

Analysis of macro metal content in chillie powder available in the Sri Lankan market

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Abstract—Chillie (*Capsicum annum* L.) powder is the dried, pulverized fruit of chillies and used as a spice to add pungency flavour to the dish. This study was conducted to investigate the macro metal content in chillie powder samples commonly available in the Sri Lankan market. Five brands of chillie powder samples named A, B, C, D and E, and an unbranded sample were selected for this study. Three batches from each brand and three samples from each batch were purchased. An ungrounded whole dry chillie sample was selected as the control. The macro metal content was determined using Atomic absorption spectrometric and X-ray fluorescence spectrometric techniques. Results were statistically analyzed using one-way ANOVA, at 0.05 probability level with MINITAB-14 software package. According to this study, there was no significant difference between the batches in each brand in mean metal contents. But there were significant differences among the brands in mean metal contents. According to the results Na content of all the chillie powder samples and Ca content of some brands (brand C-955.2 ± 1.2 µg/g, brand D-962.0 ± 0.9 µg/g and brand E-822.0 ± 1.0 µg/g) were found to be lower than the control (1495.2 ± 0.3 µg/g). The reason for this may be due to the loss caused by heat generation during the chillie grinding process or may be due to adulteration with plant materials with low Na and Ca content. The mean K content of chillie powder samples was within the range of 22 866.6 to 25 547.0 µg/g, while the mean Mg content varied in the range of 1991.3 – 2180.5 µg/g. These variations may be due to multiple factors, such as the differences in soil conditions where it was grown, variety of chillie, maturity of chillie, growing season, climatic condition, processing treatments and preservation method.

Index Terms— Calcium, Chillie powder, Magnesium, Potassium, Sodium

I. INTRODUCTION

Chillie crop is one of the most important commercial spice crops. Botanical name of chillie is *Capsicum annum* L. There are more than 400 different varieties of chillies found all over the world. Chillie belongs to the genus capsicum, under the solanaceae family. Chillies occur referred to as chillies, hot peppers, bell peppers, red peppers, pod peppers, cayenne peppers, paprika, pimento, and capsicum in different parts of the world [14].

Currently, chillies are used throughout the world as a spice, as a vegetable, and also in the making of beverages and medicines. Some varieties of chillies are famous for red colour because of the pigment 'capsanthin,' others are known for biting pungency attributed to 'capsaicin'. Chillie is very important as an anti-oxidant and anti-inflammatory agent. Chillie has become an essential ingredient in Sri Lankan meals. Per capita consumption of chillie in the form of dry chillie is estimated 2.32 kg per annum and the national annual requirement of dry chillie is around 42,634 Mt. The annual production of dry chillie in Sri Lanka is about 18,616 Mt, therefore, an amount of 31,242 Mt is being imported (Year 2007 figures). Chillie is mainly imported from India, as dry chillie [6].

In the chillie powder manufacturing process in Sri Lanka dried chillies are cleaned manually to remove impurities and then washed. After drying them, they are pulverized in two or more grinders to convert them into the powder form. Then chillies in powder form are passed through sieves to obtain uniform mesh size (45-65 mesh). Some manufactures use

metal detectors to detect and remove metal particles like iron from the ground chillie. Finally, packing is done in polythene bags using automated powder fillers, and bags are sealed. Due to major part of chillie being imported from India, many branded chillie powders available in the Sri Lankan market are products of imported dry chillies from India.

Minerals are the building blocks of the human body. They are required for body structure, fluid balance, protein structures and to produce hormones. A mineral is a compound, how the metal is found in nature. The macro metals are those found in large quantity in living bodies. They include calcium, magnesium, sodium, potassium, phosphorus and sulfur. Sodium (Na) is present mostly as an extracellular constituent and maintains the osmotic pressure of the extracellular fluid. Potassium (K) is necessary for adequate muscle contraction and rapid transmission of nerve impulses in the body. Magnesium (Mg) is used in building bones, manufacturing proteins, releasing energy from muscle storage, and regulating body temperature. Calcium (Ca) is a major mineral essential for healthy bones and teeth [16].

