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TRACE ELEMENTS IN *NUNGSHAM*, THE RED EDIBLE ALGAE OF MANIPUR

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ABTRACT: Energy Dispersive X-ray Fluorescence (EDXRF) technique was used for the determination of trace elements in wild red edible algae. The elements so far detected were discussed for their importance in the food item in addition to the monitoring the pollution levels of the fresh water where they grew as algae is one of the best bioaccumulator of heavy elements.

Key words: EDXRF, *Lemanea australis*, Trace elements, bioaccumumulator

INTRODUCTION

Plants have been used from the time of immortal at different countries as foods as well as medicine. A number of various plants are known as medicinal herbs and have been used for a very long time to cure illness. Although a curative agent is mainly with organic compounds such as glycosides or alkaloids, trace elements have additional impact. A deficit or excess in the supply of the trace element leads to a metabolic disorder. Hence the investigation of trace elements in the edible food items is very important most of the elements come from our food items. Most medicinal plants belong to a kind weed that can accumulate a greater amount of trace elements than other plants. So it can also be used to monitor environmental pollution. In polluted regions, some plants not hyperaccumulator may concentrate more trace elements indicating the type of pollutants whether toxic or not in the environment where they grow. Again by knowing the trace elemental levels in the edible plants, their role of health impacts can be analysed. A major goal for researches in biomedical science over the last few decades had been to determine the concentration of various elements in human tissues. In particular, much research has been carried out to elucidate the effects of the accumulation of certain elements by some organs which results in disease and disorders in these organs (Hofbauer et al., 1991; Scoot et al., 1982) Algal foods are rich in proteins, carbohydrates, vitamins, major and trace elements. Thus it can contribute to the nutritional requirement of humans and may be beneficial to which is responsible for human health. But if it contains toxic elements due to the environment where it grows, it may lead to hazardous effect to our body. The elemental analysis of plants with the help of EDXRF was reported by many workers (Tirasoglu et al., 2005; Ekinci et al., 2004; Budak et al.,2006).

There are several aquatic weeds which have been used as direct source of food or medicine to human beings. In addition to it some brown algae are also there Some of the edible algae are used as food items in China, Japan, Korea and they are exported to many European countries like Germany (Dawczynsky et al., 2007).

In Manipur, the north eastern state of India, Nungsham (*Lemanea australis* Atkins, Family-Rhodophyceae) the red edible algae which is grown in the fresh water river like Imphal river and Chakpi river is used as valuable food item. It grows abundantly in the river bed rocks and mostly grown on the stones at the depths of 3 to 4 feet which are situated at the big corners of the river courses. Hence the lateral meaning of Nungsham comes from "hair of stone". The size of the algae is 2.9 to 5.2 cm in length and 0.83 to 0.85 cm in breath.

MATERIALS AND METHODS

The samples are collected from the different locations of Serou area of the Chakpi river (remote area) and Koirengei area of Imphal river (just on the outskirts of Imphal city) of Manipur, India in January as this plant grows actively in winter season. A total of five samples were collected from Serou and three from Koirengei area. The samples were washed with distilled water and dried in the oven at 65^0C for 24 h. One should take care that the concentration of K can be changed if the drying temperature is higher than 70^0C and subsequently ground by an agate mortar. Grinding of the samples is necessary to have homogeneity in three dimensions. 150 mg of the sample powder were pelletised into a thin pellet of uniform thickness having 10 mm in diameter under a pressure of 100 -110 kg/cm^2 for five minutes. The pellets of the samples were analysed by the Jordan Valley Ex-3600 Energy dispersive X-ray fluorescence (EDXRF) spectrometer, which works with an oil-cooled system Rh anode X-ray tube having maximum voltage 50 kV and a current of 1 mA. The measurements were carried out in vacuum chamber with appropriate filters which were inserted in between the sample and source so as to find the optimum detection of elements in the sample. For lower Z elements ($Z< 19$), no filter was used and the anode voltage was kept at 8 kV and current at 85 mA. A 0.05 mm thick Ti filter was used for elements whose $Z \geq 19$ to $Z = 30$ with anode voltage 20 kV and current 400 mA. Again for higher Z elements, an Fe filter of 0.05 mm thickness was used at an anode voltage 35 kV and current 500 mA. The characteristic X-rays emitted from the sample were detected by the 12.5 mm^2 Si(Li) detector cooled at liquid nitrogen temperature (77 K). The spectra were analysed by the software called ExWIN integrated with the system (Raychaudhuri et al., 2008). The NIST apple leaf standard, (SRM 1515) was used as external standard reference material.

Statistical Analysis

The Standard Deviation (S.D.) of the data collected was computed by the following equation

$$S.D. = \sqrt{[(X_i - X_m)^2/(n-1)]}$$

where X_i is the ith observation, X_m is mean of the observations and n is the number of observations.

