
EVALUATION OF PROXIMATE COMPOSITION OF *CALLINECTES SAPIDUS*, *PROCAMBARUS CLARKII* AND SEDIMENT FROM QUA IBOE RIVER, NIGERIA

¹Edidiong E. Ikpe, ²Aniekan E. Akpakpan, ³Patrick Akpan, ⁴Etiowo G.Ukpong, ⁵Okon E.Okon

^{1,2}Department of Chemistry,

Akwa Ibom State University, Ikot Akpaden, Nigeria

^{3,4,5}Department of Science Technology,

Akwa Ibom State Polytechnic, Ikot Osurua, Nigeria

ABSTRACT

Proximate composition of *Callinectes sapidus* (blue crab), *procambarus clarkii* (cray fish) collected from Mkpanak area of Qua Iboe River, Ibeno local government area of Akwa Ibom State, was evaluated. Proximate composition of the samples were determined in triplicate and the mean result showed that *Callinectes sapidus* has moisture content of (6.08 ± 0.20), Ash (15.02 ± 0.03), fat (20.71 ± 0.20), crude fibre (0.419±0.10), protein (3.850 ± 0.01) and carbohydrate (46.15 ± 0.04) while *Procambarus clarkii* had a moisture content of (9.67 ± 0.0), Ash (4.40 ± 0.04), fat (15.43 ± 0.10), crude fibre (0.26 ± 0.02), Protein (2.94 ± 0.01) and carbohydrate of (67.33 ± 0.00). However, the metal analysis were determined using Atomic Absorption Spectrophotometer model, Buck scientific 210 VGP and result obtained were as follows; for *callinectes sapidus* Cd concentration was below detectable limit (BDL), Pb (10.02± 0.04), Cu (56.0± 0.04) and Ni (BDL). For *Procambarus clarkii*, Cd (10.2± 0.20), Pb (BDL), Cu (BDL) and Ni (21.04 ± 0.01) pH analysis of the samples falls within acidic region of 4.75-5.76. The value obtained during metal analysis in *Callinectes sapidus* and *Procambarus clarkii* exceeded WHO permissible limit(6.5-9.2) for aquatic food, while the metal content in sediment were within the permissible limit. Hence, the entire metal analysis revealed that Mkpanak area of Qua Iboe River is under pollution threat and underscores the need for early remediation if adverse effect is to be avoided.

KEYWORDS: *Callinectes sapidus*, *Procambarus clarkii*, proximate composition, sediment.

INTRODUCTION

Researchers on the impact of poisonous of heavy metals in aquatic ecosystems comprising of rivers, streams, lakes, aquatic organism (crab, crayfish, mollusk etc) and sediment have become a major global environmental concern. In nature aquatic organisms are constantly exposed to pollutants such as metals and hydrocarbons due to natural geochemical processes such as weathering of rock and leaching, as well as anthropogenic activity resulting from increase in urbanization, industrialization, agricultural practices, oil exploration and production activities (Vaikosen *et al.*, 2014). Mostly such increases in anthropogenic activities, usually results to accelerated deposits of chemical pollutant into the aquatic environment which poses to their proved toxicity persistence, bioaccumulation and biomagnification in the food chain (Ikpe *et al.*, 2016). Since pollutant have the tendency of accumulating in biota, it is imperative to monitor the concentration of these pollutant in the environment and to investigate bioaccumulation process in order to know the possible impact on human and health risk which man faces in such an environment.

The toxicity of poisonous metals such as Cd, Pb, and Ni to human cannot be overemphasized as individuals may become exposed to these metals. The metals are absorbed through the skin and into the blood streams where they accumulate in tissues and organs like liver and kidney causing adverse effect to man (Adewuyi *et al.*, 2011). The toxicity of heavy metals varies from one heavy metal to another and in the same element may have different toxic effects depending on its chemical form and its speciation (Kakulu, 1985, WHO 1991, Aremu,1998). This study is aim at assessing the level of some poisonous metal and proximate composition of fauna and sediment along Mkpanak area of Qua Iboe river, Ibeno local government area in order to ascertain its potential health benefit based on the level of proximate composition and the level of pollution of

the river based on the level of heavy metal concentration, thus providing a cleaner and healthier environment.