Chillie is an essential ingredient in Sri Lankan meals and as chillie powder is daily consumed by Sri Lankans through the meals, it is very important to gather information about the mineral composition present in chillie powder from a nutritional point of view. Therefore this study was carried out to determine the macro metal content in commercially available chillie powder samples in the Sri Lankan market.

The major objective of this study was to determine the content of some selected macro metals in chillie powder. The specific objectives of this study were to determine the Na, K, Mg and Ca contents in chillie powder available in the Sri Lankan market, using selected brands; to find out whether there is a significant difference in metal content among the brands, with the unbranded sample and with the control; and to find out whether there is a significant difference among batches tested in a same brand.

II. MATERIALS AND METHODS

Sample selection

Five brands of chillie powder samples named A, B, C, D and E, and an unbranded sample were selected

for this study. Three batches from each brand and three samples from each batch were purchased from the local markets in Sri Lanka, within the time period of January to April in 2015. Three samples of the unbranded sample were also collected. An unground whole dry chillie sample was selected as the control.

Sample preparation for atomic absorption spectrometric analysis

Wet digestion method was carried out according to the AOAC official method 975.03 [1].

Method

Chillie powder sample was placed in a porcelain dish and oven dried at 105 °C for 2 hours, until obtaining a constant weight. Accurately about 1 g of oven dried chillie powder sample was weighed into a 150 mL Pyrex beaker. Then 10 mL of HNO₃ (1+1) was added. It was allowed to soak thoroughly and 3 mL of 60% HClO₄ was added. Then it was heated on a hot plate, placed in a fume hood, slowly at first, until frothing ceased. The heating was carried out at 150 °C, until HNO₃ was almost evaporated. Then it was cooled and 10 mL of HNO₃ was added again and continued heating due to the charring of the sample. The addition of 10 mL of HNO₃ and heating were repeated another four times, until obtaining a clear solution, without charring. It was heated to white fumes of HClO₄; cooled; 10 mL of HCl (1+1) was added and quantitatively transferred into 50 mL volumetric flask. To the solution in 50 mL volumetric flask, 10 mL of 5 % Lanthanum (La) solution was added and diluted to volume using de-ionized water. It was kept for a while to settle silica. Then it was filtered through an acid washed No. 41 filter paper after discarding the first few drops of filtrate. This digested solution was stored in a polypropylene bottle for the AAS analysis. A blank was prepared by following all the above steps, at the same conditions, without adding the sample.

Flame Atomic absorption spectrometric (AAS) analysis

The standard series of each metal was prepared, using the stock solutions. When preparing Ca standard series, 10 mL of 5% Lanthanum (La) solution was added to the standard in 50 mL volumetric flask, so that final dilutions contained 1% La.

The metals in the digested solutions of chillie powder samples, namely Na, K, Mg and Ca were analyzed using the flame AAS technique. This metal analysis

was performed using Thermo Scientific, iCE 3000 series model double beam atomic absorption spectrometer, with deuterium background correction. The instrument was set up using the operating parameters given in the Table I. The Solaar software

was used to run the programme. Hollow cathode lamp with each metal was used at the specific wavelength.

Table I: Operating parameters of element for flame AAS

Element	Wave length (nm)	Flame type	Fuel flow rate (L/ min)	Lamp current (mA)	Slit width (nm)
Na	589.6	Air / acetylene	0.9 – 1.2	5	1.0
K	766.5	Air / acetylene	1.1 -1.3	5	1.0
Mg	285.2	Air / acetylene	0.9 – 1.2	4	0.5
Ca	422.7	Air / acetylene	4.0 -4.4	10	0.5

standard series. Then digested solutions of chillie powder samples were aspirated and the concentration was read from the plot of absorbance against concentration (µg/mL). The prepared blank was used to establish the 0 absorption point. The burner was flushed off with de-ionized water between samples. For the K determination only, the samples were diluted 50 times with 10% HCl to obtain solutions within the range of instrument. Dilution factor was considered in the K analysis.

