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Determination of Multi -elemental Profile of Nakima (*Tupistra nutans*)

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Abstract
Nakima (*Tupistra nutans*) is one of the indigenous underutilized vegetable crops of the State of Sikkim and the inflorescence part is used for consumption. The study has been conducted at Dept. of Horticulture, Sikkim University, Gangtok during 2014-15 with an objective to determine the multi-elemental profile of Nakima (*Tupistra nutans*). The Nakima inflorescences were collected from all the four districts of Sikkim i.e. East, West, North and South. There were altogether eight samples i.e. two samples from each of the four districts of Sikkim. Three replications were taken for each sample and the statistical design used was Completely Randomized Design (CRD). The determination of elements was done through ICPMS (Inductively Coupled Plasma Mass Spectrophotometry) after microwave digestion. Among Major elements P, K, Mg and Mn, Fe, Zn micro elements were found in higher concentrations. Nakima has been found to be good source of major and minor nutrients, so commercial production Nakima should be promoted in the state of Sikkim. Large scale commercial production would provide economic benefits to the farming community and will be helpful in reduction of cost of underutilizated vegetable.

Keywords: Nakima, multi-element, nutritional profile

Introduction
Nakima (*Tupistra Nutans*) is an genus of about 20 species of flowering plants. It is an unusual rhizome geophyte from the eastern Himalayas with long strap like leaves up to 1-2 metre in length forms a tall lush clump of evergreen foliage resembling a curious Aspidistra. It has an intresting club like flower that brings to mind somewhat the equally curious flower of *Molieria crassitolia* but is larger. Flowers are short duration to produce marketable price, inflorescence of attractive colour, shape, size and nice keeping quality. Its inflorescence is used in curry as well as medicinal purpose. These are eaten in Sikkim as spicy vegetables. It can be cultivated throughout the Sikkim especially in temperate regions. Nakima plant is propagated through sucker. It does not need so much care at the time of planting but field should be free from weed at the time of fruiting. Planting distance is kept 1-2m.

These species are mostly found in the moist and shady place and available in local market during the month of September-October. Many local people collect them from the forest and use them as vegetables. This is also available in the private forest in some villages.

The main importance of this work is to determine the multi-elemental value of this vegetable which is grown in Sikkim and is liked by local people also. By keeping in view, the above mentioned points the present sturdy was undertaken with the following objectives. To estimate the multi-element profile of Nakima inflorescence (*Tupistra nutans).*To analyze the various dietary nutrients present in the inflorescence of Nakima (*Tupistra nutans).*

Materials and methods
The present research work entitled "Determination of multi-elemental Profile of Nakima (*Tupistra nutans*)" was carried out during the year 2013-2015 in the P.G Laboratory, Department of Horticulture, Sikkim University, 6th mile Samdur, Tadong, Gangtok at the altitude of 1610 m and with latitude and longitude as $N^0 27^0 18.495'$ and $E^0 88^0 35.307'$. The details of materials used and methodology employed during the course of investigation are being described as follows:

Experimental material and sample collection
Nakima (*Tupistra nutans)* inflorescences were used as experimental material for the present study. Nakima (*Tupistra nutans*) inflorescences were used as experimental material in the present study and sample collection was done during October-November, 2014.

The Samples 1and 2 were collected from Naga village and Singik (North Sikkim) and Samples 3 and 4 were collected from Assamlingzey and Samsing (East Sikkim). Samples 5 and 6 were collected from Lower Perbing and Lower Chuba (South Sikkim) and Sample 7 and 8 was collected from Gairigaon and Martam (West Sikkim).

