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Research Paper

Ethno-Medicinal Importance of Viviparous Gastropod, Bellamya bengalensis (Lamarck, 1822) in terms of Estimation of its Proximate Protein and Amino acid Composition

KEYWORDS : Bellamya bengalensis, Ethno-medicinal importance, Protein , Essential amino acid.

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ABSTRACT

People of North Eastern states of India, like West Bengal, Bihar, Nagaland, Mizoram etc depend on Bellamya bengalensis since an immemorial time beings. For its not only easy availability and cheap rate, in terms of money but also it has great nutritional importance. Moreover, people are very conscious about its ethno-medicinal values. Protein and its small unit amino acid play vital role, regarding to perform several physiological activities in the living animal including human. So in this context, study was made on the analysis of qualitative and quantitative protein and amino acid composition, in the flesh of B. bengalensis. Using Kjeldahl technique and HPLC methodology, protein and amino acids content were estimated, respectively. Beside the observation of rich source of digestible protein, all indispensable amino acids were isolated from the flesh of B. bengalensis, which ensured about the daily requirement for an adult human as per WHO's guide line. As this fresh water snail contains enormous source of nutrition in terms of protein, so here it is needed to turn into commercial utilization of this snail for socio economical development.

1. Introduction

Proteins participate in every single process within living cells. Where as Amino Acids are the building blocks of "Protein". Protein and amino acids have structural and mechanical function within the cytoskeleton of cells and muscles respectively in every living being. Not only that they play vital roles to maintain shape of the cells but also involve in cellular communication, immune responses, cells binding, cell replication, catalysts in metabolism etc. Out of the 20 amino acids, our body can make almost half of them internally. But the other half of amino acids are essential, meaning they cannot be synthesized (made or manufactured) by our body. Not only that, there are several indispensable amino acids, which are very effective for the children, to expedite their growth and maturation. As we know, several types of protein contain several types of amino acid, so, in that case, we have to choose that type of edible food materials which can provide our required level of amino acids in terms of protein. Nevertheless, in a number of times, regarding high market price, people can not reach to those food items, which they desire. So, in that case, they search for alternate food items, which have same or nearest same food value, comparison to their desire items. That is why, people of North Eastern states of India, like West Bengal, Bihar, Nagaland, Mizoram etc depend on *B. bengalensis* since a immemorial time beings. For its not only easy availability and cheap rate in terms of money, but also it has great nutritional significance (Mukherjee & Basu 2008). People observed that *B.bengalensis* has good taste in terms of its edibility, easy to digest and it has noble medicinal importance to those people who are suffering from several diseases (Roy & Singh 2007). *B.bengalensis* (Phylum: Mollusca, Class: Gastropoda, Order: Mesogastropoda, Family: Viviperidae), commonly known as "Gugli " (Bengali word) which is inhabitant of benthic region of fresh water body. It was usually observed that it attached either with the substances, which were immersed, or in floating condition over the water surface. However, regarding the knowledge of *B.bengalensis* there are few informations. Though some works were done on its taxonomy, eco-biological impact, distribution, availability, socioeconomic and marketing importance (Subba & Mitra,1982;Tanveer,1992; Subba & Pandey 2005; Farahnak, *et al*,2006**;** Roy & Singh,2007;Pir *et.al*.,2010; Baby *et.al*.,2010 Chakraborty, Mukherjee & Maity,2014). However, in present study, the objective was to determine Protein and Amino Acid composition of *B. bengalensis* in order to address the nutrition as well as ethno-medicinal importance of this species which makes the impression to its socio-economic contribution in terms of availability, desired amount of protein and amino acid.

2. Material and Methods

2.1 Sample Collection:

Healthy matured samples of *B. bengalensis* were collected at random from the different market places of Kolkata, West Bengal.