Determination of the original metal content in the sample on dry basis

$$\text{Metal content in the original sample } (\mu\text{g/g, DW}) = \frac{C \times V}{W}$$

C = Concentration of the metal given by AAS reading (µg/mL)

V = Final volume of the digested sample (mL)

W = Weight of the dried sample (g)

Sample preparation for X-ray fluorescence spectrometric analysis

For the sample preparation, palletizing technique was used, after dry ashing the sample at 300 °C, according to the validated method of Atomic Energy Board of Sri-Lanka.

Method

Chillie powder sample was placed in a silica dish and oven dried at 105 °C for 2 hours, until obtaining a constant weight. Accurately about 20 g of oven dried chillie powder sample was weighed into another silica dish and ashed in the muffle furnace (Thermolyne, F62730) at 300 °C for 16 hours. Then the dish was weighed again to obtain the ash weight. From the ashed sample, 0.50 g of ash was weighed into a clean oil paper. Then it was transferred into a hard plastic mortar and ground into a homogeneous powder. Then this powder was transferred into a cylindrical pellet die, as evenly distributed and pressed using a manually operated bench top pelletizing press (Spectro Press, 4312 E) to form a supported pellet which ideally had a smooth, homogeneous sample surface and good physical stability.

X-ray fluorescence (XRF) spectrometric analysis

This method was used to analyze K and Ca contents in the chillie powder samples. One sample from the each batch, with the unbranded sample and the control was analyzed.

Method

The instrument was switched on and wait one hour to warm up before proceeding, to stabilize the machine. The prepared pellets were loaded into the sample holder. The instrument was set up using the operating parameters given in the Table II. The AXIL software was used to run the programme.

Table II: Operating parameters for XRF

Operating parameter	
Run time / s	500
X-ray tube voltage / kV	40
X-ray photon emitting current / mA	20
Tube current / mA	10
Detector voltage / V	-500
Target	Mo

The sample was run 500 seconds using the molybdenum (Mo) target and the spectrum of count per channel against channel number was obtained. The metals were identified using their characteristic fluorescent energy value. The quantification of identified metals was carried out by the calibration curve method, obtained with standard samples.

Determination of the original metal content in the sample on dry basis

$$V \text{ Metal content in the original sample } (\mu\text{g/g, DW}) = \frac{C \times W_a}{W_t}$$

- C = Concentration of the metal given by XRF (μg/g)
- Wt = Weight of the dried sample (g)
- Wa = Weight of the ash obtained (g)

Statistical analysis

Results were statistically analyzed using one-way analysis of variance (ANOVA), at 0.05 probability level with MINITAB-14 software package. The Bartlett's test (normal distribution) was performed to check whether there is any significant difference between the batches in each brand in mean metal contents. One-way ANOVA was used to check whether there is a significant difference among the brands in mean metal contents. Tukey's pairwise comparison was performed to select the brand pairs which are significantly different in mean metal contents. Dunnett's comparison test was performed to check whether selected chillie powder samples are significantly different from the control sample in the mean metal content. The results of metals obtained by the AAS and XRF techniques were compared using the paired t test, to check whether there is any significant difference between these two methods.

III. RESULTS AND DISCUSSION

In this study, macro metal contents of chillie powder samples were analyzed using selected five brands of chillie power samples and an unbranded sample, commonly available in the Sri-Lankan market. An ungrounded whole dry chillie sample was selected as the control.

The macro metals in chillie powder samples were analyzed using flame AAS and XRF spectrometric techniques. Table III shows the mean Na, K, Mg and Ca contents in each batch of five chillie powder brands, namely A, B, C, D and E, obtained using flame AAS technique. The results are expressed as µg metal /g of dried chillie powder (ppm).

