °C–28 °C) for one hour. Absorbance was measured at 517 nm by using a spectrophotometer (DAD 8453, Agilent). Methanol (82%) was used as a blank, while a mixture of 200 µl of methanol and 2800 µl of DPPH solution was taken as a negative control. Ascorbic acid was used as a positive control. Each test was performed in triplicate and the inhibition % was measured according to the formula given below:

$$\%FRSA = 1 – Ab_s/Ab_c \times 100$$

Where, Ab_s absorbs the test sample, while Ab_c absorbs the negative control containing DPPH and methanol instead of the sample.

Total antioxidant capacity (TAC)

Phosphomolybdenum-based chlometric assay was performed to determine the total antioxidant potential and was expressed according to the microgram equivalent microgram acid per mg of dry plant weight (g aAE / mg DW). Each test extract contains 0.1 ml (4 mg / ml DMSO) and positive control (ascorbic acid, 4 mg / ml DMSO) to 0.9 ml reagent (0.6 M sulfuric acid, 28 mm sodium phosphate) and was mixed with (4 mm) ammonium (molybdate solution in H2O). The blank contained 0.9 ml of reagent solution and 0.1 ml of DMSO without any liqueur. All tubes were placed in a water bath for 90 min at 95 °C and then cooled to room temperature from where 200 µl of each sample was thoroughly transferred to the 96 well plates. The gravity was measured at 630 nm using a microplate reader (Biotech USA, Microplate Reader Alex 800). A calibration curve of ascorbic acid (y = 0.0212 × + 0.0926, R2 = 0.9913) with a final number of 100, 50, 25, 12.5, 6.25, 3.12 / g / ml was prepared and the experiment was performed in a repetitive form.

Analysis and evaluation

Proper exposition of nutrient examination results cannot be achieved unless appropriate sampling methods have been used. The plant age can have a notable effect on various nutrients amount. Plant's growth continually influences nutrient levels until it reaches its critical point. We collected the samples from a wild vegetable at its middle age before they reach their critical point of growth age. All the determination was carried out in triplicate and the results of the samples were subjected to standard error and means statistically in Microsoft software excels.

Results

Proximate and mineral analysis such as ash, moisture, protein, fats, fiber, carbohydrates and energy; macro, micro and trace element were estimated in the chosen wild vegetables (i.e. *Allium griffithianum, Buglossoides arvensis*, *Caralluma tuberculata*, *Chaerophyllum reflexum* and *Stellaria media*). Proximate analyses were carried out according to the standard methods of (AOAC, 2000) and Atomics Absorptions Spectrometers (Shimadzu AA-670) for minerals analysis.

Total moisture, ash, fiber, protein, lipids, carbohydrates and calorific contents were reported as the % composition, according to the methods mentioned above. Table 1 summarized the moisture, ash, fiber, protein, lipids, carbohydrates and calorific content in (%) of the selected wild vegetables.

Mositure

The moisture amount in the selected 5 wild vegetables ranged from (11.85% to 8.95%). The highest moisture was detected in *Allium griffithianum* (11.85%) and the lowest was observed in *Stellaria media* (8.95%). The moisture content of *Buglossoides arvensis*, *Caralluma tuberculata* and *Chaerophyllum reflexum* was found 11.44%, 9.90% and 10.89% respectively as shown in Table 1.

Ash content

Ash content ranged from 11.38% to 22.77% was found maximum in *Stellaria media* (L.) (22.77%) and lowest in *Allium griffithianum* Boiss (11.38%). Other wild vegetables *Buglossoides arvensis* (L), *Caralluma tuberculata* (N.E.Br) and *Chaerophyllum reflexum* (Aitch) contained (17.5%), (15.76%) and (12.07%) respectively.

Crude fiber

Crude fiber contents were found varied from 3.46% to 27.36%. The highest materials of fibers were recorded in *Caralluma tuberculata* and lowest in *Buglossoides arvensis* while in *Stellaria media*, *Chaerophyllum reflexum* and *Allium griffithianum* were 12.13%, 10.44% and 7.96% in order.

Crude protein

Chaerophyllum reflexum ranked the highest crude protein (10.5%) and the lowest crude protein content was observed in *Buglossoides arvensis,* which is (5.25%). The crude protein content was found similar in *Caralluma tuberculata* and *Allium griffithianum* that is (8.75%). *Stellaria media* have the third-highest protein content among other wild vegetables as shown in Table 1.

Table 1. Proximate composition in the local wild plants (% dry matter). All the data expressed as means ± SD of the triplicate experiments. All of the below contents (i.e., Ash, Crude fiber, crude protein, crude lipids, and Carbohydrates) are expressed on a dry matter basis