2.2 Sample preparation and preservation:

At first collected samples of *B. bengalensis* were properly washed with double distill water. Then length and weight was measured separately. Flesh portion was carefully removed from the shell, using cleaned forceps and scalpels and thus the edible flesh parts were separated. After separation of flesh, removing of unwanted matters, proper washing was taken place with double distill water. The flesh was dried, crushed and powdered and this dried powder was preserved in sterilized vials (Terson made) for analysis. The sample vials were then preserved in a desiccator.

2.3 Analysis of Protein Content:

N_2 content was determined according to Kjeldahl procedure (AOCS, 1991).The percentage (%) of nitrogen was calculated and the % of protein sample was obtained by multiplying % of N_2 with 6.25 .

2.4 Analysis of Amino acid Profile:

Amino acid profile was determined by High Performance Liquid Chromatography (HPLC) (Waters 2487 with UV detector, column, Milford, MA, USA; Nova-Pak C_{18}, 3.9 X 150 mm) analysis after hydrolysis of protein and derivation of amino acid was done by using diethyl-ethoxy-methylenemalonate (Alaiz *et.al*.,1992).

2.5 Statistical analysis:

Statistical Analysis mean, standard deviation and coefficient of variation were applied. For all determinations ten replicates were carried out. All analysis were carried out in triplicate.

3. Result

Here healthy mature adult specimen almost same sized of *B. bengalensis* was chosen for the analysis protein and amino acid content. The estimated length and weight of *B. bengalensis* were 3.84 ± 0.26 cm and 3.76 ± 0.15 gm respectively. The estimated protein content was 48.65 ± 0.85% (dry weight basis). Among the essential amino acids, Threonine (18.89± 0.88 %) gave

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highest result while sulphur containing Methionine (0.61 ± 0.39 %) indicated lowest result. Among non essential amino acid Glutamic acid (7.93 ± 0.61%) made highest result, while Glycine (1.85 ± 0.36%) indicated lowest result. The amount of amino acids Tryptophan and Proline could not be determined. Thus significant level (P <0.05) of essential amino acid were 66.96 ± 0.68 %, was isolated from *B. bengalensis*. So according to chemical structure of amino acid monoamino carboxylic (glycine, alanine, valine); mono-amino dicarboxylic (aspartic and glutamic acids); ß-hydroxyamino carboxylic (serine and threonine); basic (lysine, histidine, arginine); sulfur-containing (methionine and cysteine); ring-containing (phenylalanine, tyrosine) and leucines (leucine and isoleucine) (Zhelyazkova *et. al.* 2006) confirmed their presence (Table-1).

Table 1: Isolation of Protein and Amino acid content in Bellamya bengalensis and a parallel comparison was made with the (WHO et.al., 2007) report, on the daily requirement of protein and amino acid for child and adult human.

Item Name	Quantity	Requirement for Adult Human (gm/kg body wt./day) (WHO et.al., 2007)
Length measurement (cm)	3.84 ± 0.26	
Weight measurement (gm)	3.76 ± 0.15	
Crude Protein Content (% dry weight)	48.65 ± 0.85	0.66 to 0.75
Name of Amino Acid (% dry weight)	Quantity (gm./ 100 gm. of Protein)	Requirement for adult human (gm/ kg body wt./day) (WHO et.al., 2007)
Aspartic acid*	6.59± 0.72	--
Glutamic acid**	7.93 ± 0.61	--
Serine	7.62 ± 0.45	--
Histidine^{eaa}	6.19 ± 0.50	0.010
Glycine	1.85 ± 0.36	--
Threonine^{eaa}	18.89± 0.88	0.015
Arginine ^{eaac}	5.61 ± 0.59	--
Alanine	8.64 ± 0.23	--
Tyrosine^{eaac}	1.90 ± 0.60	Tyro+Phyala = 0.025
Valine^{eaa}	3.66 ± 0.12	0.026
Methionine^{eaas}	0.61 ± 0.39	0.010
Cysteine$^{eaac\ \&\ eaas}$	0.84 ± 0.82	0.004
Isoleucine^{eaa}	2.70 ± 0.33	0.020
Leucine^{eaa}	7.51 ± 0.26	0.039
Phenylalanine^{eaa}	2.10 ± 0.36	Tyro+phyala = 0.025
Lysine^{eaa}	16.13 ± 0.51	0.030
% of essential amino acid	66.96 (including Cysteine)	

*aspartic acid + aspargine, **glutamic acid + glutamine, eaa- essential amino acid, eaa**c**- essential amino acid for children, eaa**s**- sulphur containing essential amino acid, Tyro+Phyala = Tyrosine+Phenylalanine, Values are mean ± *SD*, n=10. (--) no information is available.