RESULTS AND DISSCUSSION

The elements detected in the algae sample were analysed from the spectrum and the concentration of them were determined. The elements so far detected along with their concentrations are shown in Table 1 and Table 2 for Serou area and Koirengei area respectively. The elemental concentration in the edible algae so far analysed can be divided into major and trace elements. Elements like K, Fe, Al, Si, P, S, Cl, can be grouped in major elements and the remaining in trace elements. In the elements also, Al, Si, P, S, Cl, Mg, Mn were detected in the samples collected from Koirengei area and V, Se were detected in the samples of Serou. The elements viz. K, Ca, Ti, Cr, Mn, Fe, Cu, Zn, As, Br, Rb, Sr were common to both the sample from Korengei and Serou area. The trace elements as well as major elements in the algae food products can be discussed from two angles viz. 1. as food nutrient 2. as biomonitor.

Table-1: Mean element concentration ± standard deviation (mg/kg) of *Lemania austrlis* of Serou area for three replicates in each sample.

Element	Samples				
	S1	S2	S3	S4	S5
K	45598.5±678.1	48116.7±393.8	45168.9±671.4	45973.3±1198.6	43128.4±1855.6
Ca	319.5±42.2	549.4±26.9	389.1±62.1	894.0±43.9	852.4±35.4
Ti	94.8±10.9	62.6±16.2	152.6±7.0	134.7±13.8	85.1±6.8
V	2.5±2.3	0.9±1.2	5.6±2.4	1.5±0.9	1.8 ±1.2
Cr	49.0±10.7	51.1±10.4	79.2±6.9	74.2±7.8	39.4±12.8
Mn	194.5±7.5	175.4±4.9	258.8±13.9	260.1±9.2	222.6±6.1
Fe	1669.6±58.5	1150.3±40.4	2512.0±152.3	2081.5±147.7	1407.8±70.6
Cu	5.7±0.8	6.5±1.0	7.2±0.6	6.1±0.4	7.1±1.0
Zn	28.8±1.1	30.2±1.9	42.0±2.4	32.3±1.2	28.8±1.4
As	71.9±4.5	79.0±5.9	74.6±4.2	75.1±4.7	82.0±5.8
Se	0.09±0.1	nd	nd	0.02±0.03	nd
Br	50.9±0.1	49.1±0.5	50.6±0.7	49.6±0.9	56.8±1.4
Rb	51.4±2.9	52.4±2.7	52.9±3.5	54.7±3.3	42.1±16.8
Sr	12.8±2.9	14.1±1.7	12.4±2.5	15.9±3.7	8.7±5.2

Table-2: Mean element concentration ± standard deviation (mg/kg) of *Lemania austratis* of Koirengei area for three replicates in each sample.

Element	Samples		
	K1	K2	K3
Mg	619.3±154.0	837.8±51.5	510.3±96.7
Al	14512.0±1363.8	1099.1±432.2	2939.4±257.2
Si	48773.1±4999.9	6325.3±763.0	10959.9±933.3
P	3067.7±130.6	4401.1±148.0	2827.1±55.2
S	28193.1±720.4	8837.7±321.5	6415.3±105.4
Cl	3565.7±82.5	15215.4±323.1	13169.4±177.5
K	42667.0±903.9	46763.0±1204.3	42879.9±461.4
Ca	4174.6±62.9	943.7±46.6	354.5±44.0
Ti	912.2±78.4	83.5±8.4	159.2±14.0
Cr	118.8±33.0	35.6±8.2	71.5±19.1
Mn	786.7±24.0	230.5±6.2	285.2±9.4
Fe	12481.5±436.5	1538.5±83.9	2599.4±98.4
Cu	3.3±0.7	7.3±0.7	5.9±0.5
Zn	32.8±2.0	29.5±1.6	34.2±0.5
As	23.9±3.4	90.9±2.8	57.0±8.4
Br	7.0±0.1	62.1±1.4	39.9±0.4
Rb	107.2±1.5	55.6±2.9	44.9±3.8
Sr	137.6±6.5	15.2±2.3	15.8±2.8

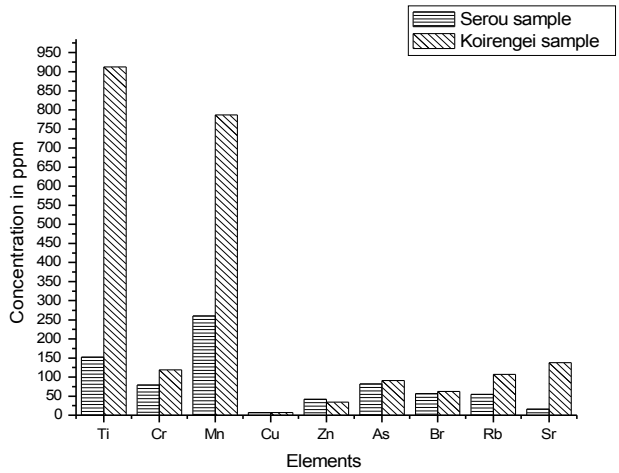

Fig.1 Concentration of Elements in *Lemanea australis*

As food nutrient

As evident from the tables, the main constituent heavy elements in all the samples were found to be K, Ca and Fe followed by the remaining elements in trace levels. The high concentration K in the algae leads to the conclusion of possibility of using it to reduce high blood pressure. Each element is use for the smooth functioning of our body except for Rb and Sr and Al whose functions are not clear.

