Jurnal Sains Nuklear Malaysia, 2018, 30(1): 30 – 35
ISSN: 2232-0946

A PRELIMINARILY STUDY OF ELEMENTAL CHARACTERIZATION FOR GEOCHEMICAL MARKERS OF HOUSE AND CAVE EDIBLE BIRD'S NEST USING NAA TECHNIQUE

Nazaratul Ashifa Abdullah Salim[1], Zainon Othman[2], Nur Afiqah Harun[1], Salmah Moosa[2], Siti Aminah Omar[1], Muhammad Azfar Azman[1], Md Suhaimi Elias[1], Shamsiah Abdul Rahman[1], Lim Ching Choah[1] and Zawiyah Sharif[3]

[1]Waste and Environmental Technology Division,
Malaysian Nuclear Agency, Bangi, 43000 Kajang, Selangor
[2]Agrotechnology and Biosciences Division,
Malaysian Nuclear Agency, Bangi, 43000 Kajang, Selangor
[3]Food Safety and Quality Division, Ministry of Health, Aras 4, Menara Prisma, No 26, Jalan Persiaran Perdana, Presint 3, Pusat Pentadbiran Kerajaan Persekutuan,
62675 Wilayah Persekutuan Putrajaya
Correspondence author: shifa@nuclearmalaysia.gov.my

ABSTRACT

For addressing the issue of adulteration and counterfeiting of the sources of origin for the edible bird's nests (EBNs), a preliminary study of elemental characterization for their geochemical markers was performed. This study aims to characterize the elemental profiles as geochemical markers and evaluate the relationships between these markers which could be used for the verification of the geographical origin of EBN in Malaysia. EBN samples from house and cave nests that represent a range of geographical and environmental characteristics were analysed using Neutron Activation Analysis (NAA) technique. The method was validated by analysing the Standard Reference Material SRM-1515 (Apple Leaves) and SRM-1573a (Tomato Leaves) of NIST. A total of 18 elements were determined. The results showed a statistical analysis of one way ANOVA has proven that there have a significant different elemental profiles in both EBN, clearly V, As, Ba, Sc, Sm and Th were not detectable in the house EBNs. Therefore, initial finding could be concluded that those elements can be identified as the key elements for geochemical marker to differentiate the type of EBN. Furthermore the differnces probably due to natural presence of those elements as well as regional or location and seasonal variation factors affecting the dietry habits of the swiftlets.

Keywords: Edible bird's nest, elements, geochemical markers, Neutron Activation Analysis, Statistic

INTRODUCTION

The capability to certify food origin is becoming significant economic importance beside the issue in quality control and safety of food. In several countries, the use of geographical indications allows producers to obtain market recognition and often a premium price. For example, some food products such as wines, cheeses, oils, honey, beers, meats, pistachios and potatoes are marketed using labels (e.g. GI, Geographic Indication) that are based on standards of identity or composition related to a very specific production area. Europe has developed a system which provides legal protection to regional foods through the Protected Designation of Origin (PDO) labels (EU Regulation, 1982) to ensure the safety and quality of the products protecting them against fraud and imitation (Gonzalvez et. al., 2009).

Edible bird’s nest (EBN) is one the most highly valued food products of South East Asia that widely consumed as a health food due to its high beneficial effect on human health.. The nest is made by two main swiftlets species; the White-nest swiflet (*Aerodramus fuciphagus*) and the Black-nest swiflet (*Aerodramus maximus*) with a high protein glutinous secretion produced by their salivary glands. Their habitats range from the Nicobar Islands in the Indian Ocean to sea caves in the coastal regions of Thailand, Vietnam, Indonesia, Borneo and the Palawan Islands in the Philippines. Malaysia is situated right at the heart of the “golden triangle”of swiftlet bird nest production, making it a strong producer in this lucrative agriculture industry (McAfee, 2011). The edible bird’s nest (EBN) industry has been identified as one of the National Key Economic Area (NKEA) for the agricultural sector under the Economic Transformation Programme (ETP) of Malaysia. EBN is a very lucrative agricultural industry in Malaysia that mostly harvested in wild or farmed in special house, fetching over US $6 billion worth of global sales.

