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Hydrocarbon Levels in the Organs of Fish and Shellfish off the South-Eastern Atlantic Coastline of Nigeria

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Abstract

Observations during a 12-month assessment of bioaccumulation potential of hydrocarbons by marine organisms using gravimetric and Gas Chromatographic techniques indicate a possible influence of climate change with regards to the ingestion of residual and volatile organics in organs of marine organisms. Along the southeastern Atlantic coastline of Nigeria, seasonal variability in total Hydrocarbon (THC), low molecular weight (LMW) (C6 – C12) and high molecular weight (HMW) (C14 – C32) aliphatic hydrocarbons (AH) occurred in the tissue, stomach, liver, gonads and gills of three fish species; *Galeoides decadactylus*, *Pseudotolithus elongatus*, *Pseudotolithus typus* and tissues of two crab species *Callinectes amnicola* and *Cronius ruber* samples collected from the nearshore zone. The highest THC concentration of 33828.56 ± 629.71 $\mu\text{g}/\text{kg}$ wwt was obtained in the tissue of *P. typus*, while the least value of 4142.35 ± 77.10 $\mu\text{g}/\text{kg}$ wwt was obtained in the gills of *G. decadactylus* during the wet season. Enhanced THC values occurred in the dry season over the wet season in gonads (47637.10 ± 886.75 $\mu\text{g}/\text{kg}$ wwt) and gills (4142.35 ± 77.10 $\mu\text{g}/\text{kg}$ wwt) of *G. decadactylus*. Of the two crab species, *C. amnicola* had higher values ($21,401.00 \pm 398.38$ $\mu\text{g}/\text{kg}$ wwt and $27,615.02 \pm 514.05$ $\mu\text{g}/\text{kg}$ wwt) respectively in both dry and wet seasons. The C6 – C12 dominated the various organs in the wet season than the C14 – C32 hydrocarbons during the period. Extreme temperatures in the tropical ocean generated low volatile hydrocarbons from point and fluvial sources which subsequently were concentrated in marine organisms. This infers volatile organics and methane especially in marshy areas should be under surveillance for the purpose of determining the level of organism contamination in the nearshore zone of Nigeria.

Keywords: Hydrocarbon levels, marine organisms, southeast Atlantic coast, Nigeria.

Introduction

Hydrocarbons are organic compounds composed of carbon and hydrogen atoms arranged in varying structural configurations. At a simple level, they may be divided into two families: aliphatics and aromatics (WHO, 2005). Marine organisms are capable of accumulating hydrocarbons in their body sometimes above the threshold or permissible limits. Achudume (2009) studied the impact of oil spill and concluded that Niger Delta are important breeding ground for marine organisms and continuous absence of fish and other aquatic organisms was a result of polluted water. Ibeno marine waters, southeastern Nigeria, are susceptible to hydrocarbon contamination due to exploration activities of crude oil in its vicinity. Marine organisms there have the tendency to take up hydrocarbon, which might eventually be cycled through the food web. Olajide *et al.* (2009) reported 0.172 ppm concentration of crude oil in the mouth of *Pseudotolithus sp.*, while Mironov *et al.* (1981) reported hydrocarbon contents of 12–90 mg/kg wwt in fishes and 7–200 mg/kg wwt in benthic animals.

The levels at which hydrocarbons are concentrated in marine organisms are controlled by certain factors. Ng and Gray (2011) and Armitage *et al.* (2011) link high temperature of marine waters to high concentration potential of chemicals in marine organisms. Yuan *et al.* (2008) and Zhang and Tao (2008) reported that where temperature is higher, the higher molecular weight hydrocarbons peak because the lower molecular weights are more volatile. To further buttress this point, Eisler (1987) observed that the more soluble molecular weight PAHs had a shorter half-life. Macdonald *et al.* (2003) observed that during warmer seasons, consumption rates increase. This causes an increase in uptake of the chemicals with food. Therefore, resulting increases or decreases in environmental concentrations could be compounded by the seasonal variability in uptake and loss rates.