Plant name & vouchar number	Family	Local name	Consumed part	Moisture (%)	Ash (%)	Crude fiber (%)	Crude protein (%)	Cruid lipids (%)	Carbo-Hydrates (%)	Total energy
Allium griffithianum Boiss Anwar .05.GPGC.PCR	Amaryllidaceae	Pizaki	Whole plant	11.85 ± 0.4	11.38 ± 0.4	7.96 ± 0.4	8.75 ± 0.4	1.88 ± 0.1	70.03 ± 0.5	34.48 ± 1.7
Buglossoides arvensis (L.) I.M.Johnst., Anwar .02.GPGC.PCR	Boraginaceae	Noraki Saba	Leaves	11.44 ± 0.5	17.5 ± 0.3	3.46 ± 0.1	2.25 ± 0.1	4.8 ± 0.2	71.99 ± 0.5	34.02 ± 1.2
Caralluma tuberculate N.E.Br, Anwar .03.GPGC.PCR	Apocynaceae	Pawani	Whole plant	9.9 ± 0.6	15.76 ± 0.1	27.36 ± 0.8	8.75 ± 0.1	4.77 ± 0.1	43.36 ± 0.9	25.01 ± 3.6
Chaerophyllum reflexum Aitch.,Anwar .04.GPGC.PCR	Apiaceae	Zanky	Leaves, young stem	10.89 ± 0.1	12.07 ± 0.1	10.44 ± 0.2	10.5 ± 0.4	2.88 ± 0.2	64.11 ± 0.8	33.68 ± 0.3
Stellaria media (L.) Vill., Anwar.01.GPGC.PCR	Caryophyllaceae	Wilaghari/ Badshah Saba	Leaves, young stems	8.95 ± 0.1	22.77 ± 0.2	12.13 ± 0.017 ± 0. 5		4.87 ± 0.1	53.23 ± 0.4	29.23 ± 0.97

Crude fat

The crude fat content in *Stellaria media*, *Buglossoides arvensis* and *Caralluma tuberculata* were detected at a close value with each other while in *Chaerophyllum reflexum* and *Allium griffithianum* contained (2.88%) and (1.88%) crude fat respectively.

Carbohydrate

The carbohydrates concentration was recorded 53.23 % in *Stellaria media*, 71.99 % in *Buglossoides arvensis,* 43.36% in *Caralluma tuberculata,* 64.11 % in *Chaerophyllum reflexum* and 70.03 % in *Allium griffithianum*. The maximum concentration of carbohydrates was noted in *Buglossoides arvensis,* which is 71.99 %, while the lowest value was detected at 43.36 % in *Caralluma tuberculata*.

Energy contents

The total energy content of *Buglossoides arvensis* showed the highest energy level 349.024 kcal/100 g and *Caralluma tuberculata* attended to the lowest level 258.012 kcal/100 g, as shown in Table 1. While the other three wild vegetables also showed a significantly higher value of energy that is 340.485 kcal/100 g, 332.685 kcal/100 g and 292.234 kcal/100 g in *Allium griffithianum, Chaerophyllum reflexum* and *Stellaria media* respectively.

Minerals composition

Minerals are inorganic mass found almost in every tissue and fluid of the body. Their presence is required to keep body chemicals and physical functions normal. The minerals may be macro, micro and trace. The trace minerals are needed for the body in less than a few mg per day in quantity.

Macro minerals

Calcium content

The results of the Calcium (Ca) of selected wild vegetables are shown in Table 2. They ranged from (2.195 µg/g) in *Stellaria media* up to (1506 µg/g) in *Caralluma tuberculata* which was followed by *Buglossoides arvensis* (459.34 µg/g), and then *Allium griffithianum* (405.16 µg/g), and the content of calcium in *Chaerophyllum reflexum* showed (19.13 µg/g).

Magnesium content

The result showed that Magnesium (Mg) ranged from (0.635 µg/g) to (368.03 µg/g) in selected samples. The maximum Mg content was reported in *Buglossoides arvensis* (368.03 µg/g). The remaining plants, i.e. *Caralluma tuberculata* show (176.45 µg/g), *Allium griffithianum* show (405.16 µg/g) and *Chaerophyllum reflexum* have (16.103 µg/g) content of Mg. The lowest Mg content was reported in *Stellaria media* (0.635 µg/g) as displayed in Table 2.

Phosphorous content

Phosphorous (P) is required for different functions and development in the body. From Table 2 it is clear that *Caralluma tuberculata* has the highest amount of phosphorous (180.01 µg/g) as compared to all others. *Allium griffithianum* has second-highest phosphorous content (61.26 µg/g), *Buglossoides arvensis* has the third (41.43 µg/g) while are the fourth-highest content followed by *Stellaria media* and *Chaerophyllum reflexum* with (2.98 µg/g).

Potassium content

The highest level (1272.06 µg/g) of Potassium (K) was in *Caralluma tuberculata*. The results were followed by *Allium griffithianum* (426.061 µg/g), *Buglossoides arvensis* (87.90 µg/g), *Stellaria media* (5.775 µg/g) while the lowest K content was found in (3.018 µg/g) in *Chaerophyllum reflexum* as showed in Table 2.

Micro minerals

Iron content

The highest (1050.5 µg/g) amount of Iron was recorded in *Buglossoides arvensis* which was followed by *Allium griffithianum* with (1002.021 µg/g), *Stellaria media* with (901.76 µg/

Table 2. Composition of mineral elements in the local wild plants (µg/g dry weight). The data are averages of three replicates with ± standard error (SR)

