CHAPTER III

Determination of Metal Contents of Wild Edible Plants

The mineral elements of any foodstuff are very important and play a vital role in human health of acid-base balance, osmotic regulation of fluid and oxygen transport in the body, catalytic processes within enzymatic activities, endocrine and immune systems, bone formation and growth [1, 2]. It has been reported that about 33% population of the world is affected by anaemia which is due to iron deficiency [3]. Wild edible vegetables are good sources of macro- and micro-elements. Therefore, the daily eating of fresh fruits and vegetables, more than 400 g/day is suggested to prevent many major non-communicable diseases such as cardiovascular diseases and certain cancers [4]. However, due to rapid industrialization, many wild edible vegetables get contaminated with heavy metals like cadmium, chromium and lead, which can have profoundly deleterious effects on human health, including kidney problems, neurobehavioral and developmental disorders, high blood pressure and potentially even lung cancer [5]. The excess amount of micronutrient consumption is also harmful, e.g. excess zinc uptake can decrease the copper and iron absorption. Similarly, excess copper causes toxicity in the human body as it generates reactive oxygen species like superoxide, hydrogen peroxide (H_2O_2), or the hydroxyl radical ($OH^{\cdot}$) that might damage proteins, lipids and DNA [6]. Hence, investigation of such minerals in any food item is very essential before it is being recommended for human consumption.

In this chapter, the selected 17 wild edible plants consumed by the Bodos of Assam, North East India were investigated for the macro- and micro-elements and reported herein.

III.1 Materials and Methods

III.1.1 Sample preparation

The powdered samples prepared as per the procedure mentioned in **Section II.2.3** (**Page No. 73**) were digested by following the AOAC [7] method. Briefly 0.5 g of dry sample was taken in silica crucible and it was incinerated in muffle furnace at 500^{o}C for 2 h. After that, it was cooled in a desiccator and then digested repeatedly with concentrated HNO_3 by evaporating in a hot plate till it became colourless. After that, it was dissolved in 50 mL double distilled water in a volumetric flask, filtered with Whatman No. 1 filter paper and it was submitted for metal analysis.

III.1.2 Analyses of metals

Metals such as magnesium, calcium, copper, iron, zinc, manganese and nickel were analysed by using Graphite Furnace-Atomic Absorption Spectrometer (Analytik Jena Vario-6) at SAIF, North Eastern Hill University (Meghalaya) and other metals such as potassium, sodium, chromium, selenium, cobalt, cadmium, and arsenic were analysed by using Atomic Absorption Spectrometer (Thermo Scientific, AAS-ICE 3500, UK) at SAIC, Tezpur University, Assam. The results were expressed in mg/100 g dry weight (DW) of sample.

III.1.3 Statistical analysis

All the experiments were carried out for three independent replicates and the data were represented in terms of mean ± standard deviation. OriginPro 8.5 software (MA 01060, OriginLab Corporation, USA) was used for statistical analysis and executed by the one-way ANOVA *t*-test at $p < 0.05$.

III.2 Results and Discussion

The results of macro-elements analysis of the seventeen wild edible plants in mg/100 g of dry weight are presented in **Table III.1**. The levels of sodium in the plant species varied from 18.88 ± 0.01 mg/100 g DW in *C. sinensis* (lowest) to 290.54 ± 0.03 mg/100 g DW in *E. fluctuans* (highest). High levels of sodium were also found in *E. foetidum* (139.44 ± 0.02 mg), *C. hirsuta* (100.48 ± 0.20 mg), *N. herpeticum* (92.15 ± 0.02 mg), *S. peguensis* (136.71 ± 0.20 mg) and in *S. media* (72.91 ± 0.03 mg) per 100 g DW of samples. The results of current study were found similar to that of some leafy vegetables like *Delonix elata, Polygala erioptera,* and *Digera arvensis* reported by Gupta *et al*. [8]. On the other hand, the results were significantly higher in comparison to the Mediterranean herbs [9], wild edible fruits, roots and some spices reported by Agrahar-Murugkar *et al*. [10] and some wild vegetables such as *Lasia spinosa*, *Oxalis corniculata* etc. of North East India [11]. It has been reported that the consumption of high amounts of sodium contributes to hypertension in susceptible individual and leads to increased calcium loss in urine [12].