Against this backdrop, this paper attempts to document the level of hydrocarbons in organs of marine organisms off the southeastern coast of Nigeria. It is assumed that marine organisms take up available hydrocarbon in water and sediment which are eventually concentrated in their various organs. Also concentration higher than the threshold could pose deleterious health effect on the organisms and on man who depends on seafood for consumption. This finding therefore would help in understanding the changes that may occur ecologically within the marine ecosystem. It would also establish the background levels and composition of hydrocarbons in marine fishes prior to further degradation of the region.

Materials and Methods

The study area is located on the southeastern segment of the Nigerian coast which is within the equatorial region. It is located within latitudes $04^{\circ}3'$ and $04^{\circ}31'N$ and longitudes $07^{\circ}56'$ and $08^{\circ}00'E$. The region is tropical, having an annual rainfall of about 14 inches (355.6 mm) which supports a mangrove system dominated by *Nypa fruticans*, wild oil palms and salt resistant woody shrubs.

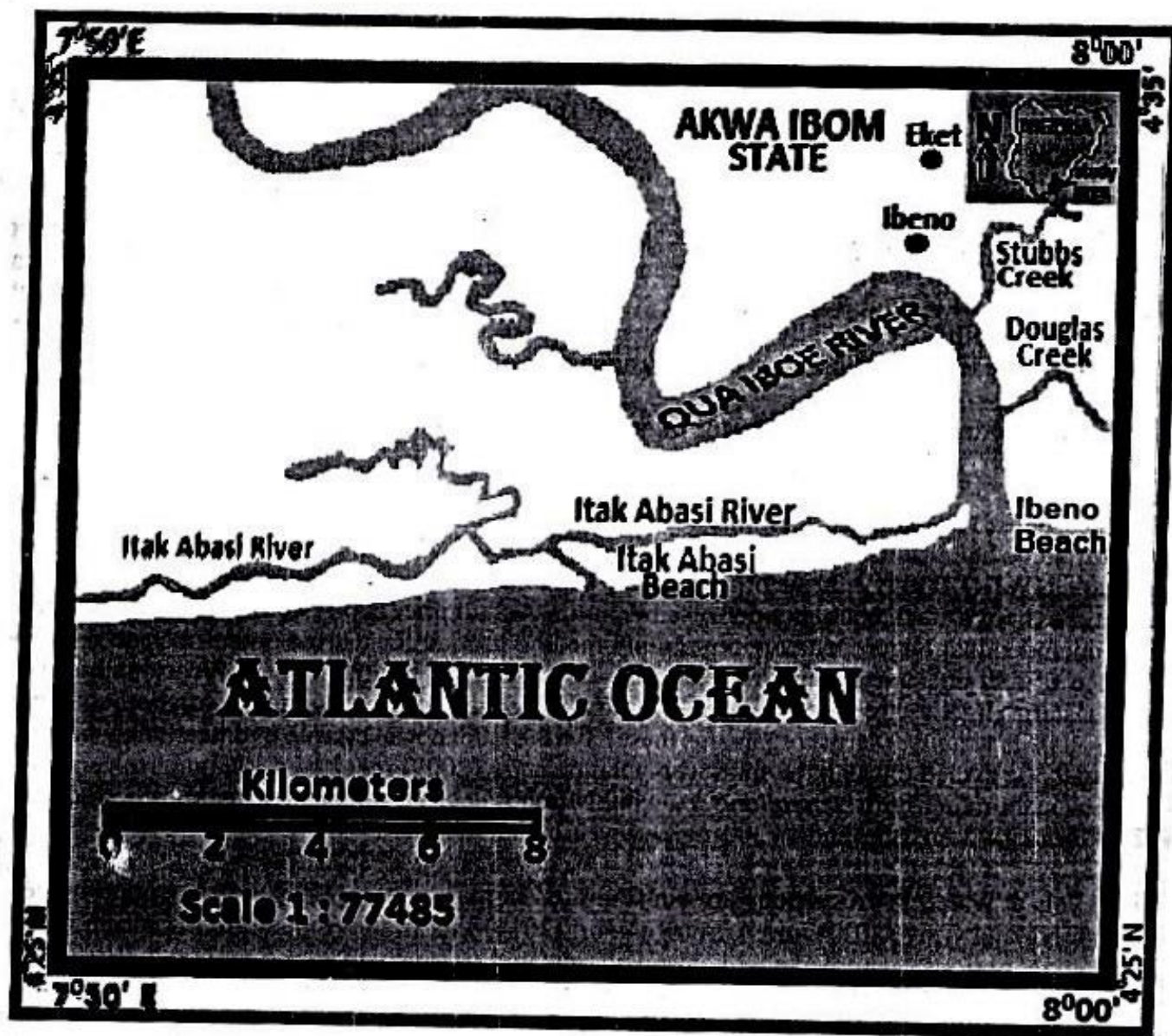


Fig. 1. Map of study area.