Mineral elements	Stellaria media	*Buglossoides arvensis*	*Caralluma tuberculata*	*Chaerophyllum reflexum*	*Allium griffithianum*
Calcium (Ca)	2.2 ± 0.5	459.34 ± 0.2	1506 ± 0.1	19.13 ± 0.1	405.16 ± 0.5
Magnesium (Mg)	0.635 ± 0.01	368.03 ± 0.6	176.45 ± 0.1	16.103 ± 0.3	405.16 ± 0.6
Phosphorus (P)	2.895 ± 0.3	41.43 ± 0.7	180.01 ± 0.1	2.98 ± 0.01	61.261 ± 0.1
Potassium (K)	5.775 ± 0.3	87.9 ± 0.4	1272.06 ± 0.1	3.018 ± 0.1	426.061 ± 0.5
Iron (Fe)	901.76 ± 0.3	1050.5 ± 0.4	249.61 ± 0.3	321.8 ± 0.5	1002.021 ± 0.1
Zinc (Zn)	70.01 ± 0.6	23.1 ± 0.6	49.26 ± 0.1	18.02 ± 0.1	46.03 ± 0.3
Chromium (Cr)	38.27 ± 0.6	6.2 ± 0.5	42.9 ± 0.5	0.98 ± 0.1	27.604 ± 0.7
Cobalt (Co)	2.1 ± 0.6	0.18 ± 0.1	7.703333 ± 0.2	1.28 ± 0.1	3.69 ± 0.1
Manganese (Mn)	130.95 ± 0.3	10.68 ± 0.4	113.85 ± 0.3	17.06 ± 0.1	96.054 ± 0.2
Sodium (Na)	0.335 ± 0.1	15.87 ± 0.3	205.5 ± 0.4	0.78 ± 0.1	45.88 ± 0.4
Copper (Cu)	9.586667 ± 0.4	5.5 ± 0.4	35.06 ± 0.7	13 ± 0.1	19.012 ± 0.2

g) and *Chaerophyllum reflexum* (321.8 µg/g). The lowest iron content was recorded in *Caralluma tuberculata* with (249.61 µg/g) as shown in Table 2.

Zinc content

The result of Zinc (Zn) contents is shown in Table 2. The content of zinc ranged from (70.01 µg/g) to (18.02 µg/g). Highest zinc content (70.01 µg/g) was found in *Stellaria media*. In contrast, the lowest zinc content was recorded in *Chaerophyllum reflexum* (18.02 µg/g). *Buglossoides arvensis, Caralluma tuberculata* and *Allium griffithianum* zinc content were found (23.1 µg/g), (49.26 µg/g) and (46.03 µg/g) respectively.

Chromium content

Among the selected wild vegetables, Chromium (Cr) was present in the highest amount in *Caralluma tuberculata* (42.9 µg/g), then the higher value contained by *Stellaria media* (38.27 µg/g), which is followed by *Allium griffithianum* (27.604 µg/g), while *Buglossoides arvensis* with (6.20 µg/g) and the *Chaerophyllum reflexum* with (0.98 µg/g).

Cobalt content

The result showed that the Cobalt (Co) amount of these selected wild vegetables ranged from (7.7 µg/g) to (0.18 µg/g) in the selected wild vegetables. From the result, it was concluded that the highest content (7.7 µg/g) of Cobalt is present in *Caralluma tuberculata* while others in descending order as *Allium griffithianum* (3.69 µg/g), *Stellaria media* (2.10 µg/g), and *Chaerophyllum reflexum* (1.28 µg/g). The lowest content (0.18 µg/g) was observed in *Buglossoides arvensis.*

Manganese content

The current results showed that Manganese (Mn) concentration ranged from (10.68 µg/g) to (130.95 µg/g) in five selected wild vegetables. The highest level (130.95 µg/g) was found in *Stellaria media* followed by *Caralluma tuberculata* with (113.85 µg/g), *Allium griffithianum* with (96.054 µg/g) and *Chaerophyllum reflexum* with (17.06 µg/g). At the same time, the lowest Mn content was evident in *Buglossoides arvensis* (10.68 µg/g).

Copper content

It is an essential trace having fundamental importance in the establishment of red blood cells, maintaining nerve cells, and the immune system. From the result of 5 selected wild vegetables, the copper content was found to be highest in *Caralluma tuberculata* (35.06 µg/g) which was followed by *Allium griffithianum* (19.012 µg/g), *Chaerophyllum reflexum* (13.0 µg/g), *Stellaria media* (9.32 µg/g) and lowest in *Buglossoides arvensis* with (5.5 µg/g) as shown in Table 2.

Trace mineral

The Sodium (Na) contents of wild vegetables are presented in Table 2, which shows it ranged from (0.78 µg/g) in *Chaerophyllum reflexum* up to (205.53 µg/g) in *Caralluma tuberculata* followed by *Allium griffithianum* with (45.88 µg/g) and then, *Buglossoides arvensis* which had (15.87 µg/g) content. *Stellaria media* showed (0.335 µg/g) content of Na.

Antioxidant assays

Determination of % Free radical scavenging activity (% FRSA)

The percentage of free radical scavenging activity (% FRSA) of the selected wild vegetables samples, calculated by measuring the discoloration of the DPPH solution is shown in Fig. 2. The test protocol is based on the conversion of the stable purple colored DPPH radical into its diphenylpicryl yellow colored hydrazine molecule accepting electrons or hydrogen radicals from the donor antioxidant. The DPPH molecule is considered as a stable free radical due to the presence of a delocalized reserve electron over the entire molecule which provides a characteristic absorption band at 517 nm. Highest free radical scavenging activity was shown by ethanol extract of *Allium griffithianum* which was 44%, followed by *Stellaria media* 24%, *Caralluma tuberculata* 22%, while the lowest activity shown in the wild vegetables of *Chaerophyllum reflexum* 12% followed by and *Buglossoides arvensis* which is 18%.