Table 1: Details of samples collection from various places in Sikkim

Name of Samples		Name of treatments	Replications	Date of collection
1	Sample 1	T_1-Naga village (North Sikkim)	T_1R_1, T_1R_2, T_1R_3	25/11/2014
		T_2- Singik-(North Sikkim)	T_2R_1, T_2R_2, T_2R_3	15/10/2014
2	Sample 2	T_3-Assamlingzey-(Sikkim East)	T_3R_1, T_3R_2, T_3R_3	13/10/2014
		T_4- Samsing-(East Sikkim)	T_4R_1, T_4R_2, T_4R_3	22/11/2014
3	Sample 3	T_5-Lower-Perbing (South Sikkim)	T_5R_1, T_5R_2, T_5R_3	6.10.2014
		T_6 Lower Chuba -(South Sikkim)	T_6R_1, T_6R_2, T_6R_3	8/10/2014
4	Sample 4	T_7-Gairigaon-(West Sikkim)	T_7T_1, T_7R_2, T_7R_3	5/10/2014
		T_8- Martam- (West Sikkim)	T_8R_1, T_8R_2, T_8R_3	5/10/2014

Fig 1: Nakima (*Tupistra nutans*) plants and its inflorescences

Instruments used

ICP-MS (Inductively Coupled Plasma Mass Spectrophotometry) Perkin Elmer, Nex ION 300 X. and micro-wave digestion system (Anton par microwave 3000) which were used to determine the different minerals present in nakima (*Tupistra nutans)* like N, P, K, Ca, Na, Mn, Mg, Fe, Cu, Mo, I, Cl and Zn and the instruments were available in Department of Horticulture, Sikkim University.

Experimental methods

The present research was conducted in PG Laboratory, Department of Horticulture, Sikkim University, 6th mile Tadong, Samdur, Gangtok. The dietary nutrients were analyzed by following methods as given below:-

Determination of multi elements

Multi elements was extracted throught ICP-MS (Inductively Coupled Plasma Mass Spectrophotometry) Perkin Elmer, Nex ION 300 X. and micro-wave digestion system (Anton par microwave 3000) which were used to determine the different minerals present in Nakima (*Tupistra nutans)* like P, K, Ca, Na, Mn, Mg, Fe, Cu, I, Cl, Zn Pb Si, Ni etc and the instruments were available in Department of Horticulture, Sikkim University. The micro wave digestion system (Anton par microwave 3000) was used for sample digestion as per the following steps. 0.5 gm sample were taken.9ml of 69% nitric acid and 2ml HCl were added into the digestion tube. The Ramp -5, Fan -1 power 1200 watt, Rate 0.3 bar per sec and Hold -5were maintained. The total time taken was 40 minutes for digestion. The digested samples were transferred into 100ml volumetric flask when the temperature of the sample was reduced and distilled water was added for making the volume of 10 ml. The liquid sample was transferred into narrow mouth bottle before the minerals were determined in ICP-MS The minerals present in the sample were analyzed by ICP-MS, Perklin Elmer Nex ION 300X. Digested sample were analyzed for the ionic constitution using elements standards such as Ag, Al, Ba, Bi, Ca, Cd, Co, Cr, Cs, Cu, Fe, Ga, Ln, K, Li, Mg, Mn, Na, Nb, N, Ti, Pb, Rb, Ge, Se, Si Sr, Tn, U, V, W, Zn and Zr were used as a standards.

Results and discussion

There were eight samples (treatments) used for conducting the experiment for analyzing the Multi-elemental profile of Nakima (*Tupistra nutans*) inflorescence. In Multi-elements mainly Major, Minor and Toxic elements were estimated.

Determination of Major elements

Determination of phosphorus (P) Nakima

From Table no.(2), the finding shows that the maximum amount of phosphorus (P) content was found in treatment (T2) (5.48 mg L^{-1}) which was collected from Singik North Sikkim whereas lowest phosphorus content was found in treatment T4 (3.37 mgL^{-1}) that was collected from Samsing East Sikkim. Similar finding were reported by Aberoumand (2014) [1].

Determination of) Potassium (K) content in Nakima

Potassium (K) content in different treatments (T1) to (T8). The highest potassium content was found in treatment (T1) (11.11 mgL^{-1}) which was collected from Naga village, North Sikkim. The lowest potassium content was found in treatment

T8 (0.08 mg L^{-1}) that was from Martam West Sikkim. Similar results were reported by Tapan. *et al.* (2014). The reason might be due to similar fertilizer / manure Application, similar planting time and proper care at the time of harvesting. Another reason might be due to genetic similarity among the samples.