The Ibéno marine waters are bordered shore-wards with a gently sloping beach profile with variations in morphology. It is a high energy area which generates waves whose breaker heights are usually 30-200 cm (Antia, 1989). Wind velocities and longshore currents range between 5-10 m/s and 0.15-0.9 m/s respectively (Antia, 1989). Inhabitants of the area are mainly the Ibéno and Eket speaking people of Akwa Ibom State, with a major occupation of fishing, lumbering, farming and thatch making. Oil exploration with offshore and onshore activities at the estuary mouth in Mkpának is a major activity in the area.

Fresh samples of 3 fish species and 2 crab species, *Galeoides decadactylus* (12-14 cm), *Pseudotolithus typus* (18-20 cm) and *Pseudotolithus elongatus* (12-14 cm), *Cronius ruber* (3-4 cm) and *Callinectes amnicola* (3-5 cm) were obtained monthly from the local fishermen for twelve months (January - December 2006), totaling 108 fish specimens and 72 crab specimens. The organs of each of the fish specimens; tissue, liver, gonad, stomach, gills and tissue of the crab specimens were removed, crushed and homogenized in order to get an average representative sample.

Three grams wet weight of the crushed samples were each put in a flask with 20 ml of alcoholic methanol and 5 ml brine solution and saponified and then refluxed on a heating sand bath and condenser to digest the samples. Subsequently, 15 ml of hexane was added to the digested sample in a separating funnel and extracted by shaking vigorously and left to separate the hydrocarbon component from the other components of sample. The hydrocarbon component was left to evaporate to near dryness and then the hydrocarbon content was determined gravimetrically by a Voltcraft Plus Ps 20 weighing balance.

The pretreatment of samples proceeded as follows: 1 cm of moderately packed glass wool was placed at the bottom of 10 mm, I.D x 250 mm long Chromatographic Column. Slurry of 2 g activated silica gel in 10 ml methylene chloride was prepared and placed into the Chromatographic Column. To the top of the column was added 0.5 cm of sodium sulfate. The column was rinsed with additional 10 ml of Methylene Chloride.

The column was pre-eluted with 20 ml of Pentane. This was allowed to flow through the column at a rate of about 2 minutes until the liquid in the column was just above the sulfate layer. Immediately, 1 ml of the extracted sample was transferred into the column, the extracted bottle was rinsed with 1 ml of Pentane and added to the column as well. The stop-cork of the column was opened and the eluent was collected with 10 ml graduated cylinder. Just prior to exposure of the sodium sulfate layer to air, Pentane was added to the column in 1-2 ml increments. Accurately, measured volume of 8-10 ml of eluent was collected and labeled "aliphatic". The aliphatic fraction was concentrated to 1 ml for Gas chromatography (GC) Analysis.

Gas Chromatography (GC) Analysis proceeded as follows: The concentrated aliphatic fractions were transferred into labeled glass vials with Teflon Rubber Crimp caps for GC analysis. 1 µl of each of the concentrated samples were rapidly injected by means of hypodermic syringe through a rubber septum into the column. The sample injection port, column and detector were usually kept somewhat warmer than the column to promote rapid vapourization of the injected sample and prevent sample condensation in the detector.

Separation occurs as the vapour constituent partition between the gas and liquid phases. The sample was automatically detected as it emerges from the column (with a constant flow rate) with FID (Flame Injector Detector) whose response is dependent upon the composition of the vapour by measuring the retention time, which was minutes between sample injection time and Chromatographic peak recording.

Results and Discussion

Table 1 and Fig. 2 show concentration and seasonal trend of total hydrocarbon in fish organs and tissues of crabs collected from Ibéno marine waters.