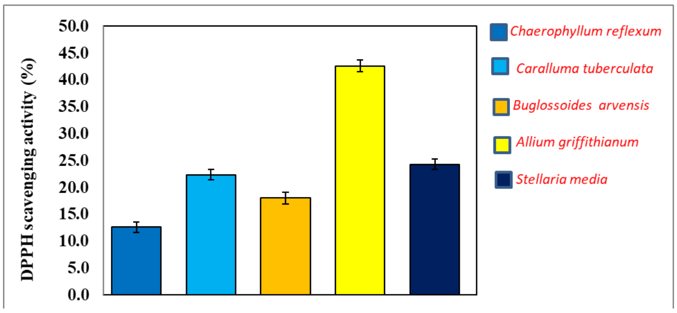

Fig. 2. Showing the %FRSA activity in selected wild vegetables.

Total antioxidant capacity (TAC)

The total antioxidant capacity of the wild vegetables estimated according to the standard methods (54, 55). In these wild vegetables all showed a significant difference among each other in the total antioxidant capacity. Among these 5 wild vegetables the wild vegetables *Chaerophyllum reflexum* showed the highest value that is 1.49 mg/ml followed by *Caralluma tuberculata* and then *Buglossoides arvensis* as shown in Fig. 3. While the wild fruit *Allium griffithianum* showed the lowest value (0.74 mg/ml) among all 5 fruits followed by wild vegetable *Stellaria media.*

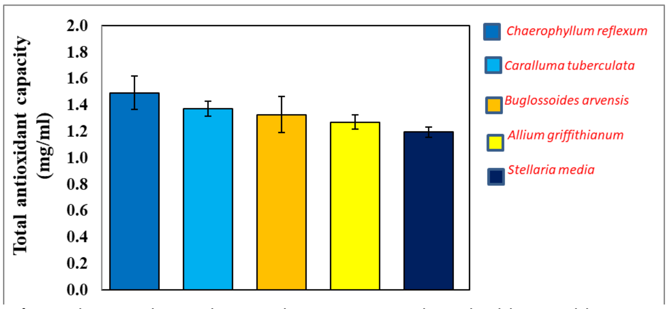

Fig. 3. Showing the total antioxidant activity in selected wild vegetables.

Statistical Analysis

The standard error declares the scatteredness in the proximate composition of the five selected wild vegetables. This study found the highest scatteredness in the *Caralluma tuberculata*. However, the vibration in the moisture and ash is negligible for all the 5 wild vegetables.

The scatteredness in the *Stellaria media* triplicates is higher for Co, Cr, Zn and Ca. However, the lower standard error (0.002 and 0.003 for Na and P respectively) in *Stellaria media,* reveal that all the triplicates of *Stellaria media* have almost same composition of Na and P. The 3 copies of *Buglossoides arvensis* show higher standard values for all the elements, other than Co, which disclose a clear difference in the composition of above mentioned elements. In the triplicates of *Caralluma tuberculata*, the composition of Mg, Fe, Cr, Na, Mn and Cu show the great difference as the standard value for these elements is high. However, the composition of K and Ca remains the same for all the triplicates as the standard error for them is lower. The standard error of elements composition in the triplicates of *Chaerophyllum reflexum* shows a higher difference in Fe and Mg. However, the composition is almost similar to all the other elements as their standard errors are negligible. For *Allium griffithianum*, the compositions of the elements in the triplicates are highly different as the standard error of Ca, Mg, K, Cr and Na are higher.

Discussion

The current study was carried out to correlate the chemical constituents of selected wild vegetables with their beneficial uses. The selection of the wild vegetables was carried out through available literature (22, 25, 29, 33, 38 – 41). The result obtained from the proximate analysis showed that the highest moisture content was found in *Allium griffithianum* (11.85%) on a dry basis. Moisture is regarded as an important source of water. It is needed that 20 % of the total consumption of water must be from food moisture (33). The moisture content observed in this study was higher than the moisture content (22). Like moisture, the ash content is also essential biochemically. Among the selected wild vegetables, the ash content was observed high 22.77% in *Stellaria media.* The ash content of these wild vegetables was also higher than the results reported (2, 17). The fiber content of *Caralluma tuberculata* was high at (27.36%). It plays a vital role in preventing heart attacks, obesity, intestinal cancer, serum cholesterol and hypertension (2). Traditionally *Caralluma tuberculata* was reported for the cure of diabetes, high blood pressure, intestinal cleaner and anti-rheumatic (42). The values reported during the current results were standard as compare to the reported values from different parts of Pakistan and the world, while few studies were not in line with existing results (34, 43). Agriculture is the primary source of food and income for the people of Kurram. Among the population of Kurram, 70-80% depends on wild vegetables and farm-grown crops for daily bread/income and fodder for their domestic animals. Among the tested wild vegetables, the highest crude protein was found in *Chaerophyllum reflexum* (10.5%); this result revealed that these are a good source of energy-rich compounds i.e. protein. Leafy vegetables with high crude protein content can be used as a source of inexpensive protein (44). Protein is vital for growth, resistance to infections, replacement of used up blood and one g of protein provides about 4 kcal energy to the body (45). The data of the present work is supported by (31), who reported the protein content of the wild vegetables consumed in the Lebialem Highlands south-western Cameroon. Fat content in the diet is regarded as a major source of energy, as vegetables with fat are recommended for obesity. The higher crude fat content was reported in *Stellaria media* (4.87%), and ethnically *Stellaria media* is used as food in the rural area of the understudy. By reviewing the literature, it has been confirmed that present results are in line with (34). The present study revealed that nitrogen-free extract (NFE) was found as the main constituent of the proximate analysis of the selected wild vegetables. Due to the higher nitrogen-free extract in these species, they are considered a good source of food. The maximum carbohydrates (71.99%) were observed in *Buglossoides arvensis*. It has a specific cultural interest to present during a special religious and cultural festival of EID-e-NOWROZ and also has medicinal value as used for the cure of anaemia (29). The present results are in line with (17), whose reported carbohydrate content in wild vegetables was in the range of 52.78 to76.34 %). Higher energy values propose that these plants may be used in the formulation of various dietary supplements. The maximum amount of energy was recorded in *Buglossoides arvensis* vs. 349.024 kcal/100 g. These results are in line with (31) reported by whose results ranged from 109.78 kcal/100 g to 364.98 kcal/100 g. The standard deviation shows the scatteredness in the proximate composition of wild vegetables. The study analyses the highest deviation in the energy content and carbohydrates; however, the variation in the moisture content and ash is negligible for all the 5 wild vegetables.

