WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 8.084
Volume 9, Issue 8, 2031-2041. Research Article ISSN 2277– 7105

BIOCHEMICAL (PROXIMATE AND ELEMENTAL) ANALYSIS OF FRESH WATER CRABS BARYTELPHUSA CUNICULARIS WHICH ENHANCE TO FOOD DOMAIN

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Article Received on
14 June 2020,

Revised on 04 July 2020,
Accepted on 24 July 2020

DOI: 10.20959/wjpr20208-18281

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ABSTRACT

The biochemistry of tissue of *Barytelphusa Cunicularis* freshwater crab analyzed the proximate composition and elemental content from inner part of the abdominal cavity. The focus of study was to outline chemical characteristics and edible property of *Barytelphusa Cunicularis.* On principle component analysis show the significant and non significant variants in the moisture and ash content were observed. Protein content was higher as compare to rest of the composition. Carbon content significant lower than Ca, K, and Mg respectively. Among the micro elements B, Ba, Cd, Cr, Cu, Li, Pb and V show extremely lower concentration that is traceable amount. Zn and Sr characterized higher composition. The analysis of crab data interpret on practical basis, but lack of availability of literature of *Barytelphusa Cunicularis* mean while data compare with other species to elaborated information on their macro and micro elemental composition. The study of chemical substances and vital process occurring in *Barytelphusa Cunicularis*, the chemical composition of fresh water species destined for human consumption influenced for nutritional preference according there variety and environmental condition. Constancy of product quality commercial market requires constant supply of the substantial quantity of crab for which harvestable area must be identified. Hence the objective of this study was to access chemical composition of the fresh water crab.

KEYWORD: *Biochemistry, freshwater, food domain, proximate, fingerprinting.*

1. INTRODUCTION

There are 1306 freshwater crab species in India; tropical region has maximum diversity of crab in the world.[1,2] About 389 crab species were reproduced in northern region of Maharashtra state. Godavari, Tapi, and Narmada are most elaborated river basin support the major freshwater food providing the animal's fish, crab, prawns. Along the fishes has large extent, freshwater crab also marketed in local market of those places in northern region of Maharashtra (west wood 1836). The chemical composition of *Barytelphusa Cunicularis* not reported in the literature, specific literature were not reported so compare the data with other freshwater crab species e.g. Brachyuran. Several studies have been assessed with nutritive value of meat.[3,4,5] Importance of the freshwater crab is food has good nutritive value, but this species has less importance in Maharashtra except konkan region because habitat of this species is rest of the Maharashtra. The crab has been analysed biochemically on large scale worldwide.[6] Specifically research has been perform on *Barytelphusa Cunicularis* to access the nutritive value of the tissue of the thoracic cavity the data will be provide by using multi-element fingerprinting analysis - i.e., use of elemental profile of an organism as a natural base of its origin. It is powerful technique to investigation of nutritional value of and raises the geographical traceability of the agricultural products.[10,11,12] The present study focused on advance resolution the quality of *Barytelphusa Cunicularis* marketed in market of northern regions Maharashtra.[16,19] In India rest of the coastal area Freshwater crab is not proper food, its population increases due to low marketed, only poor people's are utilized as food so there were buried at that places and they decomposes that site. Study of freshwater crab is very essential to increases advance resolution quality. Important study was multi element analysis complimentary with conversion to proximate component analysis to profile the chemical characteristic and nutritive value of *Barytelphusa Cunicularis.*

2. MATERIALS METHODS

Sampling permission was obtained from relevant institution, all applicable national or international guideline for the care and use of animal were followed. All procedures are performed with ethical standard.

2.1. Sample collection

Barytephusa Cuniclaris was collected from northern region of Maharashtra state, India. The adult species was used for extraction, isolation and proximate study because adult species has more bioactive compounds as compare to juvenile.[26,27] After collection, the species were

transfer alive in refrigerated containers to the laboratory. Crabs were then euthanized by thermal shock for 15 min. At -30^{0}c from each collected species dissect individual and collect the tissue of abdomen cavity of the crab, the crude extract was prepared from this tissue.[2] The extract was store in dip freezer and use whenever need.

2.2. Proximate analysis

The proximate compounds were analysed using reputed literature procedure.