Table III.1: Macro-elements of wild edible plants (mg/100 g DW)

Plants	**Na**	**K**	**Ca**	**Mg**
Sz	48.73±0.13^{a}	7684.29±0.15^{a}	6.22±0.03^{a}	4.90±0.01^{a}
Ch	100.48±0.20^{b}	10462.28±0.13^{b}	6.20±0.14^{a}	5.60±0.02^{b}
Nh	92.15±0.02^{c}	8436.10±0.09^{c}	5.47±0.02^{b}	4.58±0.03^{a,c,}
Bl	33.91±0.02^{d}	7468.14±0.10^{d}	7.41±0.01^{c}	8.10±0.06^{d}
Sp	136.71±0.20^{e}	10164.25±0.10^{e}	5.00±0.01^{b}	4.10±0.02^{c}
Ta	34.00±0.01^{d}	7698.19±0.12^{f}	4.51±0.02^{d}	8.06±0.05^{d}
Oj	45.54±0.01^{e}	8528.41±0.10^{g}	4.20±0.05^{d}	5.60±0.07^{e}
Mp	55.35±0.30^{f}	5886.17±0.16^{h}	6.14±0.04^{a}	5.10±0.01^{e}
Dc	59.45±0.02^{g}	11784.13±0.11^{i}	5.52±0.02^{b}	8.02±0.02^{d}
Cs	18.88±0.01^{h}	7262.16±0.04^{j}	5.73±0.04^{b}	4.40±0.03^{a,c}
Sm	72.91±0.03^{i}	6376.16±0.15^{k}	4.20±0.05^{d}	7.40±0.04^{f}
Pc	53.35±0.06^{j}	1155.28±0.21^{l}	5.11±0.02^{b}	4.20±0.06^{c}
Aa	23.48±0.02^{k}	6786.16±0.10^{m}	6.70±0.02^{a}	9.00±0.01^{g}
Efo	139.44±0.02^{l}	6822.11±0.10^{n}	4.70±0.05^{d}	6.80±0.06^{h}
Lj	52.99±0.02^{m}	8828.24±0.10^{p}	5.60±0.06^{b}	7.00±0.02^{f}
Pp	37.02±0.02^{n}	9566.23±0.13^{q}	4.92±0.02^{e}	7.10±0.02^{f}
Ef	290.54±0.03^{p}	9166.26±0.10^{r}	4.00±0.06^{d}	6.50±0.03^{h}

Sz = S. zeylanica, Ch = C. hirsuta, Nh =N. herpeticum, Bl = B. lanceolaria, Sp = S. peguensis, Ta = T. angustifolium, Oj = O. javanica, Mp = M. perpusilla, Dc = D. cordata, Cs = C. sinensis, Sm = S. media, Pc = P. chinensis, Aa =A. acidum, Efo = E. foetidum, Lj = L. javanica, Pp = P. perfoliatum and Ef = E. fluctuans, DW = Dry weight. Values were expressed as mean of three replicates ± standard deviation and the data with different letters in a column are significantly different from each other at $p < 0.05$.