Table III: Mean macro metal content in chillie powder samples (flame AAS) – batchwise

Brand	Batch	Mean metal content (µg/g, DW)			
		Na	K	Mg	Ca
A	1	290.7 ± 1.5 ^a	24310.1 ± 1.4 ^a	1991.8 ± 0.8 ^a	1048.8 ± 0.4 ^a
	2	289.9 ± 1.2 ^a	24316.5 ± 1.3 ^a	1991.3 ± 0.8 ^a	1046.7 ± 0.8 ^a
	3	291.3 ± 1.3 ^a	24310.5 ± 2.0 ^a	1990.9 ± 0.4 ^a	1048.0 ± 0.8 ^a
B	1	256.9 ± 1.3 ^b	33922.1 ± 1.2 ^b	2181.7 ± 0.9 ^b	1534.8 ± 0.4 ^b
	2	254.9 ± 0.3 ^b	33935.0 ± 1.3 ^b	2179.3 ± 1.1 ^b	1533.1 ± 0.7 ^b
	3	255.8 ± 0.4 ^b	33929.0 ± 1.2 ^b	2180.7 ± 0.5 ^b	1534.2 ± 0.7 ^b
C	1	183.2 ± 0.3 ^c	32735.2 ± 1.3 ^c	2173.2 ± 0.3 ^c	955.5 ± 0.7 ^c
	2	181.4 ± 0.6 ^c	32741.9 ± 1.2 ^c	2171.8 ± 0.8 ^c	953.8 ± 0.6 ^c
	3	182.6 ± 0.8 ^c	32738.5 ± 0.9 ^c	2172.7 ± 0.4 ^c	956.2 ± 0.3 ^c
D	1	309.8 ± 0.3 ^d	23369.4 ± 0.7 ^d	2154.9 ± 0.1 ^d	961.0 ± 0.2 ^d
	2	309.0 ± 0.2 ^d	23372.7 ± 1.0 ^d	2153.9 ± 0.2 ^d	962.2 ± 0.4 ^d
	3	308.7 ± 1.1 ^d	23375.0 ± 1.1 ^d	2154.9 ± 0.2 ^d	962.9 ± 0.3 ^d
E	1	265.3 ± 0.3 ^e	22865.6 ± 1.2 ^e	2046.1 ± 0.2 ^e	821.5 ± 0.9 ^e
	2	263.9 ± 0.3 ^e	22869.5 ± 0.6 ^e	2045.8 ± 0.2 ^e	822.9 ± 0.4 ^e
	3	264.8 ± 0.3 ^e	22864.7 ± 1.2 ^e	2045.0 ± 0.2 ^e	821.6 ± 1.0 ^e

Results are mean ± standard deviation of three replicates and calculated on dry weight basis. Means within the same column that have same common letters are not significantly different (p >0.05).

Analysis of variance (ANOVA) at 0.05 probability level was carried out using the MINITAB-14 software package. According to the Bartlett's test (normal distribution) results, there was no significant difference (p >0.05) between the batches in each

brand, A, B, C, D and E, in mean Na, K, Mg and Ca contents. Table IV shows the mean Na, K, Mg and Ca contents in each chillie powder brand and unbranded sample with the control, obtained using flame AAS technique.

Table IV: Mean macro metal content in chillie powder samples (flame AAS) - brandwise

Chillie powder sample	Mean metal content (µg/g, DW)			
	Na	K	Mg	Ca
Brand A	290.6 ± 3.2 ^a	24312.4 ± 3.4 ^a	1991.3 ± 0.7 ^a	1047.8 ± 1.1 ^a
Brand B	255.9 ± 2.7 ^b	33928.7 ± 5.7 ^b	2180.5 ± 1.3 ^b	1534.0 ± 0.9 ^b
Brand C	182.4 ± 2.3 ^c	32738.5 ± 3.0 ^c	2172.6 ± 0.7 ^c	955.2 ± 1.2 ^c
Brand D	309.2 ± 1.9 ^d	23372.4 ± 2.6 ^d	2154.6 ± 0.5 ^d	962.0 ± 0.9 ^d
Brand E	264.6 ± 1.6 ^e	22866.6 ± 2.4 ^e	2045.7 ± 0.5 ^e	822.0 ± 1.0 ^e
Unbranded sample	510.0 ± 0.2 ^f	25547.0 ± 1.0 ^f	2075.4 ± 0.2 ^f	1085.8 ± 0.5 ^f
Control	705.6 ± 0.4 ^g	25166.7 ± 1.5 ^g	2067.8 ± 0.2 ^g	1495.2 ± 0.3 ^g

Results are mean ± standard deviation of nine replicates and calculated on dry weight basis.