4. Discussion

Protein is an indispensable nutrient, which is a structural and functional unit of all living organisms. In our present study it was observed that crude protein content in *B. bengalensis* was 48.65 ± 0.85%. Whereas protein content in other edible Molluscs were 8.27 %, 12.36 %, 6.46 %, 12.92 % and 8.64 % in *Pila globosa*, *Melania tuberculata Lamellidens marginalis*, *Anisus convexiusculus* and *Helix* sp. respectively (Baby *et.al.*,2010).On the other hand it was observed that the avg. protein content in several fishes like; Carp 42-43 %, (Dabrowski, 1977), Pomfret 19% [4], Salmon 30% [4], Mackerel 30% [4]. While estimated protein content was reported in red meat group was, chicken 30% (Dolson, 2014), Mutton 21% (Williams,2007), Pork 22% (Dolson,2014), Beef 28% (Dolson,2014) etc. Therefore, in this regards it was cleared, this significant amount of protein which were not less than (rather, grater than) the fish and red meat communities. More than that, from the monetary point of view, it held much cheaper market rate than fish and red meat groups (Chakraborty, Mukherjee & Maity,2014).

It was narrated by (WHO *et.al*., 2007) that mean protein requirement for older, adults and the elderly lied in between 0.66 to 0.75 gm / kg body wt. /day. After making comparison with the (WHO's 2007) report, it was assumed, *B. bengalensis* confirmed such amount of protein and amino acid, which were essential for older, adults and the elderly to maintain their daily physiologic process.

Though for human, essential amino acids are Phenylalanine, Valine, Threonine, Tryptophan, Isoleucine, Methionine, Ieucine, Lysine, and Histidine (Young,1994),[(2)] and along with Cysteine, Argenine**,** Tyrosine are required for infants and growing children (WHO *et.al.,*2007). Where as Alanine, Serine, Glycine, Glutamine acid etc are the non essential amino acid. In addition to that Arginine, Glycine, Glutamine, Serin etc are considered as "Conditionally" essential amino acid (Reeds, 2000 and Fürst *et.*al, 2004). However, in this experiment a lots of important amino acids were detected (Table-1), these amino acids play vital contribution in controlling human physiological process. There presence in this experiment (in percentage) & importance is discussed as follows:

Argenine (5.61 ± 0.59 %) important for production of T-cells, good for people suffering from arthritis, helps in releasing the insulin from pancreas [(1)]. **Histidine** (6.19 ± 0.50 %) associate with pain control, neural deafness, production of white blood cells, aiding early digestion by stimulating stomach acid secretion treating[(1)]. **Isoleucine** (2.70 ± 0.33 %) is important for the formation of hemoglobin, stabilization and its regularization of blood sugar, repairing of muscle tissues, it works with Valine [(1,3)] . **Leucine** (7.51 ± 0.26 %) plays its role to releases a lots of energy during any form of exercise, tissue repair after surgery, blood sugar (diabetics) control, it also works with Valine[(1,3)].**Lysine** (16.13 ± 0.51%) improves concentration, preventing fertility problem, also helps in calcium absorption [(1)]. **Methionine** (0.61 ± 0.39 %) assists gall bladder and liver functions, balances the pH of urinary tract, beneficial for those who are suffering from osteoporosis, and gives rise to "Taurine" (an important neuron-regulator in the brain)[(1,3)]. **Phenylalanine** (2.10 ± 0.36 %) related to appetite control, blood pressure in hypotension, works with minerals in skin and hair pigmentation, produces adrenalin and noradrenalin, also used for rheumatoid arthritis, migraine pain[(1)]. **Threonine** (18.89± 0.88%) enhances the immune system, collagen production, maintains a proper protein balance in the body, does not allow fats to build up on the liver, mood elevation or depression and skin pigmentation, manufactures adrenalin, and precursors of thyroid hormone**(1)**. **Valine** (3.66 ± 0.12 %) regulates the metabolism, useful for the people who have just come out of drug addiction as they help to correct the amino acids deficiencies, caused due to drugs, also acts as a stimulant and provide energy during times of fasting or between meals[(1)]. **Tyrosine** (1.90 ± 0.60 %) is needed especially for infants regarding proper growth and development, also require for pituitary gland and the thyroid gland and it defends the skin against radiation[(1)]. **Cystiene** (0.84 ± 0.82 %) is needed to infants and children; it is synthesized from Methionine, functional component of proteins and enzymes [(1)]. Among the dispensable amino acid, **Serine** (7.62 ± 0.45 %) which most important amino acid, is useful for brain and central nervous system, memory power, brain development, releasing hormone etc (Mothet *et al*.,2000).

5. Summary and Conclusion

From the above discussions, it was observed that, a large number of amino acid and protein, which are very essential for our body, to promote our physiological activities, were identified (qualitatively and quantitatively) in *B.bengalensis*. Moreover, from the immemorial time beings, people are very conscious about the ethno-medicinal values of *B. bengalensis*. They strongly believe about the functions *B. bengalensis*, which can cure several diseases such as controlling conjunctivitis, night blindness, diarrhea, stomach upset and chronic gastric disorders, arthritis, joint pain rheumatism, cardiac diseases, controlling blood pressure, asthma, rickets, calcium metabolism, nervousness, giddiness etc. (Mahata, 2002, Pravakar & Roy,2009). The interesting points were noticed that the people who are living, beneath the poverty level of north-eastern part of our country (Chhattisgarh, Jharkhand, Bihar, West Bengal, Sikkim, Tripura, Manipur, Mizoram etc) depends directly on *B. bengalensis*, as their main earning and edible source. In this regards there are a lots of evidence, such as: (1) The South-West region of West Bengal (West Midnapore,Bankura,Purulia etc) a lots of people engage with the marketing process of *B. bengalensis* (Chakraborty *et. al*., 2013 & Chakraborty *et.al* 2014). (2) It was also investigated about the ethno-medicinal uses of *B. bengalensis* by the tribal people of Santhal Pargana, Jharkhand,India (Roy and Singh 2007). (3) The uses of *B. bengalensis* with other shell fishes, were the traditional medical practice among the people of Kosi basin of North-Bihar, Khagaria, Supaul, Saharsa, Madhepura, Purnea etc areas. They used it for curing of rickets in children, night blindness, controlling and curing of blood pressure and cardiac problem etc (Pravakar & Roy, 2009).

So, here *B. bengalensis* can, not only provide a cheap protein rich edible product but also supplements several deficiencies. Specially those poor people who can not effort to buy fishes, meats every day, can not effort to pay to doctor, medicines and also not able to reach hospital and health care centre (which are far from their living areas). For those type of rural region *B. bengalensis* is the only solution for providing protein-rich food to the people. Therefore, fisheries with *B. bengalensis* and its allied genera can be a solution to the world food problem, regarding increasing population and demand of foodstuff. And thus, there is a considerable scope for judicious utilization of this snail regarding their edible values. That is why, this resources need to be turn into commercial utilization, to generate employment opportunity and enhancement the income of the poor people. Side by side, conservation and management of this snail is necessary for the sustenance of local people also (Rao 1969, Mukherjee & Basu, 2008). Though it is just a beginning, there is a long way remaining to move on, regarding, gathering of knowledge about the ethno-medicinal, biochemical, socio-bio-economical importance etc on, *Bellamya bengalensis*.

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