Table III.2: Micro-elements present in the wild edible plants in mg/100 g DW

Plants	Fe	Cu	Zn	Mn	Co	Cr	Se	Ni	Cd and As
Sz	1.83±0.05[a]	2.58±0.07[a]	0.70±0.05[a]	0.54±0.04[a]	0.39±0.11[a]	0.58±0.03[a]	0.81±0.40[a]	2.60±0.02[a]	Nd
Ch	6.10±0.01[b]	1.62±0.03[b]	0.30±0.01[a]	0.64±0.03[a]	0.57±0.02[a]	0.87±0.02[a]	1.83±0.60[b]	3.60±0.03[b]	Nd
Nh	2.48±0.01[c]	2.10±0.01[a]	0.70±0.02[a]	0.70±0.01a	0.59±0.12[a]	0.74±0.09[a]	0.78±0.41[a]	4.70±0.06[c]	Nd
Bl	13.90±0.02[d]	1.62±0.02[b]	1.11±0.03[b]	4.80±0.02[b]	0.43±0.04[a]	1.68±0.09[b]	1.01±0.06[a]	0.17±0.09[d]	Nd
Sp	2.40±0.06[c]	2.11±0.02[a]	0.20±0.01[a]	0.62±0.03[a]	0.57±0.16[a]	2.36±0.01[c]	0.45± 0.90[a]	4.20±0.02[c]	Nd
Ta	0.90±0.005[e]	1.81±0.01[b]	1.70±0.02[c]	3.40±0.02[c]	0.54±0.01[a]	1.65±0.04[b]	0.18±0.06[c]	0.25±0.07[d]	Nd
Oj	12.60±0.06[f]	2.14±0.05[a]	1.20±0.03[b]	5.00±0.06[d]	0.51±0.01[a]	7.53±0.13[d]	0.84±0.03[a]	0.25±0.03[d]	Nd
Mp	1.70±0.07[a]	2.60±0.01[a]	0.20±0.03[a]	0.80±0.14[a]	0.26±0.09[a]	1.32±0.12[b]	0.37±0.53[a]	3.80±0.02[b]	Nd
Dc	10.80±0.01[g]	1.90±0.07[b]	1.40±0.03[b]	5.20±0.04[d]	0.71±0.01[a]	1.61±0.15[b]	1.79±0.02[b]	0.43±0.02[d]	Nd
Cs	0.60±0.03[e]	3.21±0.01[c]	1.00±0.01[b]	5.60±0.02[d]	0.61±0.09[a]	1.74±0.05[b]	0.51±0.01[a]	0.23±0.01[d]	Nd
Sm	13.42±0.02[h]	1.50±0.02[b]	0.90±0.02[a]	4.50±0.06[b]	0.64±0.01[a]	2.61±0.11[c]	0.74±0.02[a]	0.35±0.02[d]	Nd
Pc	1.60±0.16[a]	2.60±0.41[a]	0.30±0.05[a]	0.60±0.02[a]	0.36±0.07[a]	2.38±0.19[c]	0.84± 0.24[a]	4.70±0.08[c]	Nd
Aa	0.90±0.061[e]	1.70±0.05[b]	1.20±0.02[b]	3.90±0.03[c]	0.45±0.01[a]	2.34±0.13[c]	0.72±0.07[a]	0.13±0.04[d]	Nd
Efo	12.50±0.01[f]	2.60±0.07[a]	1.00±0.06[b]	5.13±0.03[d]	0.56±0.01[a]	1.19±0.09[b]	1.15±0.04[d]	0.28±0.09[d]	Nd
Lj	10.70±0.01[g]	2.40±0.03[a]	0.40±0.01[a]	5.61±0.01[d]	0.31±0.09[a]	2.37±0.08[c]	0.98±0.01[a]	0.15±0.04[d]	Nd
Pp	12.50±0.01[f]	1.40±0.06[b]	1.60±0.04[c]	4.80±0.01[b]	0.46±0.07[a]	2.27±0.09[c]	0.94±0.05[a]	0.21±0.01[d]	Nd
Ef	15.10±0.02[i]	1.70±0.04[b]	1.70±0.04[c]	3.30±0.06[c]	0.67±0.01[a]	2.46±0.09[c]	0.24±0.01[c]	0.16±0.01[d]	Nd

DW = Dry weight, Values were expressed as mean of three replicates ± standard deviation and the data with different letters in a column are significantly different from each other at $p < 0.05$.