Issues facing the EBN industry include unknown source of origin with various quality grades, thus affecting export market and production subjected to fraud such as adulteration and counterfeiting. In addition, consumers in key markets are increasingly concerned with the origin of their food and are willing to pay more if there can be assured of its origin. Therefore, development of appropriate analytical tools to ensure geographical traceability is indeed essential. In addressing food safety issues in Malaysia, the Food Hygiene Regulations 2009 has provision for traceability but mechanism on how traceability can be achieved is not prescribed. Present mechanism is mainly paper-based system that pass information along with the commodity. However, such system is subject to failure either inadvertently or deliberately (fraud). Thus there is a need for an independent system to verify the origin of food and hence audit the traceability control systems.

Nuclear techniques involving elemental and stable isotopes fingerprinting provide a robust analytical tool for use in conjunction with food safety surveillance programmes to provide independent verification of food traceability systems (IAEA, 2013). Techniques such as Neutron Activation Analysis (NAA) for determining elemental composition and isotope ratio mass spectrometry (IRMS) to measure stable isotope ratio (SIR). It is well known that the content of selected minerals and trace elements in foods clearly reflects the soil type and the environmental growing conditions that able to be used in assuring the geographical origin of food samples (Gonzalvez , 2011).

This study aims to characterize the elemental profiles as geochemical markers and evaluate the relationships between these markers which could later be used for the verification of the geographical origin of EBN in Malaysia. Hypothesis of this study is that the trace-element of the EBN from two sources will provide unique and representative fingerprints that make it possible to differentiate between samples of different origins.

METHODOLOGY

Samples were received from Ministry of Health in total of 17 samples consisted of 7 samples from house nest and 10 samples from cave nest of unknown specific location. The samples were already in dried condition and ground into powder form. Approximately 500 mg of powdered EBN samples were weighted into polyethylene irradiation vials. Standard reference materials (SRMs) were co-irradiated with the samples, standard and blank to ensure accuracy and precision of data. The irradiations for NAA were performed in the TRIGA Mark II reactor at Malaysian Nuclear Agency that operated at 750 kW with a thermal neutron flux of about 2.5×10^{12} $cm^{-2}s^{-1}$. For short irradiation, all samples, standard, blank and SRMs were irradiated for a period of 60 seconds, and the cooling time for 10 min for first counting, and the second counting conducted after 24 hrs. In long irradiation, each batch containing the EBN samples, standard, blank and were irradiated for 6 hrs. The cooling times took 3 days for first counting, and 3 weeks for the second counting. A high resolution coaxial Ortec Hyper-Pure Germanium detector (HPGe) with a resolution of 1.9 keV at 1332 keV gamma-rays line of Co-60 was used for counting. The spectral data of 18 elements were processed using the spectrum analysis software namely Gamma Visions. Quantification of major, minor and trace elemental concentrations were based on relative method and data were reported in dry weight (d.w.).

Statistical Analysis

The elemental values were analyzed using one way analyisis variance (ANOVA) at $p < 0.05$ followed by Kruskal-Wallis H test using Past3.12 software (folk.uio.no/ohammer/past) to compare the values between the cave and the house nest. Kruskal-Wallis H test was choosen because it does not require the assumption of data normality.

RESULTS AND DISCUSSION

Quality Assessment

Standard reference material namely Tomato Leave 1573a and Apple Leaves 1515 provided by National Institute of Standard and Technology (NIST) were applied as quality control material of each analytical regiment implemented. The accuracy of the analytical technique was evaluated by analyzing the SRM using a Z-score calculation (Crosby et. al., 1995). The Z-score of an element concentration was computed based on the following equation:

$$Z - score = \frac{x - c}{\sqrt{u_x^2 + u_c^2}}$$

Where x is the analytical results; c is the certified value, u_x is the uncertainty of analytical results, and u_c is the uncertainty of certified value. The uncertainty of the analytical results were obtained after estimating the contribution from the sources in example errors in weighing, spectral peak counts, background counts and uncertainties in standard value whilst the uncertainty of the certified value was based on the certificate. For acceptance of results: $-2 < Z < 2$ is anticipated. However, if $Z < 3$ and $Z > 3$, it is consider that the result is "out-of-control" and corrective action will be taken, respectively.Whilst the precision simply described by the relative standard deviation of the measurement of the proposed SRMs.

Results obtained for the SRMs are presented in Table 1. Data obtained indicate good precision and good agreement with the certified values. Analytical results disclosed that the precision of the analysis is satisfactory where percentage relative standard deviations (RSD) of the SRMs results were lower than 20% and relative errors varied from 0 to 19.2%. The Z-score values obtained for most of the elements analyzed from both SRMs were |Z-score| ≤ 2, indicating that the results are satisfactory and are within the ranges of certified data at the 95% confidence level. However, some elements were not indicated any Z-score value as in the cerificate they are given only information value of concentrations.