Surprisingly, iodine was not detected in our experiment which was contradictory to the earlier findings of sea algae in which it was observed in large concentration (Dawczynsky et al., 2007). This may be due to the difference in oceanic origin and fresh water origin as elemental uptake by the plants depends on regional soil characteristics and climate condition (Zaidi, 2004). The variation in elemental concentration of the sample is also due to the different collecting site. The Se containing in the algae of Serou area is remarkable as this element has anti-oxidant property so that it can be used for cancer treatment. The daily intake requirement of it is 0.05-0.2 mg per day. The high amount of Fe in all the samples signifies that high concentration of it in the water where they grow and can be used them as food items for those persons having iron deficiency syndrome. The daily intake requirement of Fe is 10 mg per day for male and 15 mg per day. Chromium is essential functioning as a glucose tolerance factor. The function of Vanadium will be probably part of protection system against injury of tissues. Zinc deficiency is characterized by recurrent infections. For Nickel, Urease, the Nickel containing enzyme is used hydrolysis of urea. The functional values of Manganese are as a Lewis acid and catalyst for oxidation. The function of Titanium is not known yet. It is harmless to our body. Bromine may be essential to in mammals. It is non-toxic except in oxidizing forms. The total As in the algae samples ranges from 23.9 -90.9 mg per kg of dry weight which is well below the maximum value as reported from Spain (Almela, 2002). Both inorganic and organic concentrations of it are responsible for it. Here It is worth to mention that the organic compounds are considered to be nontoxic or minimally toxic in living systems, whereas in recent years, evidence has accumulated that dimethylarsinic acid (DMA), the major As metabolite formed after exposure to inorganic As via ingestion or inhalation in both humans and rodents, has cytotoxic and carcinogenic properties (Dawczynsky et al., 2007). The inorganic As is more severe than organic As. To evaluate the health risk of As, its chemical composition in the red algae is very important to know.

The detection of phosphorus in the samples of Koirengei area may be due to the seepage of long used phosphate fertilizer in paddy fields and it indicates pollution level of the water. Si in the form of compounds of $Si(OH)_4$ is an extremely abundant element in minerals and soils, its role in living system has been poorly examined since it is required only as trace elements in most higher animals, although there as much Si present in man as there is magnesium. By contrast, in plant life it is a major element. In the samples from Korengei, Al is also detected in huge amount in addition to Si. It may be due to the adsorption or due to the contamination polluted water. Si takes a role in bone formation, in connective tissue and it is found that Al does accumulate with silica in the needle leaves of fir trees, it is uncertain that there association in a particular compound (Frausto et al., 2006). Some of the daily intake requirements of elements are given as follows: P -700 mg, Cl – 750 mg, Ca – 1000 to 1200 mg, Cr – 0.05 to 0.2 mg, Cu – 1.5 to 3 mg (Raychaudhuri et al., 2008). The richness in Fe is significant for using it in Fe deficient persons. As Nungsham is used in small amount in one of the food items (Chatani) by Manipuris, it will not have hazardous effect to body due to excess of elemental concentration.

As biomonitor

As, algae being one of the best bioaccumulator of various elements, it can be used to study the environment where it grows. A large number of investigations into the effect of heavy metals on water environments and their accumulation in various hydrobionts can be found in literature data (Levkov et al. 2002; Kelly et al. 1989).

Algae and molluscs are among the organisms most used for this purpose (Conti et al., 2003). Macroalgae are able to accumulate trace metals, reaching concentration values that are thousands of times higher than the corresponding concentrations in sea water (Bryan et al., 1992; Fo¨ster, 1976; Rai et al., 1981). Algae bind only free metal ions, the concentrations of which depend on the nature of suspended particulate matter (Luoma, 1983; Seeliger et al., 1977; Volterra et al., 2000), which, in turn, is formed by both organic and inorganic complexes.

So all the dissolved salts and compounds forming the metal ions in the river water are responsible for the detection of the elements from which the idea of constituent elements in the rocks where the rivers flow can be known. The detection of Al, Si, P, S, Cl, Mg, Mn in the Koirengi samples suggests the availability of such elements originating either from rocks and artificial fertilizer or household and other industrial effluents as the site is at the outskirt of the Imphal city. The non-detection of Pb, Hg and Cd leads to the fact that the plants are grown in the pollution free of such elements. The maximum concentration levels of the common elements on the two places (Serou and Koirengei) are shown in Fig.1 and it was found that the highest concentration of almost the elements in Koirengei samples is higher from that of the Serou samples except some elements like K. From the comparison of the concentration of the elements in the two tables, it was found that the water in the Korengei area is polluted more as compare to Serou area.

CONCLUSION

The major and trace elements in the *Lemanea australis* were determined by using EDXRF technique. The number of elements detected in outskirts of the Imphal city was greater than that observed in remote area. From the observation, it is concluded that using of *Lemanea australis* as food item, from Serou area is better for health as compare to those from Koirengei area.

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