Means within the same column that have no common letters are significantly different (p <0.05).

One way ANOVA was used to check whether there is a significant difference between brands in mean metal contents. Due to p-value is less than 0.05, there are significant differences among brands in the mean Na, K, Mg and Ca contents. Tukey's pairwise comparison was performed to select the brand pairs which are significantly different in mean metal contents. According to the test results, all brands are significantly different to each other and with the unbranded sample in mean Na, K, Mg and Ca

contents. Dunnett's comparison test was performed to check whether selected chillie powder samples are significantly different from the control sample in the mean metal content. According to the test results, all brands are significantly different from the control in mean Na, K, Mg and Ca contents. Mean Na, Mg and Ca content in chillie powder samples are graphically represented in the Figure I, while mean K content is represented in the Figure II.

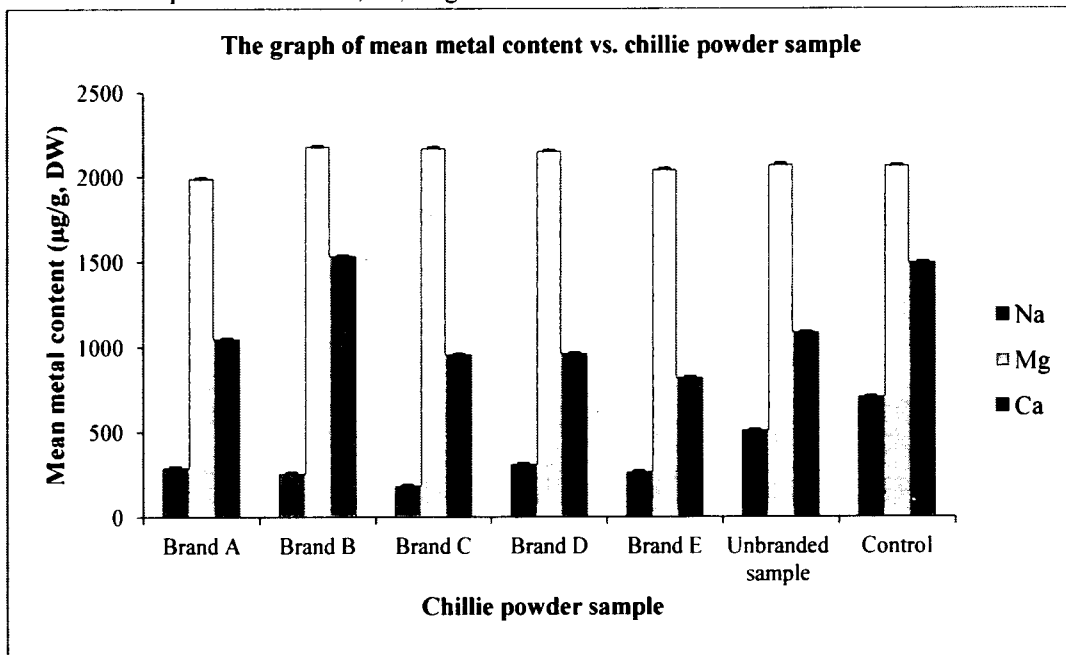


Figure I: Variation of Na, Mg and Ca contents in chillie powder samples (flame AAS)