Tables 1. Concentrations of THC in fish organs and crab tissues.

DRY SEASON					
Species	Tissue ($\mu\text{g}/\text{kg}$ wwt) \pm SD	Liver ($\mu\text{g}/\text{kg}$ wwt) \pm SD	Gonad ($\mu\text{g}/\text{kg}$ wwt) \pm SD	Stomach ($\mu\text{g}/\text{kg}$ wwt) \pm SD	Gills ($\mu\text{g}/\text{kg}$ wwt) \pm SD
<i>Galeoides decadactylus</i>	5522.80 \pm 102.81	18640.60 \pm 346.98	47637.10 \pm 886.75	12427.07 \pm 231.32	4142.35 \pm 77.10
<i>Pseudotolithus typus</i>	18641.00 \pm 346.99	44184.44 \pm 822.48	34519.29 \pm 642.56	12530.62 \pm 233.25	6550.10 \pm 121.92
<i>Pseudotolithus elongatus</i>	10010.00 \pm 186.33	20021.04 \pm 372.68	23472.66 \pm 436.93	9320.30 \pm 173.49	5867.64 \pm 109.22
<i>Callinectes amnicola</i>	21401.00 \pm 398.38				
<i>Cronilus ruber</i>	9665.15 \pm 179.91				
WET SEASON					
Species	Tissue ($\mu\text{g}/\text{kg}$ wwt) \pm SD	Liver ($\mu\text{g}/\text{kg}$ wwt) \pm SD	Gonad ($\mu\text{g}/\text{kg}$ wwt) \pm SD	Stomach ($\mu\text{g}/\text{kg}$ wwt) \pm SD	Gills ($\mu\text{g}/\text{kg}$ wwt) \pm SD
<i>Galeoides decadactylus</i>	5867.65 \pm 109.22	27270.17 \pm 507.62	30032.08 \pm 559.03	12427.07 \pm 726.09	4142.35 \pm 77.10
<i>Pseudotolithus typus</i>	33828.56 \pm 629.71	27615.02 \pm 514.04	33828.55 \pm 629.70	12530.62 \pm 257.02	6550.10 \pm 141.35
<i>Pseudotolithus elongatus</i>	16223.54 \pm 302.00	20021.04 \pm 372.68	34519.29 \pm 642.56	9320.30 \pm 244.16	5867.64 \pm 128.50
<i>Callinectes amnicola</i>	27615.02 \pm 514.05				
<i>Cronilus ruber</i>	11045.59 \pm 205.61				

In the dry season *G. decadactylus* had THC value of 5522.80 \pm 102.81 $\mu\text{g}/\text{kg}$ wwt in its tissues, while in the wet season, a concentration of 5867.65 \pm 109.22 $\mu\text{g}/\text{kg}$ wwt was obtained. In the liver, THC concentration was 18640.60 \pm 346.98 $\mu\text{g}/\text{kg}$ wwt, but higher value of 27270.17 \pm 507.62 $\mu\text{g}/\text{kg}$ wwt occurred in the wet season. For both seasons, stomach concentrations were 12427.07 \pm 231.32 $\mu\text{g}/\text{kg}$ wwt (dry season), 12427.07 \pm 726.09 $\mu\text{g}/\text{kg}$ wwt (wet season). In the gills, dry and wet season concentrations were similar with the value of 4142.35 \pm 77.10 $\mu\text{g}/\text{kg}$ wwt. THC concentration in gonad was 47637.10 \pm 886.75 $\mu\text{g}/\text{kg}$ wwt (dry season), with concentration of 30032.08 \pm 559.03 $\mu\text{g}/\text{kg}$ wwt in wet season. The trend of accumulation in this species increases from gills to tissue to stomach to liver to gonads in both seasons.