The mineral composition was analyzed to find the correlations between traditional knowledge of wild vegetables and their mineral composition such as calcium, magnesium, phosphorous, potassium, iron, zinc chromium cobalt, manganese, sodium and copper. The calcium concentration was found highest in *Caralluma tuberculata* (1506 µg/g). Our results are very co-related to some of the wild vegetables consumed in Nigeria and North-eastern part (a hilly and rainy area like Kurram) of the India but much higher than the vegetables consumed as food in some other part of Pakistan (36, 46, 47). Similarly, magnesium concentration was recorded maximum in *Buglossoides arvensis,* i.e. (378.03 µg/g). In the comparison of some leafy vegetables as referenced in (46), the content of magnesium in wild vegetables of Kurram was much higher. The maximum amount of phosphorus was found in *Caralluma tuberculata* (180.01 µg/g). Phosphorus is an important element that plays a role in various normal physiological functions of the body. A study from Turkey (48) reported similar phosphorus results for the same wild edible vegetables grown there. The average amount of potassium and sodium was reported in selected wild vegetable species. Both potassium and sodium ratio helps in preventing high blood pressure in the human body (49). The concentration of Iron in these vegetables was found to be highest in *Buglossoides arvensis* (1050 µg/g). in the formation of haemoglobin, Iron is considered as an essential trace element. It transports oxygen to different parts of the body and guarantees a healthy immune system (50). Our results indeed agree with folk knowledge reported that the *Buglossoides arvensis* is locally used in the remedy for the treatment of anaemia (29). The zinc content in *Caralluma tuberculata* was recorded at (49.26 µg/g). "It plays a vital role in diabetes as a cofactor for insulin but at an optimum concentration" (43), which is in line with the findings that *Caralluma tuberculata* is used for diabetes in the district of Kurram (29). The highest amount of Cr was reported, i.e. (42.9 µg/g) in *Caralluma tuberculata.* The highest amount of the Co was also found in *Caralluma tuberculata* 7.7 µg/g. "It is an essential trace element for the human body, which is an integral part of vitamin B_{12} and plays a vital role in the formation of many amino acids" (51). The maximum concentration of manganese content was observed, i.e. (130 µg/g) in *Stellaria media* and it is another essential microelement for human nutrition. The highest amount of sodium was reported in *Caralluma tuberculata* (205.53 µg/g). Sodium regulates blood pressure and movement of fluid in and out of body cells. Sodium is also involved in the regulation of acid-base balance, the transport of metabolites, nerve and muscle contraction" (52). The highest content of copper was reported in *Caralluma tuberculata* (35.06 µg/g). The primary function of Cu is that it is an important component of an enzyme, which helps in the incorporation of Fe in red blood cells and prevent anaemia (53). The total antioxidant capacity record highest in the *Chaerophyllum reflexum* followed by *Caralluma tuberculata*, while *Stellaria media* showed the lowest value of antioxidant among all the 5 wild vegetables. The highest DPPH values showed by *Allium griffithianum*, while the *Chaerophyllum reflexum* showed the lowest values. The highest antioxidant activity in the wild vegetables help in the reducing oxidative stress that's why they mostly used for the treatments of several human diseases such as diabetes mellitus, chronic inflammations, atherosclerosis (54-56). The standard deviation for the mineral composition shows that there is a wide scatteredness in the composition of iron, magnesium, sodium and phosphorous in selected wild vegetables. However, the level of cobalt and copper can be considered as equal in all the wild vegetables due to the lesser deviation in the composition. This is the ever first study recording the proximate analysis as well as minerals compositions of vs. *Allium graffithianum*, *Chaerophyllum reflexum* and *Buglossoides arvensis* in Pakistan and all over the world.

Conclusion

The results of the current study publicize that the chosen wild vegetables, which were traditionally used for various medicinal purposes and sources of food for centuries, are a good source of food (vital nutrients and minerals) regarding human health. In the conclusion of the current study, we can say that there was a strong connection between folk knowledge and phytochemicals of the selected wild species. The current study is a basis for evidence that folk knowledge makes a significant contribution to the balance of food consumption in the district of Kurram. To meet food needs, these wild vegetables are commonly used in daily eating and need to grow them widely. These wild vegetables can ensure a balanced diet by its various species and can prevent mineral deficiencies and malnutrition. From the result, it was concluded that most of the wild edible vegetables have considerable amounts of nutritional values and commonly used in the daily diet among the rural population. These 5 wild vegetables have massive amount of minerals, crude fat, crude protein and crude fiber which are also helpful for the local people to the treatment of many diseases.