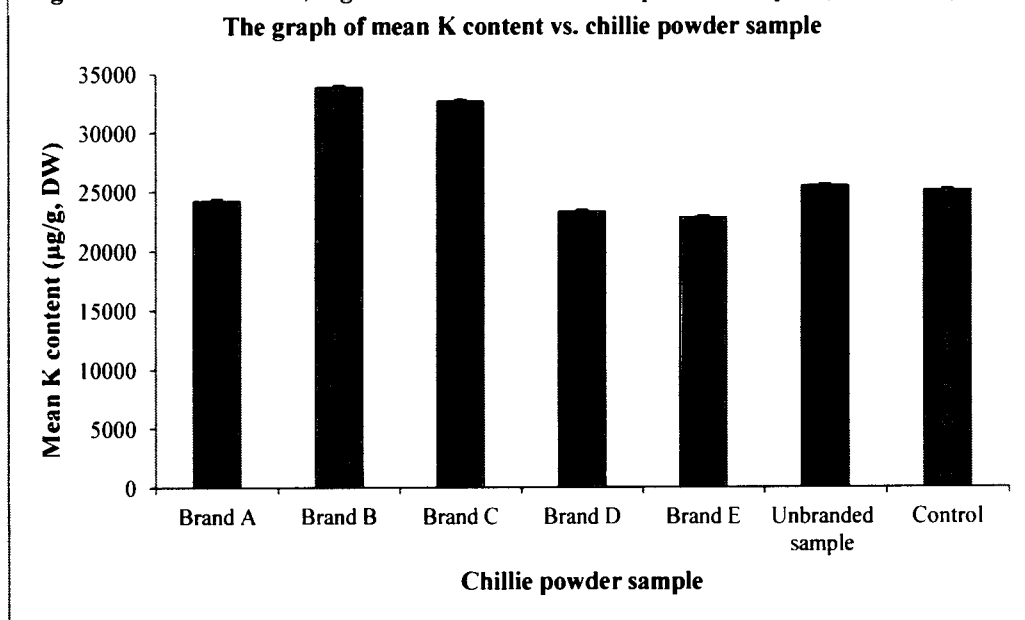


Figure II: Variation of K content in chillie powder samples (flame AAS)

Table V shows the K and Ca contents in chillie powder samples, according to the XRF spectrometric

results. The results are expressed as μg metal /g of dried chillie powder (ppm).

Table V: Macro metal content in chillie powder samples (XRF) - brandwise

Chillie powder sample	Metal content ($\mu\text{g/g}$, DW)	
	K	Ca
Brand A	24 318.1 \pm 337.3	1 049.6 \pm 72.4
Brand B	33 940.2 \pm 195.1	1 536.4 \pm 52.6
Brand C	32 746.9 \pm 240.9	956.0 \pm 52.5
Brand D	23 379.7 \pm 160.0	965.9 \pm 31.8
Brand E	22 865.3 \pm 149.5	821.0 \pm 32.8
Unbranded sample	25 553.8 \pm 181.8	1 086.1 \pm 44.0
Control	25 165.4 \pm 264.3	1 495.9 \pm 95.7

Results are metal content \pm measurement uncertainty of each sample, calculated on dry weight basis.

The contents of K and Ca in chillie powder brands, obtained by XRF technique are graphically represented in the Figure III.