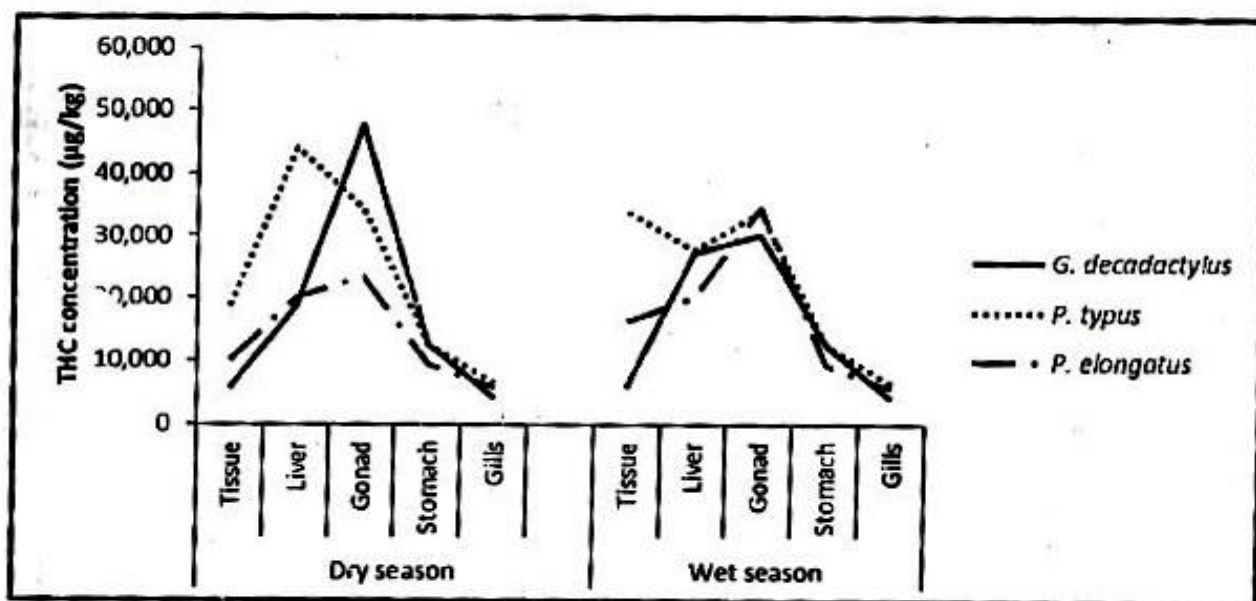


Fig. 2. Variations of THC in organs of the fish species in dry and wet season

THC concentration of 18641.00 ± 346.99 $\mu\text{g}/\text{kg}$ wwt was observed in the tissue of *P. typus* in dry season while in the wet season it was 33828.56 ± 629.71 $\mu\text{g}/\text{kg}$ wwt. In *P. typus*, liver had THC concentration of 44184.44 ± 822.48 $\mu\text{g}/\text{kg}$ wwt (dry season) and lower value of 27615.02 ± 514.04 $\mu\text{g}/\text{kg}$ wwt (wet season). THC in gonad was 34519.29 ± 642.56 $\mu\text{g}/\text{kg}$ wwt (dry season) and lower value of 33828.55 ± 629.70 $\mu\text{g}/\text{kg}$ wwt (wet season). THC concentration in stomach was similar (12530.62 ± 233.25 $\mu\text{g}/\text{kg}$ wwt, 12530.62 ± 257.02 $\mu\text{g}/\text{kg}$ wwt) in dry and wet seasons respectively.

THC concentration of 10010.00 ± 186.33 $\mu\text{g}/\text{kg}$ wwt obtained in dry season was lower than 16223.54 ± 302.00 $\mu\text{g}/\text{kg}$ wwt obtained during the wet season in tissue of *P. elongatus*. In the liver, 20021.04 ± 372.68 $\mu\text{g}/\text{kg}$ wwt was noted for both dry and wet seasons. In the gonads, THC was 23472.66 ± 436.93 $\mu\text{g}/\text{kg}$ wwt in dry season while 34519.29 ± 642.56 $\mu\text{g}/\text{kg}$ wwt was recorded for the wet season. In the stomach, 9320.30 ± 173.49 $\mu\text{g}/\text{kg}$ wwt was noted for dry season while 9320.30 ± 244.16 $\mu\text{g}/\text{kg}$ wwt was noted for the wet season. In the gills, for both dry and wet seasons, 5867.64 ± 109.22 $\mu\text{g}/\text{kg}$ wwt and 5867.64 ± 128.50 $\mu\text{g}/\text{kg}$ wwt were noted respectively.