Determination of Sodium (Na) content in Nakima

Maximum amount of sodium (Na) content was found in (T7) (0.17 mg L^{-1}) which was collected from Gairigaon, west Sikkim and lowest content of sodium was recorded in (T2) that was collected from Singik North Sikkim. The findings of present study are in line with Tapan *et al.* (2014). The reason might be slow mineralization rate in soil and different pH range values.

Determination of magnesium (Mg) content in Nakima

The highest Mg content was found in treatment (T6) (8.71 mgL^{-1}) collected from Lower Chuba, South Sikkim. The lowest Mg content was found in treatment (T3) (1.9 mg L^{-1}) collected from Assam lingzey, East Sikkim. Similar findings were reported by Tapan *et al.* (2014). This difference could be due to the external growing environment leading to change in the final quality of the inflorescence and its shelf-life after harvest.

Determination of Calcium (Ca) content in Nakima

The highest Ca content was found in treatment (T2) (3.81 mg L^{-1}) collected from Singik North Sikkim, whereas the lowest was found in treatment (T1) (1.91 mg L^{-1}) sample collected from Naga village, North Sikkim. These findings are in agreement with those of Tapan, *et al.* (2014). The calcium contents in present study were higher due to the green colour content in inflorescence of plant. Another difference might be due to variation in cultivation practices and manures and fertilizers application.

Table 2: Estimation of Major element Content P, K and Cu in Nakima *(Tupistra nutans)*

Treatment No.	Major elements content in Nakima (*Tupistra nutans*) on (dry wt. basis mg L^{-1})				
	P	K	Na	Mg	Ca
T1	4.67	11.11	0.06	2.03	1.98
T2	5.48	9.31	0.04	2.87	3.81
T3	3.11	7.87	0.06	1.91	2.35
T4	3.37	9.59	0.05	2.64	2.42
T5	4.02	9.34	0.06	2.33	2.25
T6	4.08	9.32	0.05	8.71	2.85
T7	3.55	9.07	0.17	2.54	2.04
T8	3.51	0.08	0.06	2.11	2.33
GM	4.029	9.210	0.687	2.388	2.337
CD at 5%	0.235	0.179	0.173	0.406	0.968
SEm	0.786	0.597	0.577	0.135	0.234

Determination of Minor elements

Determination of Manganese (Mn) content in Nakima

The Table (3) shows that Mn content was found maximum in (T4) and (T6) (0.04) collected from East and South Sikkim and lowest was recorded in (T1) and (T2) (0.01 mg L^{-1}) the sample was collected from North Sikkim. The similar results were also reported by Tapan *et al.* (2014). Manganese contents in present study were high and the reason might be the variation in climatic conditions.

Determination of Iron (Fe) content in Nakima

The highest Iron content was recorded in (T1) (0.11 mg L^{-1}) which was collected from Naga village, North Sikkim whereas lowest was found in (T4) (0.07 mg L^{-1}) followed by T6, T7 and T8 collected from Martam West Sikkim. The similar results were also reported by Tapan *et al.* (2014). This difference could be due to the location and season of inflorescence growth, as well as monthly and yearly variation in weather conditions. Abiotic conditions i.e. soil fertility and water availability vary from year to year and site to site and can affect the level and quality of inflorescence after harvest.

Determination of Cupper (Cu) content in Nakima

The maximum Cu was found in (T2) (0.02 mg L^{-1}) collected from Singik, North Sikkim and lowest content was found in (T1) (0.01 mg L^{-1}) the sample was collected from Naga village, North Sikkim. These findings are in agreement with those of Tapan *et al.* (2014). This difference might be due to semi-perishable nature of Nakima and they have a short life after harvest (3 to 6 days) due to their natural soft texture. Post-harvest handling and storage conditions such as packaging, relative humidity, temperature, light and storage period might also have affected the content of the minerals in Nakima.