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Authors contributions

SA, ZM carried out the field and laboratory work and wrote the initial draft. WH supervised and designed the research project. AA and JH helped in analysis of data. NA helped in experimental work. DH helped in structured and finalized the contents of the manuscript; improved the manuscript by editing and reviewing. All authors have read and agreed to the published version of the manuscript.

Compliance with ethical standards

Conflict of interest: The authors declare that they have no conflict of interest.

Ethical issues: None.

References

1. Jumrani J, Rai VN. The unacceptable status quo: Malnutrition challenges of the developed and developing world. In: Wheat and Barley Grain Biofortification. 2020; 1-25. http://doi:org/10.1016/B978-0-12-818444-8.00001-8
2. Sheikh SA, Mirani BN, Nizamani SM, Panhwar AA, Memon M. *et al.* Nutritional composition of wild-non-traditional vegetables of sindh, under different postharvest processing methods. Pak J Bot. 2019; 51(1):259-67. http://doi:10.30848/PJB2019-1(45)
3. Kalle R, Sõukand R, Pieroni A *et al*. Devil Is in the details: Use of wild food plants in historical Võromaa and Setomaa, present-day Estonia. Foods. 2020; 9: 570. http://doi:org/10.3390/foods9050570
4. Seal T. Determination of nutritive value, mineral contents and antioxidant activity of some wild edible plants from Meghalaya State, India. Asian J Appl Sci. 2011; 4: 238-46. http://doi:3923/ajaps.2011.238.246
5. Bussmann RW, Gilbreath GG, Solio J, Lutura M, Lutuluo R, Kunguru K, Mathenge, SG *et al*. Plant use of the Maasai of Sekenani Valley, Maasai Mara, Kenya. J Ethnobiol Ethnomed. 2006; 2: 22. https://doi.org/10.1186/1746-4269-2-22
6. Bussmann RW, Sharon D. Traditional medicinal plant use in Northern Peru: tracking two thousand years of healing culture. J Ethnobiol Ethnomed. 2006; 2: 47. https://doi.org/10.1186/1746-4269-2-47
7. Pieroni A, Houlihan L, Ansari N, Hussain B, Aslam S *et al*. Medicinal perceptions of vegetables traditionally consumed by South-Asian migrants living in Bradford, Northern England. J Ethnopharmacol. 2007; 113: 100-10. https://doi:org/10.1016/j.jep.2007.05.009
8. Mattelia G, Corvo P, Pieroni A *et al*. The virtues of being peripheral, recreational and transnational: local wild food and medicinal plant knowledge in selected remote municipalities of Calabria, Southern Italy. Ethnobot Res Appl. 2020; 19: 1-20. http://orcid.org/0000-0002-1947-7007
9. Füleky, György. "Cultivated plants, primarily as food sources." Encyclopedia of Life Support Systems 1 (2009); 1-47.
10. Brahma S, Narzary H, Basumatary S *et al*. Wild edible fruits of Kokrajhar district of Assam, North-East India. Asian J Plant Sci Res. 2013; 3: 95-100
11. Heywood VH. Use and potential of wild plants in farm households (No. 15). Food & Agriculture Org. 1999.
12. Termote C, Van Damme P, Djailo BDA *et al*. Eating from the wild: Turumbu, Mbole and Bali traditional knowledge on non-cultivated edible plants, District Tshopo, DRCongo. Genetic Resources CropEv. 2011; 58: 585-618. https://doi:org/10.1007/s10722-010-9602-4
13. Boedecker J, Termote C, Assogbadjo AE, Van Damme P, Lachat C et al. Dietary contribution of wild edible plants to 'women's diets in the buffer zone around the Lama forest, Benin–an underutilized potential. Food Secur. 2014; 6: 833-49. https://doi.org/10.1007/s12571-014-0396-7
14. Garekae H, Shackleton CM. Foraging Wild food in urban spaces: The contribution of wild foods to urban dietary diversity in South Africa. Sustainability. 2020; 12: 678. https://doi:org/10.3390/su12020678
15. Iqbal J, Shah A, Sarvat R, Adnan M, Parveen N, Nuzhat R et al. Documentation of folk herbal uses of medicinally important wild vegetables used by the tribal communities of Sargodha Region, Pakistan. Planta Daninha. 2019; 37. https://doi:org/10.1590/s0100-83582019370100135
16. Simpson BB, Ogorzaly MC. Spices, herbs and perfumes. Economic Botany: Plants in Our World, McGraw-Hill, New York. 1995; 278-301.
17. Satter MMA, Khan MMRL, Jabin SA, Abedin N, Islam MF, Shaha B et al. Nutritional quality and safety aspects of wild vegetables consume in Bangladesh. Asian Pac J Trop Biomed. 2016; 6: 125-31.https://doi:org/10.1016/j.apjtb.2015.11.004
18. Achaglinkame MA, Aderibigbe RO, Hensel O, Sturm B, Korese JK et al. Nutritional characteristics of four underutilized edible wild fruits of dietary interest in Ghana. Foods. 2019; 8: 104. https://doi:org/10.3390/foods8030104
19. World Health Organization. The World Health Report 2000: Health systems: Improving performance. World Health Organization. 2000.
20. Larsen T, Thilsted SH, Biswas SK, Tetens I, et al. The leafy vegetable amaranth (*Amaranthus gangeticus*) is a potent inhibitor of calcium availability and retention in rice-based diets. British Journal of Nutrition. 2003; 90: 521-27. https://doi:org/10.1079/BJN2003923
21. Lenzi A, Orlandini A, Bulgari R, Ferrante A, Bruschi P et al. Antioxidant and mineral composition of three wild leafy species: A comparison between microgreens and baby greens. Foods. 2019; 8: 487. https://doi:org/10.3390/foods8100487
22. Ullah I, Gul S, Rehman HU, Ahmad N, Ullah I, Aziz-ud-Din SMJ, Akbar MU et al. Analysis of nutrients and minerals of some wild edible plants. International Journal of Fauna and Biological Studies. 2017; 4: 35-39.
23. Arias-Rico J, Macías-León FJ, Alanís-García E, Cruz-Cansino NDS, Jaramillo-Morales OA, Barrera-Gálvez R, Ramírez-Moreno E et al. Study of edible plants: Effects of boiling on nutritional, antioxidant and physicochemical properties. Foods. 2020; 9: 599. https://doi:org/10.3390/foods9050599
24. Narzary H, Swargiary A, Basumatary S et al. Proximate and vitamin C analysis of wild edible plants consumed by Bodos of Assam, India. J Mol Pathophysiol. 2015; 4: 128-33. https://doi:10.5455/jmp.20151111030040
25. Aziz MA, Abbasi AM, Ullah Z, Pieroni A et al. Shared but threatened: The heritage of wild food plant gathering among different linguistic and religious Groups in the Ishkoman and Yasin Valleys, North Pakistan. Foods. 2020; 9: 601. https://doi:org/10.3390/foods9050601
26. AOAC. Official mehods of analysis of the association of official analytical chemists (17th ed.). Washington: Association of Official Analytical Chemists. 2000.
27. Hussain W, Badshah L, Ullah M, Ali M, Ali A, Hussain F et al. Quantitative study of medicinal plants used by the communities residing in Koh-e-Safaid Range, northern Pakistani-Afghan borders. J Ethnobiol Ethnomed. 2018; 14: 30. https://doi:org/10.1186/s13002-018-0229-4
28. Hussain W, Badshah L, Ali A *et al*. Quantitative aspects of the Koh -e-Safaid Range vegetation across the altiti tudinal gradient in Upper Kurram Valley, Pakistan. Appl Ecol Evciron Res. 2019;17:9905-24. https://doi:org/10.15666/aeer/1704 99059924
29. Abbas W, Hussain W, Badshah L, Hussain K, Pieroni A et al. Traditional wild vegetables gathered by four religious groups in Kurram District, Khyber Pakhtunkhwa, North-West Pakistan. Genetic Resources Crop Ev. 2020; 4: 1-16. https://doi:org/10.1016/B978-0-12-818444-8.00001-8
30. Hussain W, Badshah L, Hussain F, Ali A et al. Floristic configuration and ecological characteristics of plants of Koh-e-Safaid range, northern Pakistani-Afghan borders. Acta Ecologica Sinica. 2020; 40: 221-36. https://doi:org/10.1016/j.chnaes.2020.04.006
31. Ali M, Yar P, Salman K, Hussain W, Hussain K, Hussain G, Bussmann RW. Land use and land cover modification and its impact on biodiversity and the ecosystem services in District Kurram, Pakistan. BLACPMA.2022; 21: 365-88. https://doi:org/10.37360/blacpma.22.21.3.22
32. Datta S, Sinha BK, Bhattacharjee S, Seal T *et al*. Nutritional composition, mineral content, antioxidant activity and quantitative