In all fish species, highest concentrations were in the gonads while least concentrations were in the gills. This could have been due to the presence of relatively small amount of lipids in the gills and due to constant flushing of water through the gills during respiration. Therefore the gonad had the highest concentration than the other organs.

For dry and wet seasons, *C. amnicola* had 21401.00 ± 398.38 $\mu\text{g}/\text{kg}$ wwt and 27615.02 ± 514.05 $\mu\text{g}/\text{kg}$ wwt respectively in its tissues. Wet season uptake was higher than dry season uptake, which can be attributed to wet season runoffs into coastal sediments, high lipid content in crab tissues and interaction with HC particles.

For dry and wet season, *C. ruber* had 9665.15 ± 179.91 $\mu\text{g}/\text{kg}$ wwt and 11045.59 ± 205.61 $\mu\text{g}/\text{kg}$ wwt respectively in its tissues. This lower concentration compared to *C. amnicola*, was probably because of less ability to uptake THC from sediments, strong influence of physical process such as waves, water washing and gravity affect the settling of HC unto sediments, and therefore only a little would be available for uptake.

The relationships between water temperature and concentration of THC in tissues of marine fish and shellfish are presented in Table 2 and Fig. 3 below.

Table 2. Relationship between water temperature and concentration of THC in tissues of studied organisms.

Months	Water temp. (°C)	<i>G. decadactylus</i>	<i>P. typus</i>	<i>P. elongatus</i>	<i>C. amnicola</i>	<i>C. ruber</i>
January	31.21	5837.685	33655.81	10058.48	27474.01	11099.08
February	31.53	5955.023	34675.62	16302.1	28026.23	10880.39
March	31.06	6014.573	33992.37	15980.89	26932.66	11210.07
April	24.24	5440.193	18545.42	9762.66	21505.12	9711.957
May	24.48	5549.541	18180	9958.889	21937.38	9520.593
June	24.72	5661.087	18730.87	9860.287	20872.66	9615.799
July	24.97	5494.595	18361.8	10260.65	21292.2	9907.168
August	25.22	5605.037	18918.18	10159.06	21081.39	9426.33
September	25.47	5386.33	19107.36	16140.7	21720.17	9809.077
October	30.3	5722.66	33322.59	16465.12	27748.75	10989.19
November	30.6	5896.062	34332.29	15822.66	27201.99	11322.17
December	30.9	5779.887	32992.66	16629.77	28306.5	10772.66

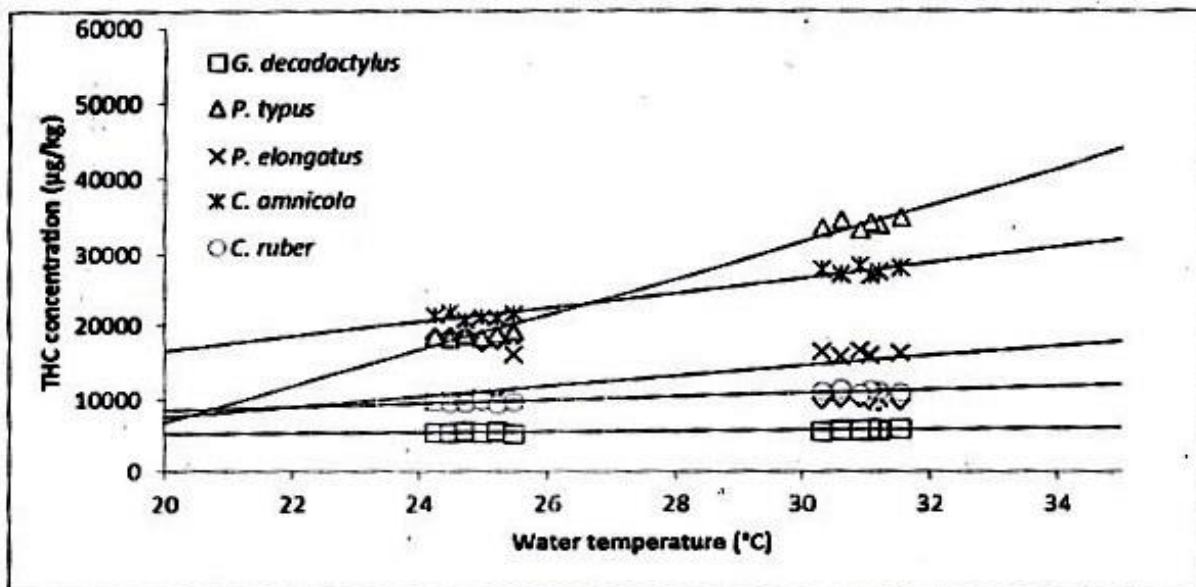


Fig. 3. A plot of water temperature against THC concentration in marine organisms.