MATERIALS AND METHODS

SAMPLING LOCATION AND PHYSIOGRAPHY

Mkpanak is a village along the Qua Iboe River in Ibeno Local Government Area of Akwa Ibom State. Ibeno is located within latitude $4^{\circ}30' - 40^{\circ} 45'N$ and longitude of $7^{\circ}30' - 8^{\circ}45'E$ on the South-Eastern coastline of Nigeria. It is one of the largest petroleum oil producing communities in Nigeria. Ibeno River is about 150 km long and empties into the Atlantic Ocean.

According to marine and fishery resources of Nigeria (MFRN) (2016), it occupied the largest Atlantic coastline of more than 129 KM, in Akwa Ibom State. However, Mkpanak (Qua Iboe River) is close proximity to Exxon-Mobil oil effluent treatment and discharge plant. The area is made up of mangrove swamp forest; it has rainfall throughout the year with its peak between May and September (MFRN, 2016). Moreover, the prime occupation of the people in the area is fishing.

COLLECTION AND PREPARATION OF SAMPLES

(a) Sediment

Three sampling location were chosen within Mkpanak downstream of Qua Iboe river at an interval of 400 metre and with specific co-ordinate as started in Table 1 below. The sampling was done in the month of June 2017. However, representative samples were taken off in triplicate into a black polythene bag to prevent photo-oxidation and contamination. The samples were taken to the laboratory and were preserved in the refrigerator at a temperature of $0-4^{\circ}C$. The samples (sediment) were air dried; grind and digested using nitric acid (HNO_3) and perchloric acid $HClO_4$ with the ratio of 2.1. Finally they were analyzed using Atomic Absorption Spectrophotometer Buck Scientific model 210 VGP (Variable Giant Pulse) method according to AOAC (1995).

Table 1: Sample Location, Geographical Coordinate and Site description

Site	Site code	Coordinate	Site description
1	A	N05 ⁰ 02 ⁰ 07.11" E007 ⁰ 58' 08.0"	NW
2	B	N04 ⁰ 33' 25.2" E008 ⁰ 00' 08.04"	SE
3	C	N04 ⁰ 33' 24.7" E008 ⁰ 00' 00.5"	NS

(b) Fauna

The blue crab (*Callinectes sapidus*) and crayfish (*Procambarus clarkii*) were collected from Qua Iboe along Mkpanak area at different points using fishing net with the assistance of fishermen. The samples were collected in a black polythene bag into an icepack cover and conferred to the laboratory for analysis using standard analytical methods as described by A.O.A.C. (1995).

CHARACTERIZATION OF SAMPLES

DETERMINATION OF pH AND TEMPERATURE

The negative logarithmic of the hydrogen ion concentration of the samples were determined with the aid of *Thomas Scientific metre, TS675* which has been previously calibrated using buffer solutions, according to standard analytical methods reported by Okieme *et al*, 2015.

Temperature of the samples were determined by dipping the temperature probe into the samples for few seconds during when the pH was investigated with the aid of dual purpose pH /temperature metre. Results were recorded in degree Celsius ($^{\circ}\text{C}$).

PROXIMATE ANALYSIS

Proximate analyses of the samples were carried in triplicate after which the mean value was calculated. The procedures are stated below:

MOISTURE CONTENT DETERMINATION

Moisture is determined by the loss in weight that occurs when a sample is dried to a constant weight in an oven. About 1g of the samples was weight into preheated evaporating dish separately. The samples were dried in an oven at 105°C for 3 hours, cool in a desiccator and weighed. The drying and weighing was done until a constant weight was achieved

$$\% \text{ moisture} = \frac{\text{weight of sample} + \text{evaporating dish} - \text{weight of evap. Dish after drying}}{\text{Weight of sample taken}} \times 100$$

CRUDE FIBRE

The crude fibre content of the samples was determined by weighing 2g of the samples into 100ml conical flash. 200ml of 5% H_2SO_4 was added, boiled for 30 minutes by adding H_2O to a constant volume. It was filtered through a whatman filter paper using a vacuum pump. The filtrate was washed with hot water (distilled) to neutrality. Later tested with blue litmus paper (for acid) and the filtered were washed back into the flask. 200ml of 5% NaOH was added and boiled for 30 minutes.