Moisture: The tissue of the crab was oven dried at 135^{0}c and weighed; this method was repeated until constant weight was obtained. (AOAC 2019).

Ash: weighed 2 gm of sample in to porcelain crucible and placed in temperature controlled furnace preheated to 600^{0}c. Hold at this temperature 2 hours. Transferred crucible directly to dedicator, cool and weighed immediately % of ash.(AOAC 2019).

Total carbon and nitrogen: The freeze-dried tissue was on combustion by using elemental analyser (thermo scientific flash 1112).

Protein 100 mg tissue + 5 ml Distilled water + 5 ml 30% TCA → the protein precipitate → centrifuged at 3000 rpm→ discard supernent liquid + 0.1 N NaOH→ precipitate dissolved + 4ml $CuSO_4$ + 0.4 ml foline reagent→ **Blue coloration of the solution indicated presence of protein.**

2.3. Elemental analyzer

The tissue were mixed with 4 ml H_2O_2 and 6 ml HNO_3 at 180^{0}c for 10 digestion solvent system in microwave, cooled sample and diluted with double distilled water to make volume 25 ml filtered through Whitman no. 42 paper measured elements by ICP- AES (SHIMATDZU ICPE-9800 Series). Results expressed as mg 100 g^{-1} (Raab. et. al.2005).

2.4. Statistical analysis

Principal component analysis (PCA) and Permutational multivariative ANOVA (PERMANOVA) were used to multivariative data analysis and find the exact elemental composition of this species. In principle composition analysis matrices were constructed and performed fix factor *p* from PERMANOVA analysis these all values were calculated from raw data. PCA and PERMANOVA analysed were performed by using software (PAST326B.PCA Germany) were performed data of the species like proximate composition,

micro and macro elements after the assumption of the variance was checked with log and square root transformation.

3. RESULT AND DISCUSSION

Table 1: Biometric and proximate composition, mineral and elemental data from tissues of fresh water crab *Barytelphusa Cunicularis* under analysis.

Biometric		Proximate composition		Macro-elementsgm 100gm^{-1}		Micro-elements gm 100gm^{-1}	
Wet weight (gm)	302.56	Moisture	78.70%	C	8.37	B	0.07
Carapace Width (mm)	117.17	Ash	1.90%	N	3.14	Ba	0.067
-	-	Protein	16.70%	Ca	235.01	Cd	0.017
-	-	-	-	K	319.56	Cr	0.017
-	-	-	-	Fe	0.44	Cu	0.98
-	-	-	-	Mg	48.10	Li	0.006
-	-	-	-	Na	242.10	Mn	0.53
-	-	-	-	-	-	Ni	0.013
-	-	-	-	-	-	Pb	0.01
-	-	-	-	-	-	Sr	2.69
-	-	-	-	-	-	V	0.083
-	-	-	-	-	-	Zn	7.20

Significant study of biochemical constituent tissue of *Barytelphusa Cunicularis*, the results expressed on freeze-dried tissue wet weight basis. Tissue yielded abdominal cavity of freshwater crab. Protein, microelements and microelements were studied of freshwater crab. The protein content highest biochemical as compare to macro and microelements, moisture content is significant higher while ash content was lower. The proximate composition means percentage composition of three basic components such as moisture, ash, and protein (table). Protein is energy yielding nutrient in freshwater crab so, called freshwater food. The proximate composition may vary with different habitat while different seasonal condition (Bilgin S et. al. and Langer S, et. al. 2013). Macro elements and microelements also investigated tissue of the freshwater crab expressed as gm 100gm^{-1} wet weight.

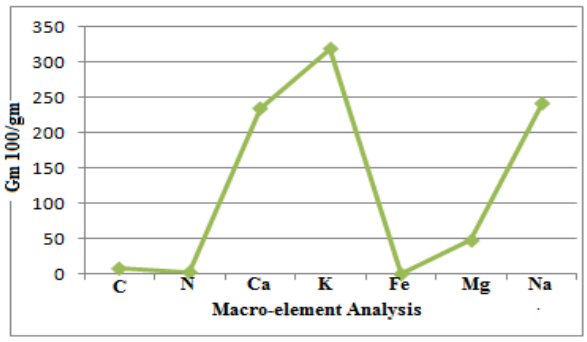

Fig. 1.