Trend line (Fig. 3) equations of regression and positive r-values (Table 3) show that THC concentration in tissue of marine organism is directly related to the temperature of marine water. This means that the THC values were higher in the tissues of marine organisms during the dry season when temperature of marine waters is at extreme and vice versa. This is in line with the reports of Ng and Gray (2011) and Armitage *et al.* (2011) that link high temperature of marine water to high concentration potential of chemicals in marine organisms.

Table 3. Regression of THC in marine organisms on water temperature.

Marine organisms	Equation of regression	Correlation coefficient, r^2
<i>G. decadactylus</i>	$y = 56.67x + 4114.6$	0.7736
<i>P. typus</i>	$y = 2462.9x - 42459$	0.9872
<i>P. elongatus</i>	$y = 696.63x - 6313.4$	0.4711
<i>C. amnicola</i>	$y = 1003.3x - 3476.1$	0.9649
<i>C. ruber</i>	$y = 222.43x + 4151.5$	0.9183

Table 4 and Fig. 4 show the concentration of aliphatic hydrocarbons in tissues of marine fish and shellfish during dry and wet seasons.

Table 4. Concentration of aliphatic hydrocarbons in tissues of studied organisms.

Aliphatic HC	---	Wet Season ($\mu\text{g}/\text{kg} \pm \text{SD}$)
C6	18.01 \pm 13.95	359.82 \pm 394.22
C7	16.80 \pm 18.42	1040.55 \pm 19.36
C8	70.31 \pm 1.30	871.75 \pm 16.22
C9	36.49 \pm 28.27	1702.41 \pm 31.68
C10	22.27 \pm 34.51	288.71 \pm 5.37
C11	105.37 \pm 1.96	466.77 \pm 8.68
C12	125.77 \pm 2.34	702.27 \pm 13.07
C14	146.86 \pm 2.73	208.70 \pm 3.88
C16	250.03 \pm 4.65	224.06 \pm 4.17
C18	85.23 \pm 1.58	7.51 \pm 11.64
C24	508.71 \pm 9.46	19.22 \pm 0.35
C28	29.23 \pm 0.54	11.64 \pm 0.21
C32	22.93 \pm 17.76	40.16 \pm 0.74