Determination of Zinc (Zn) content in Nakima

The highest Zn content was recorded in (T7) (0.07 mg L^{-1}) was collected from Gairigaon, west Sikkim and lowest was recorded in (T3) (0.03 mg L^{-1}) collected from Assam lingzey, East Sikkim. Similar findings were reported by Tapan *et al.* (2014). This reason might be due to biotic factors such as available soil moisture, temperature, relative humidity and nutrient availability influencing texture of Nakima inflorescence.

Table 3: Estimation of Minor element Fe, Mg and Mn content in Nakima *(Tupistra nutans)*

Treatment No.	Minor elements content in Nakima (*Tupistra nutans*) on (dry wt. basis mg L^{-1})			
	Mn	Fe	Cu	Zn
T1	0.01	0.11	0.01	0.04
T2	0.01	0.08	0.02	0.04
T3	0.03	0.08	0.01	0.03
T4	0.04	0.07	0.01	0.03
T5	0.01	0.08	0.02	0.04

T6	0.04	0.07	0.01	0.03
T7	0.03	0.07	0.01	0.07
T8	0.03	0.07	0.01	0.03
Gm	0.279	0.787	0.112	0.376
CD at 5%	0.145	0.173	0.105	0.221
SE m	0.485	0.577	0.408	0.739

Determination of Toxic elements

Determination of Lead (Pb) content in Nakima

The Table (3) shows the toxic elements and all the treatments were found significantly at par at 5% level of significance. In toxic elements maximum Pb content was found in (T1) (0.09 mg L^{-1}) the sample collected from Naga village, North Sikkim and lowest was found in T2(0.01 mg L^{-1}) collected from Singik, North Sikkim. The findings of present study are in line with Chove *et al.* (2006) [5]. The reasons might be due to different cultivars, purchase of the samples from different locations, growing of crop in different seasons, stage of collection i.e. early stage of inflorescence and maturity stages.

Determination of Aluminum (Al) content in Nakima

Al was found lessie. (0.01 mg L^{-1}) found in all the treatments collected from all the four districts of Sikkim. In Ti, Si, Ni maximum content was found in (T2) (0.01 mg L^{-1}) followed by (T4) whereas lowest was found in (T1) (0.02) followed by (T3), (T5), (T6), (T7) and (T8). The findings of present investigation are in concurrence with Ademilua. and Obaloba. (2008) [2], Powel *et al*. (2005) [6] and Asaiou *et al.* (2012). In case of toxic elements, the suggested safe concentration in plant species is (2 to 6 mg L^{-1}) (Broyer *et al*., 1972), so the analyzed plant species has lower level of toxic elements which further clarifies their use as food supplement or their medicinal benefits.

The present study reveals the nutritional profile of Nakima (*Tupistra nutans*). The nutrients present in Nakima are comparable to the popular vegetables of Sikkim like Asparagus etc. Commercial utilization of Nakima in Sikkim will be beneficial for farmers of Sikkim as it may help in cost reduction of present value of this underutilized vegetable.

Table 4: Estimation of content of toxic element in Nakima *(Tupistra nutans)*

Treatment No.	**Toxic elements content in Nakima *(Tupistra nutans)* on (dry wt. basis mg L^{-1})**				
	Pb (%)	**Al (%)**	**Ti (%)**	**Si**	**Ni**
T1	0.09	0.01	0.02	0.02	0.02
T2	0.01	0.01	0.01	0.01	0.01
T3	0.02	0.01	0.02	0.02	0.02
T4	0.02	0.01	0.01	0.01	0.01
T5	0.04	0.01	0.02	0.02	0.02
T6	0.08	0.01	0.02	0.02	0.02
T7	0.02	0.01	0.02	0.02	0.02
T8	0.02	0.01	0.02	0.02	0.02
GM	0.466	0.304	0.200	0.200	0.200
CD at 5%	0.141	0.109	0.176	0.176	0.0.176
SEm	0.471	0.361	0.589	0.589	0.2589

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