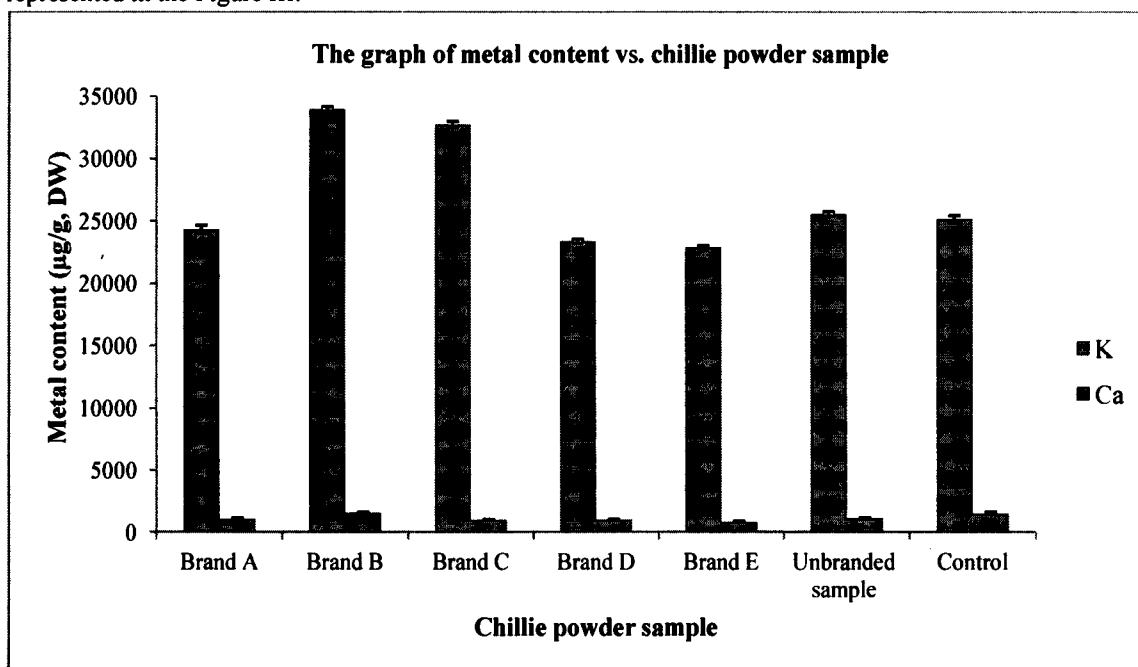


Figure III: Variation of K and Ca content in chillie powder samples (XRF)

The results of macro metals obtained by the AAS and XRF techniques were compared using the paired t test, to check whether there is any significant difference between these two methods. According to the test results, there was no significant difference ($p > 0.05$) between these two methods, AAS and XRF in K and Ca contents. The Na and Mg contents were unable to be detected by the XRF spectrometric technique, due to instrumental limitations and low X-

ray yields for the light elements. It is often difficult to quantify elements lighter than Na ($Z = 11$), unless background corrections and very comprehensive inter-element corrections are made [12].

According to the flame AAS analysis, the mean Na content of chillie powder samples was within the range of 182.4 to 510.0 $\mu\text{g/g}$ (on dry weight basis). But the control sample was having mean Na content of 705.6 $\mu\text{g/g}$. The mean Ca content of chillie powder

samples was within the range of 822.0 to 1534.0 µg/g (on dry weight basis). The reason for low Na content in chillie powder samples, compared to the control, and low Ca content in some brands (brand C, D and E), may be due to the loss caused by heat generation during the chillie grinding process or may be adulterated with plant materials with low Na and Ca content.

In the chillie industry, the main aim of grinding is to obtain smaller particle size with good product quality in terms of flavour and colour. In the normal grinding process, heat is generated when energy is used to fracture a particle into a smaller size. This generated heat usually is detrimental to the product and results in some loss of flavour and quality. During grinding, the temperature of the product rises to a level in the range of 42 – 95 °C and varies with the oil and moisture content of the chillies, but chillies lose a significant fraction of their volatile oil or flavouring components due to this temperature rise [18]. Hence minerals like Na and Ca associated with volatile oil or flavouring components may be lost during the grinding process.

In the AAS analysis phosphorous (P) interferes in Ca determination with air-acetylene flame. To eliminate this interference, La was added to the Ca standard series and also to the sample solutions.

The mean K content of chillie powder samples was within the range from 22 866.6 to 25 547.0 µg/g (on dry weight basis), while mean Mg content varies in the range of 1991.3 – 2180.5 µg/g (on dry weight basis). There are significant differences in these macro mineral contents among the brands. These variations may be due to multiple factors, such as the differences in soil conditions where it is grown, variety of chillie, maturity of chillie, growing season, climatic condition, processing treatments and preservation method [17].