The result (**Table III.1**) shows that among the macro-elements, potassium was the most abundant element found in these plant species which ranged from 1155.28 ± 0.21 mg/100 g DW in *P. chinensis* (lowest) to 11784.13 ± 0.11 mg/100 g DW in *D. cordata* (highest). The results of current study are similar to the results of some wild vegetables like *Amaranthus spinosus, Centella asiatica,* and *Chenopodium album* reported by Borah *et al*. [13], wild edible flowers and fruits [9] and condiments like *Thymus vulgaris, Lavandula officinalis,* and *Anethum graveolens* [14]. The adult human being required 4700 mg/day of potassium [15], and thus the selected plants can provide adequate amount of potassium. It was reported that high potassium content helps in lowering the effects of NaCl, and thus reduces blood pressure [16]. It is required for nerve impulses and needed to release energy from carbohydrates, protein and fat and decreases the risk of cardiovascular disease [17]. The amount of calcium contents varied from 4.00 ± 0.06 mg/100 g DW in *E. fluctuans* to 7.41 ± 0.01 mg/100 g DW in *B. lanceolaria* which are lower in comparison to the results of some Mediterranean edible herbs like *Cichorium intybus* and *Sonchus asper* reported by Volpe *et al*. [9], edible herbs, leafy vegetables, wild flowers and fruits such as *Solanum gilo, Prunus nepalensis, Castanopsis indica* etc. reported by Ozcan [18] and Agrahar-Murugkar *et al*. [10]. However, the result of the present study showed the lower amount of calcium in comparison to the daily requirement for adult human being (1000 mg/day) [15]. Calcium deficiency increases the risk of osteoporosis [19]. The deficiency of magnesium affects several medical disorders such as asthma, high blood pressure, epilepsy, and all types of musculoskeletal disorders [5]. In this study, the amounts of magnesium varied from 4.10 ± 0.03 mg in *S. peguensis* to 9.00 ± 0.01 mg in *A. acidum* and these values are also lower in comparison to the reported results of leafy vegetables, edible herbs, wild flowers and fruits [18, 10, 8, 13, 9]. The levels of magnesium detected in *B. lanceolaria, T. angustifolium, S. media, L. javanica,* and in *P. perfoliatum* were 8.10 ± 0.06 mg, 8.06 ± 0.05 mg, 7.40 ± 0.04 mg, 7.00 ± 0.02 mg and 7.10 ± 0.02 mg per 100 g DW, respectively. However, these values were also very low in comparison to the daily requirement of this element (420 mg/day) [15]. The previous study reported [13] the elements like calcium, magnesium, iron, and potassium contents in *E. foetidum* were higher and the values were 512 mg/g, 308 mg/g, 24.26 mg/g, 11.26 mg/g and 26 mg/g, respectively. However, in *S. media* the sodium and potassium contents reported by Guerreo *et al.* [20] were lower (67.30 ± 8.10 mg/100 g FW and 460.10 ± 35.11 mg/100 g FW) in comparison to the present study.

The results of micro-elements analyses of wild edible plants in mg/100 g of dry weight are shown in **Table III.2.** The iron content varied from 0.60 ± 0.03 mg/100 g DW in *C. sinensis* (lowest) to 15.10 ± 0.02 mg/100 g DW in *E. fluctuans* (highest). High levels of iron were also observed in *C. hirsuta*, *B. lanceolaria, O. javanica, D. cordata, S. media, E. foetidum, L. javanica,* and *P. perfoliatum* (**Table III.2**). The recommended daily allowance (RDA) of iron is 8 – 18 mg/day [20]. These wild plants can be accepted as an excellent source of iron and the daily requirement of iron can be fulfilled by consumption of these wild vegetables which can prevent anaemia. Similar levels of iron were reported in some leafy vegetables by Gupta *et al.* [8], plants of Turkey reported by Ozcan [18], edible herbs [9] and in wild vegetables of Assam reported by Saikia *et al*. [11]. In comparison to this study, higher levels of iron were reported in cabbage, *Portulaca oleracea, Solanum nigrum,* and *Amaranthus spinosus* and in condiments like *Thymus vulgaris* [18, 22, 23]. The copper contents in the plants were found in the range from 1.40 ± 0.06 mg (*P. perfoliatum*) to 3.21 ± 0.01 mg (*C. sinensis*). The concentrations of copper were found almost similar in all the plant samples (**Table III.2**). Similar results of copper were also reported in some Mediterranean herbs and some condiments of Turkey [9, 18]. Copper acts as cofactors for many enzymes, which helps in biological electron transport system and also helps in the production of many enzymes in the body. Its deficiency can cause irregular glucose and cholesterol metabolism, less energy production, and thus increased oxidative damages [24, 25]. In this study, the copper content was found to be ranging from 1.40 ± 0.06 mg (*P. perfoliatum*) to 3.21 ± 0.01 mg (*C. sinensis*). The concentrations of copper content were found almost similar in all the samples with an average concentration of 2.09 mg/100 g DW. The results obtained from the present study were higher than that of the leafy vegetables like *Centella asiatica, Delonix elata,* and *Amaranthus tricolor* [8, 11], some tropical fruits such as papaya, guava, star fruits, banana, etc. and some non-conventional foods like, taro, wild taro, and yam [25]. Zinc is an important element which increases the immunity, but on excess of this mineral, it becomes toxic to the health [26]. The results of the zinc content of the plant species varied from 0.20 ± 0.01 mg/100 g DW (*S. peguensis* and *M. perpusilla*) to 1.70 ± 0.04 mg/100 g DW (*E.