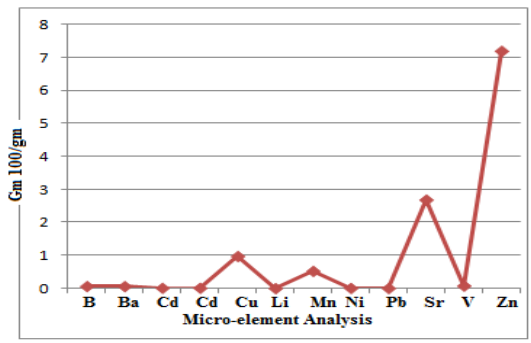

Fig. 2.

Potassium (k) has highest macro element, then sodium (Na) 242.10 has second prior calcium (ca) 235.01 third prior and magnesium (Mg) 48.10 these macro elements has highest composition, carbon (C) and Nitrogen (N) has 8.37 and 3.14 respectively less composition but ferrous (Fe) has 0.44 negligible composition. When micro-elements resulted data was observed zinc (Zn) and strontium (Sr) highest composition in freshwater crab tissue copper (Cu) and Manganese (Mn) also showed low composition while Boron (B), Barium (Ba) and Vanadium (V) has lest one but Lithium (Li), Nickel (Ni) and lead (Pb) were resulted negligible composition.

The present study concluded that the tissue of freshwater crab has good nutritional value due to protein content. The macro elements Ca, K, Mg, and Na has enriched they make vital role in living system of the human body, micro-elements like Zn and Sr most abounded, these are all biologically active elements. It means that freshwater crabs always beneficial to human life and make important role in drug design leads.

4. Principle component analysis

There are various explanatory variables in data they are correlates with other so by principle component analysis method that's components are identified from database, make standard linear method. PCA is very important tools of assessment because variable data study were large number of differences, so PCA sorted out data into groups of variable. It is also data reduction method.

4.1. One way anova test.

Test for equal means	Sum of sqrs	df	Mean square	F	p (same)
Between groups	7.286E06	3	2.42867E06	0.8778	0.4692
Within groups	5.5337E07	20	2.76685E06	-	-
Total	6.2623E07	23	0.2876	-	-

4.2. Permutation p (n=99999)

One way ANOVA test gives the value of fixed factor *p*, it was unrestricted raw data.

4.3. Components of variance (only for random effects)

Var (group) : -65809

Var (error) : 2.76685E06

ICC : -0.0243643

Omega2 : 0

Levine's test for homogeneity of variance, from means p (same): 0.01913

Levine's test, from medians p (same): 0.4698

4.4. Welch F test in the case of unequal variances

F=1.728, df = 3, p=0.3321

PC	Eigen value	% variance
1	4.89307E06	97.265
2	137166	2.7266
3	401.835	0.0079878
4	4.43117	8.8084

4.5. Score

Sr. no.	PC1	PC2	PC3	PC4
1	6787.2	-10.172	-0.50387	-0.72103
2	-1221.2	-677.28	-48.144	-0.71783
3	-1111.3	1011.6	-26.163	-0.7193
4	-947.14	-68.921	15.896	-1.0859
5	-1265.4	92.078	21.245	-0.16644
6	-1217.8	-88.62	20.441	-1.1339
7	-1024.4	-74.542	17.196	-0.58351
8	2.5557E-05	0.00010208	-0.0058507	-0.96057
9	2.5637E-05	2.5637E-05	-0.0058689	-0.96357
10	-4.5666E-05	-0.00018241	0.010454	1.7164
11	2.3694E-05	9.4644E-05	-0.0054243	-0.89057
12	-0.00016566	-0.00066169	0.037923	6.2263