Table 1: Results of analysis of Standard Reference Materials (in mg/kg)

Elements	Measurement Value	Certified or Recommended Value	Error (%)	RSD (%)	Z-score
Tomato Leave 1573a					
Al					
Ba	600 ± 11	598 ± 12	0.33	1.83	-0.12
Br	59 ± 6	63	-6.35	10.17	
Ca	1200 ± 50	1300	-7.69	4.17	2.00
Co	50800 ± 762	50500 ± 900	0.59	1.50	-0.25
Cr	0.67 ± 0.05	0.57 ± 0.02	17.5	7.46	-1.86
K	1.91 ± 0.1	1.99 ± 0.06	-4.02	5.24	0.69
Mg	26000 ± 540	27000 ± 500	-1.48	2.03	0.54
Mn	11930 ± 850	12000	-0.58	7.12	
Na	238 ± 11	246 ± 8	-3.25	4.62	0.59
Rb	144 ± 10	136 ± 4	5.88	6.94	-0.74
Sc	13.5 ± 0.7	14.89 ± 0.27	-9.34	5.19	1.85
Sm	0.101 ± 0.01	0.1	1.04	9.90	
Th	0.17 ± 0.02	0.19	-10.5	11.8	
V	0.118 ± 0.01	0.12	-1.67	8.47	
Zn	0.766 ± 0.05	0.835 ± 0.01	-8.26	6.53	1.35
	28.1 ± 2.0	30.9 ± 0.7	-9.06	7.12	1.32
Apple Leaves 1515					
Al					
Ba	288 ± 15	286 ± 9	0.70	5.21	-0.11
Br	19 ± 5.5	49	0	11.2	
Ca	1.67 ± 0.2	1.8	-7.22	12.0	
Cl	15100 ± 230	15260 ± 150	-1.05	1.52	0.58
K	562 ± 28	579	-2.94	4.98	0.61
Mg	16500 ± 260	16100 ± 200	2.48	1.58	-1.22
Mn	2880 ± 120	2710 ± 80	6.27	4.17	-1.18
Na	54 ± 6	54 ± 3	0	11.1	0.00
Rb	28.4± 3	24.4 ± 1.2	16.4	10.6	-1.24
Sc	10.2 ± 1.9	10.2 1.5	0	18.6	0.00
Sm	0.030 ± 0.005	0.03	0	16.7	
V	3.27 ± 0.20	3	9.00	6.12	
Zn	0.21 ± 0.04	0.26 ± 0.03	-19.2	19.0	1.00
	12.9 ± 0.8	12.5 ± 0.3	3.20	6.20	-0.47

Elemental Concentration in EBN

The mean, maximum and minimum values observed for 18 elements in EBN samples are presented in Table 2. Both type of EBN showed relatively high amount of essential macronutrient particularly Al, Ca, Cl, K, Mg and Na. The EBN from cave had highest concentrations of Al, Ca and Mg. Meanwhile concentrations of Cl, K and Na in house type are more abundance. The Ca levels of the EBN are among the highes of all the elements analyzed. The cave nests have significantly higher levels ($p < 0.05$) Ca compared to house nests. Since calcium in the form of calcium carbonate is the major component of cave natural structure. Thus, the effects of weathering and erosion process occurred on the cave walls, resulted calcium (calcium carbonate) is dispersed in the cave environment and then attached onto the bird nests. The element of Al, Cl, K and Na were significantly different ($p < 0.05$) among the type of EBN except for Mg. The differences of Al, Cl, K and Na concentrations in both type of EBNs could be due to natural presence of those elements and different levels exposure of those elements to the swiftlets and nests. Other factors such as regional or location and seasonal variation affected the dietry habits of the swiftlets (Chen et. al., 2014).