fluctuans and *T. angustifolium*). Similarly, Ozcan [18] and Uusiku *et al*. [27] reported comparable levels of zinc content in some leafy vegetables like *Brassica sp., Bidens pilosa,* and *Amaranthus sp.*, and in some condiments like coriander, cumin, fennel, mint, etc. The results of current study are almost similar to the zinc contents of rice, barley flour, wheat flour, and millet reported by Reinivuo *et al.* [28]. Similarly, Kawashima *et al*. [29] and Gupta *et al*. [8] also studied some vegetables and reported low levels of zinc contents in comparison to the current study. The RDA of zinc is 8–11 mg/day given by the Food and Nutrition Board (FNB) [15]. Manganese plays an important role in steroid sexual hormone production and cellular metabolism [30]. The levels of manganese in the plant species of this study varied from 0.54 ± 0.04 mg/100 g DW (*S. zeylanica*) to 5.61 ± 0.01 mg/100 g DW (*L. javanica*) which is close to the results reported by Volpe *et al*. [9] and Ozcan [18]. Similarly, the results of current study are comparable with the reported results of millet, barley flour, rice and wheat flour [28] and some leafy vegetables of Assam [11]. The cobalt is an important element which helps in the formation of vitamin B_{12} (cyanocobalamin) and also has healing properties in accurate doses [24, 26]. The level of cobalt in this study was found the lowest in *M. perpusilla* (0.26 ± 0.09 mg) and it was found the highest in *D. cordata* (0.71 ± 0.01 mg) and the results are comparable to the results of some non-conventional foods, tropical fruits and wild plants [29, 31, 22]. The chromium element is required for activation of many enzymes and hormones [11]. The highest amount of chromium was observed in *O. javanica* 7.53 ± 0.13 mg/100 g DW and the lowest was found in *S. zeylanica* as 0.58 ± 0.03 mg/100 g DW. Similar findings of chromium contents were also reported in some condiment like coriander, mint, cumin and fennel [18]. The levels of selenium were found to be varying from 0.18 ± 0.06 mg/100 g (*T. angustifolium*) to 1.83 ± 0.60 mg/100 g (*C. hirsuta*). High concentrations of selenium were also observed in *B. lanceolaria, S. media, E. foetidum, L. javanica* and in *P. perfoliatum*. Similarly, Glew *et al*. [21] reported close values of selenium contents in some condiments. However, in comparison to this study, low levels of selenium were reported in some Korean foods, edible plants, and wild fruits [32, 22, 26,]. The RDA for selenium is 0.05 mg/day given by the FNB [15]. Selenium can act as antioxidant and its deficiency can cause weak immune system, heart diseases, and hypothyroidism [33, 34]. The nickel contents detected in the plants species varied from 0.13 ± 0.04 mg/100 g in *A. acidum* (lowest) to 4.70 ± 0.08 mg/100 g *P. chinensis* and *N. herpeticum* (highest). These results are closely similar to the reported results of some non-conventional foods [25], wild vegetables [11], and edible plants [22], and are quite higher to that of some conventional vegetables like cauliflower, radish, onion and carrot [28]. The heavy metals such as mercury, lead, cadmium and arsenic are toxic metals and non-biodegradable which get contaminated to the soil due to different environmental pollutions created by industry and agriculture sectors and these are ultimately get absorbed by the plants [35, 36]. They get accumulated in the body when consumed through foods and cause serious adverse health effects on human body [37, 38]. In this study, cadmium and arsenic were found to be below the detection levels in the plant species (**Table III.2**). The study shows that the wild edible plants selected are found to be free from cadmium and arsenic contamination and found to be rich sources of minerals which can fulfil malnutrition problems on regular consumption and thereby can prevent several human diseases.

III.3 Conclusion

The present study reveals that the wild edible vegetables consumed by the Bodos of Assam contain adequate levels of minerals. Potassium was the most abundant element found in these plants which ranged from 1155.28 ± 0.21 mg/100 g DW in *P. chinensis* (lowest) to 11784.13 ± 0.11 mg/100 g DW in *D. cordata* (highest). Among the selected plants, *E. fluctuans, B. lanceolaria,* and *S. media* were found to contain good sources of iron which were 15.10 ± 0.02, 13.90 ± 0.02 and 13.42 ± 0.02 mg/100 g of DW, respectively. The plant species are also good sources of selenium which is an important element for human health. The wild edible plants selected are found to be free from cadmium and arsenic contamination. Therefore, these vegetables can be served as rich sources of minerals which can fulfil malnutrition problems on regular consumption and thereby can prevent several human diseases.

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