4.6. Score Plot

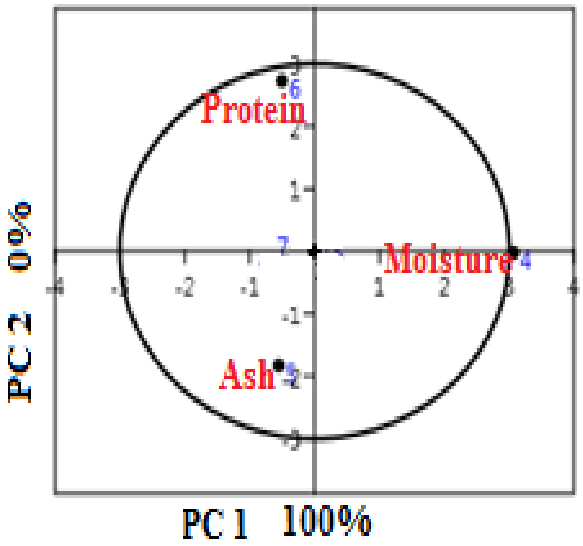

Fig. 3

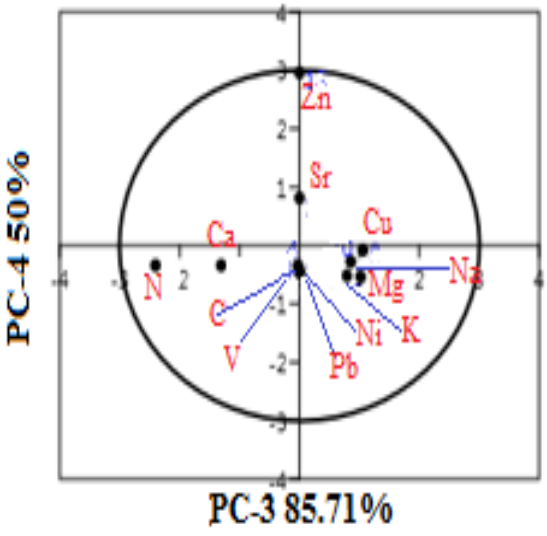

Fig. 4

Fig.3. score plot of the first two components of the principle components analysis conducted with the proximate elements are characterizing. The most significant correlation with biometric data is shown. Fig.4. score plot of the PCA of third and forth component conducted with the macro and micro elements composition data characterizing of tissue of the *Barytelphusa Cunicularis.* The most significant correlation both chemical elements are shown.

The tissue of *Barytelphusa Cunicularis data* was analysed, there were variable components, but protein has most abundant due to fact this freshwater crab species utilize to food which characterized in fig.3. When analysis of fresh water crab point to be noted that most of the difference in chemical elements that's while differs macro and micro or trace elements. The protein content was estimated using total nitrogen concentration from standard AOAC method, it is more effective method. In general the proximate parameters of the *Barytelphusa Cunicularis* observed moisture was 78.70% due to this weight of the crab is 302.56 gm for this composition study adult crab was used. In adult species protein content has most abundant (Ozogul et.al. 2013 Baklouti.et.al, 2013).

The PCA analysis performed elemental concentration of *Barytelphusa Cunicularis* major four principle components are explained first at 85.71% and second was 50% consequently *Barytelphusa Cunicularis* was indicated in PCA plot (Fig.4) it is proved by PERMANOVA test (F=1.728, p=0.3321). Further bivariance comparison indicated that significant multivariate occurs. Among, macro element significant of carbon has lower content comparison to Ca, Mg & Na respectively. In micro-elements B, Ba, Cd, Cu, Li, Ni, V & Pb has negligible, while Zn and Sr have higher content.

5. CONCLUSIONS

Here presented data was description about tissue of the *Barytelphusa Cunicularis* from abdominal cavity. The analysis focused a wide proximate composition and elemental content. Aims of this study also incorporate study of the elemental finger printing of the macro and micro elements, which provide advance biochemical screening method based on H^1NMR metabolomics and provide the standard profiling procedure towards investigation of freshwater food domains.

6. Declarations

6.1. Author contribution Statement

Sanjay V. Chavan*, Ashok A. Patil, Rajendra P. Borale** was review the literature and designs the experiments the performed, analysed and interprets the data every things was contributed like reagents, materials, analysis tool, and wrote the paper.

6.2. Funding statement

No any funding agency was supported to this research work.

6.3. Conflict of interest statement

The author declares no conflict of interest.

6.4. Additional information

No any additional information for this work.

7. Acknowledgements

This research work performed for Ph.D. work entitled "Study of natural product from freshwater crab and its biological efficiency with reference to structure activity relationship" its partial fulfilment.

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