It was filtered and washed with hot (distilled) water to neutrality. Samples were rinsed with ethyl alcohol and wash from filter paper into a known weight of evaporating dish using ethyl alcohol. Samples were dried to a constant weight after an hour and were taken to muffle furnace for ashing at 500°C for three hours. It was transferred to a desiccator to cool and the weight of the ash samples was taken.

$$\% \text{ crude fibre} = \frac{\text{Weight of sample} + \text{Weight of dish} - \text{Weight of dish} + \text{ash}}{\text{Weigh of samples}}$$

LIPID (FAT) CONTENT DETERMINATION

1g of the dry ground samples were wrapped separately in a whatmann filter paper and placed inside the thimble in a soxhlet extractor, the apparatus was connected to a round bottom flash containing 250ml petroleum ether. A complete set-up was obtained by connecting the flask to a reflux condenser and allowed to reflux for about 6 hours. Rotary evaporator was used to recover the remaining solvent. The oil obtained was weighed and the percentage determined as shown below.

$$\% \text{ Lipid} = \frac{\text{Weight of oil}}{\text{Weight of sample}} \times 100$$

ASH CONTENT DETERMINATION

Ash is the inorganic residue obtained by burning off the organic matter of sample at $400 - 600^{\circ}\text{C}$ in muffle furnace for four hours. 1g of the sample was weighed into a preheated crucible. The crucible was placed into a muffle furnace at 500°C for 3 hours until a whitish grey was obtained. The crucible was placed in a desiccator to cool before weighing.

$$\% \text{ Ash} = \frac{\text{Weight of crucible} + \text{ash} - \text{Weight of crucible}}{\text{Weight of sample}} \times 100$$

CRUDE PROTEIN

Crude protein was determined by measuring the nitrogen content of the sample and multiplying it by a factor of 6.25. This factor is based on the fact that most protein contains 16% nitrogen.

Crude protein was determined by modified kjeldahl method as reported by Ikpe and Akpabio, (2013). The method involves digestion, distillation and Titration

DIGESTION: 1g of the samples was weighed into the Kjeldahl flask. Well clamp to a retort stand under a regulated hot plate in the fumes cupboard. HNO₃ and HCl were introduced in the ratio of 1:3 respectively. The flask was heated slowly, later increased until 1/3 of the digest remains. The digest was diluted with deionized H₂O, filtered and made-up to make of 100ml standard volumetric flask.

Calculation

$$\% \text{ Nitrogen (N)} = \frac{(S-B) \times M \times 14 \times 100}{\text{Weight of sample (w)} \times 100 \times V}$$

Where

S	=	Average titre value
B	=	Blank titre value
M	=	Molarity or Normality of HCl
14	=	Atomic weight of Nitrogen
D	=	Total volume of Digest
100	=	Percentage conversion
W	=	Weight of sample
100	=	Conversion to dm ³
V	=	Volume taken for distillation

The crude protein was calculated as: 6.25 x N

DETERMINATION OF CARBOHYDRATE

Dutcher *et al* method was used as reported by Enyeribe *et al*, 2014. The total amount of carbohydrate in the sample was obtained by calculation using percentage weight difference, where it involves subtracting the percentage sum of the food nutrients, % crude protein, % crude lipid, % crude fibre, and % Ash from 100% dry weight.

Percentage carbohydrate was calculated using equation:

$$\text{Carbohydrate \%} = 100 - (\text{crude protein} + \text{crude lipid} + \text{crude fibre} + \text{Ash}).$$

RESULTS AND DISCUSSIONS

Table 2: The result of the proximate analysis

S/n	Components	<i>Callinectes sapidus</i> (blue crab) (%)	<i>Procambarus clarkii</i> (crayfish) (%)
1	Moisture	6.08 ± 0.20	9.6 ± 0.02
2	Ash	15.02 ± 0.03	4.40 ± 0.05
3	Fat	20.71 ± 0.20	15.43 ± 0.22
4	Crude fibre	0.419 ± 0.10	0.26 ± 0.20
5	Protein	3.850 ± 0.01	2.94 ± 0.21
6	Carbohydrate (CHO)	46.15 ± 0.04	67.33 ± 0.02

Table 3 : Results of some heavy metal content

Biological name	Common name	Cadmium (mg/kg)	Lead (mg/kg)	Copper (mg/kg)	Nickel (mg/kg)
<i>Callinectes sapidus</i>	Blue crab	0.00 ± 0.00	10.02 ± 0.04	56.01 ± 0.04	BDL
<i>Procambarus</i>	Crayfish	10.02 ± 0.20	10.30 ± 0.10	36.10 ± 0.01	21.04 ± 0.00