Table 2: Elemental concentrations in dry weight of EBN

Elements	**House EBN (n = 7)**				**Cave EBN (n = 10)**			
	Average	**SD**	**Min**	**Max**	**Average**	**SD**	**Min**	**Max**
Al (mg/kg)	33.7	33.9	5.58	100	3240	1435	689	4366
Ca (%)	0.69	0.06	0.59	0.75	1.36	0.71	0.77	2.59
Cl (mg/kg)	2392	1030	912	3915	42.1	13.7	19.5	58.8
K (mg/kg)	311.6	70	231	431	187	106	147	506
Mg (%)	0.12	0.02	0.10	0.15	0.17	0.05	0.09	0.21
Mn (mg/kg)	1.34	1.00	0.44	3.43	4.52	3.30	1.33	11.0
Na (%)	0.61	0.14	0.37	0.81	0.03	0.07	0.02	0.22
V (mg/kg))	<0.03				0.19	0.09	0.07	0.33
As (mg/kg)	<0.05				0.18	0.06	0.10	0.30
Ba (mg/kg)	<1.0				8.41	2.30	3.86	11.1
Br (mg/kg)	3.53	1.23	1.63	5.33	0.36	0.24	0.19	0.99
Co (mg/kg)	0.06	0.01	0.04	0.08	0.07	0.02	0.05	0.11
Cr (mg/kg)	0.55	0.09	0.46	0.68	0.40	0.28	0.17	1.17
Rb (mg/kg)	1.05	0.23	0.78	1.23	<0.1			
Sc (mg/kg)	<0.001				0.005	0.003	0.004	0.012
Sm (mg/kg)	<0.002				0.010	0.003	0.004	0.015
Th (mg/kg)	<0.01				0.02	0.01	0.01	0.04
Zn (mg/kg)	9.05	8.40	2.28	23.0	6.49	2.90	2.80	12.8

The cave nests demonstrated significant amount of minor and trace elements such as Mn, V, As, Ba, Br, Co, Cr, Sc, Sm, Th and Zn. However, statistical analysis has proven that there are no significant difference ($p > 0.05$) of Mn, Co, Cr and Zn between both type of EBN. On the other hand, only few elements showed detectable amount of concentration in the house EBN samples. Therefore, elements of V, As, Ba, Sc, Sm and Th can be identified as the key element for geochemical marker to differentiate both type of EBN. Those elements in cave nests signified their presence in the sample but not in the house nests. This may be due to the natural environment of the cave that promote some form of nutrient absorption into the birds' nest. The surrounding habitats, availability and abundance of food source could be associated to the assortment of elements found in this study (Lim, 2006).

CONCLUSIONS

The concentration levels of 18 elements were quantified in the house and cave EBNs. Several elements demonstrated significant contributions in the differentiation of two types of EBN samples. Meanwhile, six elements of V, As, Ba, Sc, Sm and Th were not detectable in the house EBNs. Thus, those elements can be identified as the key elements for geochemical marker to differentiate the type of EBN. However, it is recommended that a more comprehensive study be conducted involving more samples from different region or locations in order to confirm the present findings.

ACKNOWLEDGEMENTS

The authors wish to thank laboratory technicians from Analytical Chemistry and Applied Group for their help during sample preparation and analysis. The Reactor Group is also gratefully acknowledged for their assistance on sample irradiation.

REFERENCES

Chen, J.X.J, Lim, P.K.C., Wong, S.F. and Mark, J.W. (2014). Determination of the presence and levels of heavy metals and other elements in raw and commercial edible bird nests, *Malaysian Journal Nutrient*, 20(3): 377-391.

Crosby, N.T., Day, J.A., Hardcastle, W.A., Holcombe, D.G. and Treble, R.D. (1995). Quality in The Analytical Chemistry Laboratory, John Wiley & Son, NewYork.

Gonzalvez, A., Armenta, S. and de la Guardia, M. (2009). Trace-element composition and stable-isotope ratio for discrimination of foods with Protected Designation of Origin, *Trends in Analytical Chemistry*, 28(11): 1295-1311.

Gonzalvez, A., Armenta, S. and de la Guardia, M. (2011). Geographical traceability of ‘‘Arròs de Valencia’’ rice grain based on mineral element composition, *Food Chemistry* 126: 1254-1260.

International Atomic Energy Agency (IAEA) (2013). Food and Environmental Protection Newsletter, Vol 16, No. 1.

Lim, C.K. (2006). An overview for sustainable swiftlet farming in South-East Asia. In: Proceeding of First South-East Asia Swiftlet Farming Conference, Challenges to Sustainable Swiftlet Farming, Penang, pp:13-22.

McAfee, A. (2011). A brief look at edible-nest swiftlet industry. In: Proceedings of the International Conference and Training on Swiftlet Ranching, 17-19 July 2011, Terengganu, pp:1-11.