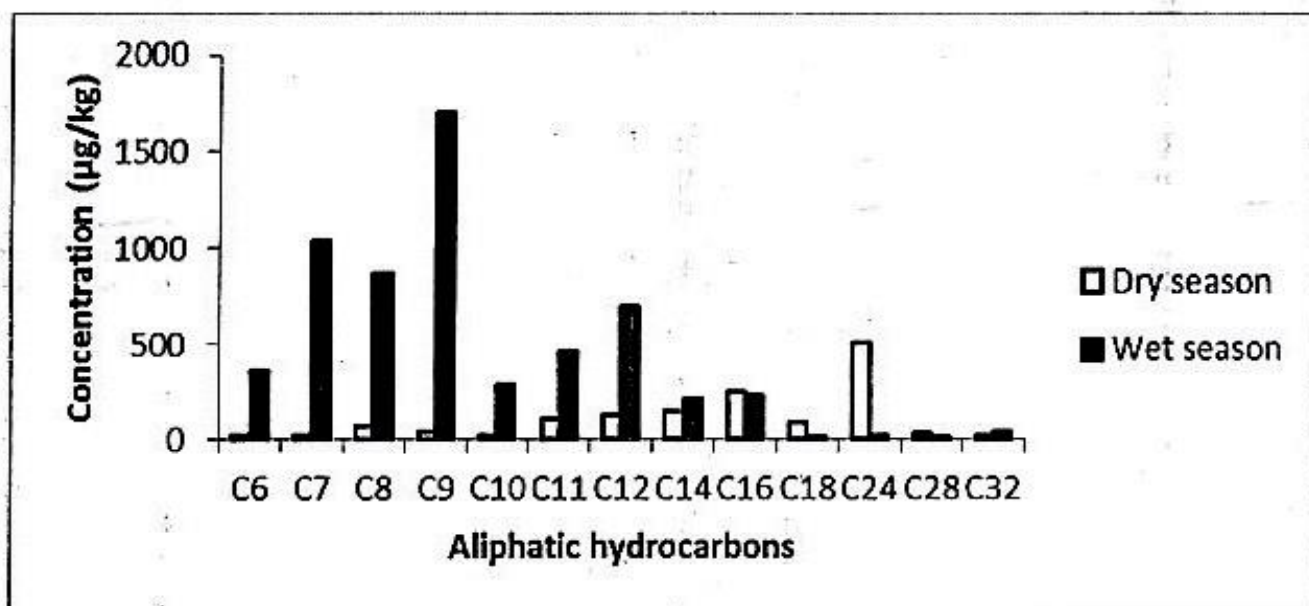


Fig. 4. Concentration of aliphatic HC in the tissues of studied fish and shellfish.

The straight chains analyzed for, ranged from $C_6 - C_{40}$. From figure 4, low molecular weight aliphatics dominated in the wet season, while high molecular aliphatics dominated the dry season. However, the low molecular peaks in wet season were more conspicuous than the high molecular peaks in the dry season.

In the wet season, C_9 had highest concentration of $1702.41 \pm 31.68 \mu\text{g/l}$ and C_{28} had least concentration of $11.64 \pm 0.21 \mu\text{g/l}$. The lower aliphatics were enriched in the surface water probably due to a decrease in temperature, which is conducive for degradation of high molecular weight aliphatics, hence the low molecular weight aliphatics were more available, less volatile due to temperature reduction, and more ready for incorporation. This could be a reason for higher concentrations for the aliphatics in the wet season in all the species which have shown the trend.

In the dry season, the highest peak was observed for C_{24} , to be $508.71 \pm 9.46 \mu\text{g/l}$. The least peak was observed for C_7 , to be $16.80 \pm 18.42 \mu\text{g/l}$. There is a reversal in the HC proportion over the seasons. Paired sample statistics showed significant difference and weak correlation between data collected during the dry and wet seasons ($t=0.047$; $r=-0.32$) ($p=0.05$). In the dry season, the higher molecular aliphatics reflected the highest peaks. The lower molecular aliphatics on the other hand, had the highest peaks in the wet season. This is also due to temperature effect in the dry season. Where temperature is higher, the higher molecular weight HC peak because the lower molecular weights are more volatile. This finding is in line with Yuan *et al.* (2008) and Zhang and Tao (2008). This was also noted by Eisler (1987) when he observed that the more soluble molecular weight PAHs had a shorter half-life. Highest n-alkane percentage was 34.96 for C_{24} followed by 17.18 for C_{16} . Least percentage was 1.15 for C_7 .

Conclusion

Hydrocarbons originate from organic matter and are found in the ambient atmosphere, land and in the marine environment. The lower molecular weight hydrocarbons are volatile and their existence in the marine environment depends on low temperature which favours their retention. The higher molecular weights are less volatile and less biodegradable. This observed variation in temperature determines the nature of hydrocarbon present in marine organisms at a particular period of time. In a period where temperatures are at extreme, low volatile hydrocarbons concentrate more than their high volatile counterparts.

Realizing that the environment is slowly and steadily degraded due to human activities, systematic monitoring should be ensured by government through relevant agencies and departments in order to determine, in quantitative terms, the rate of variation. Such information becomes important for early warnings against natural disasters of flooding, erosion and drought among others which could cause bio-amplification of chemical pollutants against their background levels. Also several greenhouse gases should be under surveillance for the purpose of determining the level of atmospheric pollution in the country. For sustainable development, there is need for the use of technologies that enable the consumption of less energy for an equal amount of services offered. The petrochemical industries should develop technology for the geologic capture and sequestration of CO₂ in order to reduce CO₂ emissions to the atmosphere.

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