estimation of water soluble vitamins and phenolics by RP-HPLC in some lesser used wild edible plants. Heliyon. 2019; 5, e01431. https://doi:org/10.1016/j.heliyon.2019.e0143

33. Khan N, Sultana A, Tahir N, Jamila N *et al*. Nutritional composition, vitamins, minerals and toxic heavy metals analysis of *Trianthema portulacastrum* L., a wild edible plant from Peshawar, Khyber Pakhtunkhwa, Pakistan. Afr J Biotechnol. 2013; 12.https://doi:10.5897/AJB2013.12972

34. Ubwa ST, Tyohemba RL, Oshido BA, Amua QM et al. Proximate and mineral analysis of some wild leafy vegetables common in Benue State, middle Belt-Nigeria. Int J Sci. 2015; 4: 25-29. https://doi.org/10.18483/ijSci.709

35. Tunçtürk M, Özgökçe F. Chemical composition of some Apiaceae plants commonly used in herby cheese in Eastern Anatolia. Turk J Agric For. 2015; 39: 55-62.https://doi:10.3906/tar-1406-153

36. Hussain J, Rehman NR, Khan AL, Hussain H, Al-Harrasi A, Ali L, Shinwari ZK *et al*. Determination of macro and micronutrients and nutritional prospects of six vegetable species of Mardan, Pakistan. Pak J Bot. 2011; 43: 2829-33.