IV. CONCLUSION

This study was carried out to investigate the macro metal content present in the selected chillie powder samples. According to the results, Na content of all the chillie powder samples were found to be lower than the control (705.6 ± 0.4 µg/g) and Ca content of some brands (brand C- 955.2 ± 1.2 µg/g, brand D- 962.0 ± 0.9 µg/g and brand E- 822.0 ± 1.0 µg/g) were also found to be lower than the control (1495.2 ± 0.3 µg/g). The reason for this may be due to the loss

caused by heat generation during the chillie grinding process or may be due to adulteration with plant materials with low Na and Ca content. The mean K content of chillie powder samples was in the range from 22 866.6 to 25 547.0 µg/g, while the mean Mg content varied in the range of 1991.3 – 2180.5 µg/g. These variations may be due to multiple factors, such as the differences in soil conditions where it was grown, variety of chillie, maturity of chillie, growing season, climatic condition, processing treatments and preservation method.

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REFERENCES

- [1] AOAC (1996), Association of Analytical Communities, Official Methods of Analysis of AOAC International, 16th edition, Vol. II
- [2] Balcerzak, M. (2002), Sample Digestion Methods for the Determination of Traces of Precious Metals by Spectrometric Techniques, *Analytical Sciences*, Vol. 18, pp. 737-750
- [3] Belay, K. (2014), Analysis of Lead, Cadmium and Chromium in Ethiopian Spices after Wet (acid) Digestion using Atomic Absorption Spectroscopy, *Global Journal of Science Frontier Research : (B) Chemistry*, Vol. 14, Iss.4 No. 1.0, pp. 1-5
- [4] Belay, K. (2014), Determination of Pb (II), Cr (VI) and Cd (III) contents in Ethiopian Spices after Microwave Digestion using Atomic Absorption Spectroscopy, *International Journal of Technology Enhancements and Emerging Engineering Research*, Vol. 2, Iss. 8, pp.87-91
- [5] Buhrke, V.E., Jenkins, R. and Smith, D.K. (1998), A Practical Guide for the Preparation of Specimens for X-Ray Fluorescence and X-Ray Diffraction Analysis, pp.39
- [6] DASL (2006), Department of Agriculture - Sri Lanka, Crop Recommendations – Chillie
- [7] Deshpande, S.S. and Dekker, M. (2002), Hand book of Food Toxicology, *New York*
- [8] FSAI (2009), Food Safety Authority of Ireland, Mercury, Lead, Cadmium, Tin and Arsenic in

- Food, *Toxicology Factsheet Series*, No. 1, pp. 1-13
- [9] FSSAI (2012), Food Safety and Standards Authority of India, Manual of Methods of Analysis of Food Metals
- [10] Garcia, R. and Baez, A.P. (2012), Atomic Absorption Spectrometry (AAS), pp. 1-11
- [11] Inam, F., Deo, S. and Narkhede, N. (2013), Analysis of Minerals and Heavy Metals in Some Spices Collected from Local Market, IOSR Journal of Pharmacy and Biological Sciences, Vol. 8, Iss. 2, pp. 40-43
- [12] Jenkins, R. (1988), X-Ray Fluorescence Spectrometry, 2nd edition, pp. 51-53
- [13] Krejpcio, Z., Krol, E. and Sionkowski, S. (2007), Evaluation of Heavy Metals contents in Spices and Herbs Available on the Polish Market, Polish Journal of Environmental Studies, Vol.16, No.1, pp. 97-100
- [14] MAGI (2009), Ministry of Agriculture – Government of India, Post Harvest Profile of Chillie, pp. 6-16
- [15] Pearson D. (1976), The Chemical Analysis of Foods, 7th edition, pp. 73-97
- [16] Ranaweera, K.K.D.S. (2007), All About Foods, Godage International Publishers (Pvt) Ltd, pp. 153-176
- [17] Sarker, M.S.H., Hasan, S.M.K. and Aziz, M.G. (2012), The effects of processing treatments on the shelf life and nutritional quality of green chilli (*Capsicum annum* L.) powder, *Journal of Tropical Agricultural Science*, No.35, Vol.4, pp. 843-852
- [18] Singh, K.K. and Goswami, T.K. (1999), Design of a cryogenic grinding system for spices, *Journal of Food Engineering*, No.39, pp.359-368