<i>clarkii</i>					
sediment	-	BDL	BDL	BDL	12.31 ± 0.01
WHO (mg/kg)	-	0.01	0.30	0.05	-

BDL = below detectable limit

DISCUSSION

PROXIMATE ANALYSIS

The results in (Table 2) revealed that *Callinectes sapidus* and *Procambarus clarkii* contains moisture $6.08 \pm 0.20\%$ and $9.67 \pm 0.02\%$ respectively. From the result *Procambarus clarkii* has a higher moisture content which suggested that it should be properly dried before storage so as to avoid the invasion of micro-organism which can lead to spoilage (Kirchmann and Kirchmann 1996). The ash content was also determined in both samples which *Callinectes sapidus* recorded a higher value of $15.02 \pm 0.03\%$ and *Procambarus clarkii* $4.40 \pm 0.04\%$, the result showed that they contained some quality of mineral element which are essential in our diet (Onwuka, 2005). However, Ash content represents the index of mineral elements present in a sample. Its amount is useful in assessing a sample and gives an idea of the minerals present in a sample (Michael and David, 2002).

Consequently, *Callinectes sapidus* has a higher value of crude fat of $20.71 \pm 0.20\%$ compared to $15.43 \pm 0.01\%$ of *Procambarus clarkii*. Lipid can provide a very good source of energy and aids in transport of fat soluble vitamins, and protects internal tissues; lipid also contributes to important cells processes (Joes *et al*, 1985 and Pamela *et al*, 2005).

Crude fibre was also determined in the samples; the results revealed that *Callinectes sapidus* had $419 \pm 0.10\%$ and *Procambarus clarkii* 0.26 ± 0.02 . The crude fibre composition was determined because of the recent interest in the potential role of dietary fibre in human nutrition. Fibre helps to maintain the health of gastrointestinal tracts, in excess may bind tract elements, leading to deficiencies of iron and zinc. Moreso, fibres lower the body cholesterol lever and consequently decrease cardiovascular disease (Gwarzo *et al.*, 2014). Also the percentage of the crude fibre present in the samples suggested that consumption could help maintain movement of food through the gut to provide energy and ensure break down of the food. Moreover, it showed that the samples contained some mineral elements which are essential for animal's nutrition (Ajiwe *et al.*, 2008). In general, fibre appears to inhabit many diseases, like colon cancer by binding the carcinogens and preventing them from entering the body while they pass through the system (Uchegbu *et al.*, 2012). Fibre cleans the digestive system flushing the residue as efficiently and quickly as possible (Jean, 1996).

The protein content of the *Callinectes sapidus* was high then *Procambarus clarkii* by 0.91%, both samples showed that they are highly proteinous and could be incorporated in the diet of both aged and young including pregnant and nursing mothers.

Procambarus clarkii had a 21.19% increase in carbohydrate content than *Callinectes sampidus*. High carbohydrate value indicated that the sample contained reasonable amount of energy and will give high amount of energy when consume.

The percentage of carbohydrate in the samples is an indication that it can be used to regulate various metabolic processes in the body, as key molecules in the central metabolic pathways of the body.

Carbohydrate also serves as stores forms of energy as glycogen in liver and muscles. It also provides major source of energy and responsible for breaking-down fatty acids and preventing ketosis, as reported by Gwarzo *et al.*, (2014).

HEAVY METAL CONTENT

Cadmium (Cd)

The highest cadmium concentration among the three samples under study was detected in *Procambarus clarkii* with 10.20 ± 0.20 (mg/kg), this might be attributed to bioaccumulation; within its habitat as well as migration to highly Cd polluted area before the sampling was done. The sediment recorded no result it might be as the result of cleaning exercise that was carried out two year ago by the operating oil company in the area, it could also affect the negligible concentration detected in *Callinectes sapidus* because of its sedentary lifestyle. The presence of high concentration of Cd in *Procambarus clarkii* could be attributed to atmospheric deposition from natural or anthropogenic sources as well as from run-off/flooding washing of cadmium containing fertilizer, cadmium-nickel battery and sewage sludge into the river (ATSDR,1997).