37. Hussain W, Badshah L, Ali A, Hussain F, Jinjie Z, Hussain H, Green IR. Studies on varying nutrient status of five plants species in pre-reproductive, reproductive and post-reproductive stages, Koh-E-Sufaid Range, Kurram Valley, Pakistan. Pak J Bot. 2021; 53: 1771-77. http://dx.doi.org/10.30848/PJB2021-5(2)

38. Ahmad B, Abbas SJ, Hussain F, Bashir S, Ahmad D *et al*. Study on *Caralluma tuberculata* nutritional composition and its importance as medicinal plant. Pak J Bot. 2014; 46: 1677-84.

39. Pieroni A, Cattero V. Wild vegetables do not lie: Comparative gastronomic ethnobotany and ethnolinguistics on the Greek traces of the Mediterranean diet of southeastern Italy. Acta Bot Brasilica. 2019; 33: 198-211. https://doi:org/10.1590/0102-33062018abb0323

40. Bhatia H, Sharma YP, Manhas RK, Kumar K *et al*. Traditionally used wild edible plants of district Udhampur, J&K, India. J Ethnobiol Ethnomed. 2018; 14: 7. https://doi.org/10.1186/s13002-018-0272-1

41. Ahmad K, Pieroni, A. Folk knowledge of wild food plants among the tribal communities of Thakht-e-Sulaiman Hills, North-West Pakistan. J Ethnobiol Ethnomed. 2016; 12: 17. https://doi.org/10.1186/s13002-016-0090-2

42. Hussain W, Hussain J, Hussain S, Shinwari ZK, Ali R, Basir A *et al*. Ethono medicinal study of Parachinar, Kurram Valley (FATA) KPK. Pak JPharm Sci. 2013; 3: 85. https://doi:10.7324/JAPS.2013.31115

43. Ahmad M, Masood S, Sultana S, Hadda TB, Bader A, Zafar M *et al*. Antioxidant and nutraceutical value of wild medicinal Rubus berries. Pak J Pharm Sci. 2015; 28: 241-47.

44. Afolayan AJ, Jimoh FO. Nutritional quality of some wild leafy vegetables in South Africa. Int J Food SciNutr. 2009; 60: 424-31. https://doi:org/10.1080/09637480701777928

45. Agea JG, Kimondo JM, Woiso DA, Okia CA, Obaa BB, Isubikalu P, Teklehaimanot Z *et al*. Proximate composition, Vitamin C and Beta-Carotene contents of fifteen selected leafy wild and semi-wild food plants (WSWFPs) from Bunyoro-Kitara Kingdom. Uganda. J Nat Prod Plant Resour. 2014; 4: 1-12.

46. Saikia P, Deka DC. Mineral content of some wild green leafy vegetables of North-East India. J Chem Pharm Res. 2013; 5(3): 117-21.

47. Nkafamiya II, Osemeahon SA, Modibbo UU, Aminu A *et al*. Nutritional status of non-conventional leafy vegetables, *Ficus asperifolia* and *Ficus sycomorus*. Afr J Food Sci. 2010; 4: 104-08. https://doi.org/10.5897/AJFS.9000206

48. Akgünlü SB. Mineral content and microbiological analysis of some wild edible vegetables consumed in Kilis and Gaziantep provinces. MSc, Kilis. 2012; 7.

49. Saupi N, Zakaria MH, Bujang JS *et al*. Analytic chemical composition and mineral content of yellow velvet leaf (*Limnocharis flava* L. Buchenau) edible parts. J Appl Sci. 2009; 9: 2969-74. https://doi.org/10.3923/jas.2009.2969.2974

50. Adeyeyei EI, Otokiti MKO. Proximate composition and some nutritionally valuable minerals of two varieties of *Capsicum annum* L. (Bell and Cherry Peppers). Discovery and Innovation. 1999; 11: 75-82. https://doi:10.4314/dai.v11i1.15537

51. Czarnek K, Terpiłowska S, Siwicki AK *et al*. Selected aspects of the action of cobalt ions in the human body. Cent Eur J Immunol. 2015; 40: 236. https://doi:10.5114/ceji.2015.52837

52. Akpanyung EO. Proximate and mineral element composition of bouillon cubes produced in Nigeria. Pak J Nutr. 2005; 4: 327-29. https://doi.org/10.3923/pjn.2005.327.329

53. Ihedioha JN, Okoye COB. Nutritional Evaluation of *Mucuna flagellipes* Leaves: An Underutilized Legume in Eastern Nigeria. American Journal of Plant Nutrition and Fertilization Technology. 2011; 201: 1. https://doi:103923/ajpnft.2011.55.63

54. Jafri L, Saleem S, Ullah N, Mirza B *et al*. *In vitro* assessment of antioxidant potential and determination of polyphenolic compounds of *Hedera nepalensis* K. Koch. Arabian Journal of Chemistry. 2017; 10: S3699-S3706. https://doi.org/10.1016/j.arabjc.2014.05.002

55. Ahmed M, Fatima H, Qasim M, Gul B *et al*. Polarity directed optimization of phytochemical and *in vitro* biological potential of an indigenous folklore: *Quercus dilatata* Lindl. ex Royle. BMC Complementary and Alternative Medicine. 2017; 17(1): 1-16. https://doi.org/10.1186/s12906-017-1894-x

56. Karadag A, Ozcelik B, Saner, S. Review of methods to determine antioxidant capacities. Food Analytical Methods. 2009; 2: 41-60. https://doi.org/10.1007/s12161-008-9067-7

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