Cadmium when ingested by humans, it accumulates in the intestine, liver and kidney (Malviya and Wagela, 2001). The health effects of chronic exposure of Cd include proximal tubular disease and Osteomalacia. Long term exposure to cadmium is associates with renal dysfunction. Cadmium is bio persistence and once absorbed remains resident for many years. High exposure can lead to obstructive lung diseases and has been linked to lungs cancer (Dauda and Odoh, 2014). Cadmium may also cause bone defect in humans and animals. The average daily intake for humans is estimated as 0.15 from air μg from air and $1\mu\text{g}$ from water (Jarup *et al.*, 1998).

Maximum limit of $0.2 \mu\text{g/g}$ Cd in plant and $5.0 \mu\text{g/g}$ Pb in plant was prescribed by WHO/FAO (WHO and FAO, 2007). The values of the standard compared to this research work indicate Cd pollution in *Procambarus clarkii*.

However, following the recommended maximum standard of 0.01mg/kg by WHO (1990), it means that *Procambarus clarkii* is polluted with cadmium, and not recommended for frequent consumption. Also, dwellers of Mkpanak that consume the sample are liable to suffer from aliment attributed to cadmium such as severe kidney, liver damage as well as bone weakness in the near future due to bioaccumulation.

Lead (Pb)

The level of Pb concentration in the faunas under study is high this could be increased by the volume of boat traffic in the area, also Pb compounds form basis of anticorrosive and primer as reported by Adewauyi *et al.*, (2011). However, bioaccumulation could also lead to increase value of lead in the samples. The sediment recorded a negligible value of 0.00 ± 0.00 and these could attributed to the cleaning activities of the operating oil company in the area. Moreso, it is envisage that metal construction work, iron bending and welding of metal for the boat making within the area could cause high lead content of $10.30 \pm 0.10\text{mg/kg}$ and $10.02 \pm 0.04\text{mg/kg}$ *Procambarus clarkii* and *Callinectes sapidus* respectively.

Nickel (Ni)

The concentration of Nickel (Ni) in sediment was $12.31 \pm 0.01\text{mg/kg}$ while that of *Procambarus clarkii* increase tremendously with value of $21.04 \pm 0.00 \text{mg/kg}$ that might be attributed to bioaccumulation. The level of nickel in *Callinectes Sapidus* was negligible with 0.00 ± 0.00 . This could be linked to Rhizoremediation, because *callinectes sapidus* is sedentary, and this enhances its opportunity to feed on the roots of some mangrove swamp floras such as *Rhizophora mangle* which is prevalent in the area of study.

However, from the statistical analysis on co-efficient of variation of Ni, among the sample signifies stability in terms of variability. Moreso, Ni content in *Procambarus clarkii* and Sediment exceeded the tolerance level of 0.5mg/l for drilling water set by WHO (1990), therefore there is need for public concern because of the detrimental effect of Ni to human and plant by which inhibits photosynthesis (NCBI, 2014).

It's noted that frequent dumping of refuse such as nickel battery, and other nickel containing substance by dwellers and oil companies in the river might cause the prevalence of Ni in the sediment and *Procambarus clarkii* within the area under study.

Copper (Cu)

According to Kukul (1985) Cu is known to be one of the active ingredients in antifouling composition. From the result of the study, it is observed that samples under study such as *callinectes sapidus*, *procambarus clarkii* showed a high level of Cu with the range of 56.01 ± 0.01 mg/kg respectively which could emanates from the operating oil company and individuals, while the sediment recorded BDL (Below detectable limit) which might be attributed to the cleaning exercise conducted by the operating oil company during oil spillage two year ago, because of bioaccumulation Cu still remains in the faunas. From the statistical point of view Cu is stable with a coefficient at variation of 0.13%. Meanwhile if Cu in the study sample compared with FAC (2007) standard of 0.2mg/kg, it proved that the samples are above the permissible unit for aquatic food.

CONCLUSION

This research study has ascertained the quality of fauna and sediment in Mkpanak area of Qua Iboe River for human consumption and aquaculture. It has created awareness on the impact of heavy metals on aquatic environment (biota) through anthropogenic activities.

Thus, providing a healthier and cleaner environment. Therefore, I recommend that further study be conducted during dry season for comparative analysis.

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