RESEARCH ARTICLES

Petroleum hydrocarbons contamination of sediments and accumulation in *Tympanotonus fuscatus* var. *radula* from the Qua Iboe Mangrove Ecosystem, Nigeria

Nsikak U. Benson[1],* and Joseph P. Essien[2]

[1]Department of Chemistry, College of Science and Technology, Covenant University, P.M.B. 1023, Ota, Nigeria and Department of Atmospheric and Oceanic Sciences, McKinley Ocean Biogeochemistry Research Group, University of Wisconsin, Madison, WI 53706, USA
[2]Department of Microbiology, Faculty of Science, University of Uyo, P.M.B. 1017, Uyo, Nigeria

The pollution of coastal ecosystems as a result of petroleum-related activities is increasing, and the literature on occurrence, levels and seasonal dynamics of petroleum hydrocarbons in sediments and biota from the impacted areas of the Niger Delta, Nigeria, is practically scanty. This study was set out to provide information on the status of contamination by petroleum hydrocarbons in *Tympanotonus fuscatus* var. *radula* and sediments from Qua Iboe Estuary, Nigeria. Analyses for total petroleum hydrocarbons (TPHs) were performed on *Tympanotonus fuscatus* var. *radula* and sediment extracts using gas chromatograph with flame ionization detector (GC-FID). Seventy-two samples each of mangrove epipellic (intertidal) and benthic (subtidal) sediments, and *T. fuscatus* var. *radula* were analysed monthly between June 2003 and February 2004, covering peak periods of the wet and dry seasons. Results revealed that the TPHs level in sediments and *T. fuscatus* were highly variable. It ranged between 18.01 ± 0.04 and 210.23 ± 1.18 µg g^{-1} dry wt of epipellic sediment, 5.00 ± 0.82 and 232.00 ± 3.23 µg g^{-1} dry wt of benthic sediment and 9.40 ± 1.0 and 23.27 ± 1.0 µg g^{-1} dry wt of *T. fuscatus*. Summary continuous descriptives and correlation analyses revealed that TPH levels of the epipellic and benthic sediments showed a significant relation ($r = 0.54$, CI = 0.18–0.78), and correlated with levels in *T. fuscatus*. The overall levels of TPHs in the Qua Iboe Estuary when compared to similar ecosystems with substantial industrial and domestic coastal activities worldwide, revealed a moderate to high level of mineral hydrocarbon pollution.

Keywords: Estuary, petroleum hydrocarbons, sediment, *Tympanotonus fuscatus*.

IN recent times, contamination of the environment by petroleum hydrocarbons is potentially widespread because modern society uses so many petroleum-based products (for example, gasoline, kerosene, fuel oil, mineral oil and asphalt). Hydrocarbons are quantitatively the most important constituents of petroleum, and arise from natural as well as anthropogenic sources[1,2]. Human-mediated sources of petroleum hydrocarbons include offshore oil production, marine transportation, atmospheric or aerial depositions from combustion of coal and gas flaring, direct ocean dumping, coastal, municipal and industrial wastes, and run-off[3]. However, among the anthropogenic sources, point discharges, contamination by urban run-offs, refineries and other coastal effluents are in aggregate substantial and are important in causing local, chronic pollution in the vicinity of estuaries, creeks, harbours and coastal settlements[4].

Studies of the accidental and intentional releases of petroleum-based products to the aquatic environment indicate that aquatic organisms are able to bioaccumulate some total petroleum hydrocarbon (TPH) fractions, particularly polycyclic aromatic hydrocarbons[5,6]. Petroleum hydrocarbons are among the most common contaminants bound to estuarine sediments[2,6,7]. These substances, mostly of organic origin, may be deleterious to aquatic plants and animals. Where the spilled petroleum washes up onto beaches or shorelines, there may be short-term damage to fish and wildlife, as well as impacts to recreational use of shoreline or riparian areas for human swimming or fishing. Some heavier petroleum fractions are capable of accumulating in environmental substrates. This can lead to stress on bentho-pelagic organisms[8]. The effects of hydrocarbon-laden sediments on flatfish tissue and the fish itself, particularly the juvenile stage have been reported by Moles and Norcoss[9].

Sediment contamination in estuaries needs to be properly and fully assessed. Therefore, in assessing and monitoring aquatic environments for hydrocarbons, it becomes necessary to adopt biological[10] and sediments assays[2,11]. Recent studies have revealed the use of biological indicators such as algae[12,13], nearshore sediments, bivalves and fish[14–18]. Aquatic assessment using the shellfish *Tympanotonus fuscatus* is rare, and considering its general consumption in the Niger Delta region, Nigeria, investigation

*For correspondence. (e-mail: nbenson@covenantuniversity.com)

using it as an indicator of pollution is warranted. *T. fuscatus* routinely filter large quantities of sea water in their feeding processes and as a result may harbour pathogenic bacteria, viruses and accumulate hydrocarbons and dioxins, which are readily taken up by marine fauna, through ingested sediment particles, food and absorption across the biomembranes[18]. Recent studies have reported incidents of oil spills from offshore production platforms of oil companies located in the study area[6,12,19], which resulted in enhanced levels of petroleum hydrocarbons in seafoods and sediments[6,20].

The present study provides reference information on the levels of TPH in sediments and a commercially benthic organism, *T. fuscatus* var. *radula* (periwinkle) obtained from the ecosystem. It has been designed to provide the baseline data required for formulation of regulations regarding managed discharges, and for optimizing environmental assessment and monitoring programme for the estuary system as well as the coastal ecozone of the Niger Delta.

Materials and methods

Study area

The Qua Iboe Estuary is a mesotidal palustrine ecotone of the parent river, Qua Iboe River. It is a major and commercially important hydrographic feature in the Niger Delta. The estuary is characterized by fine psammitic beaches, fringed with tidal mudflats and mangrove swamps. It lies within lat. 4°30′–4°45′N and long. 7°30′–8°45′E on the southeastern coastline of Nigeria (Figure 1). Like other estuaries located in the region, the Qua Iboe Estuary experiences great fluctuations in salinity between the wet and dry seasons[21]. The estuary with its associated creeks constitutes a rich assemblage of fluvial ecohydrological biotopes, dominated mainly by vast intertidal mangroves and Nypa forested wetlands. The estuary is under the constant influence of petroleum exploration and exploitation activities, and the use of icthyocides by the local fishermen. Qua Iboe Light crude oil is produced from numerous offshore fields in the Bight of Bonny, which is brought to the shore through seabed pipeline system to the Qua Iboe Terminal. The terminal (4°20′N, 7°59′E) is located on the eastern side of the Qua Iboe Estuary and contains nine crude oil storage tanks, with a total capacity of 4.5 million barrels (m bbls). Vessels are loaded from this tank farm.

The estuary area and parts of the coastal fronts of the Niger Delta are densely populated and have witnessed substantial increase in industrial and agricultural activities in the past four decades, resulting in direct discharge of organic/inorganic substances (including crude oil and refined petroleum products) into the open waters through effluents, sabotage to oil installation facilities and
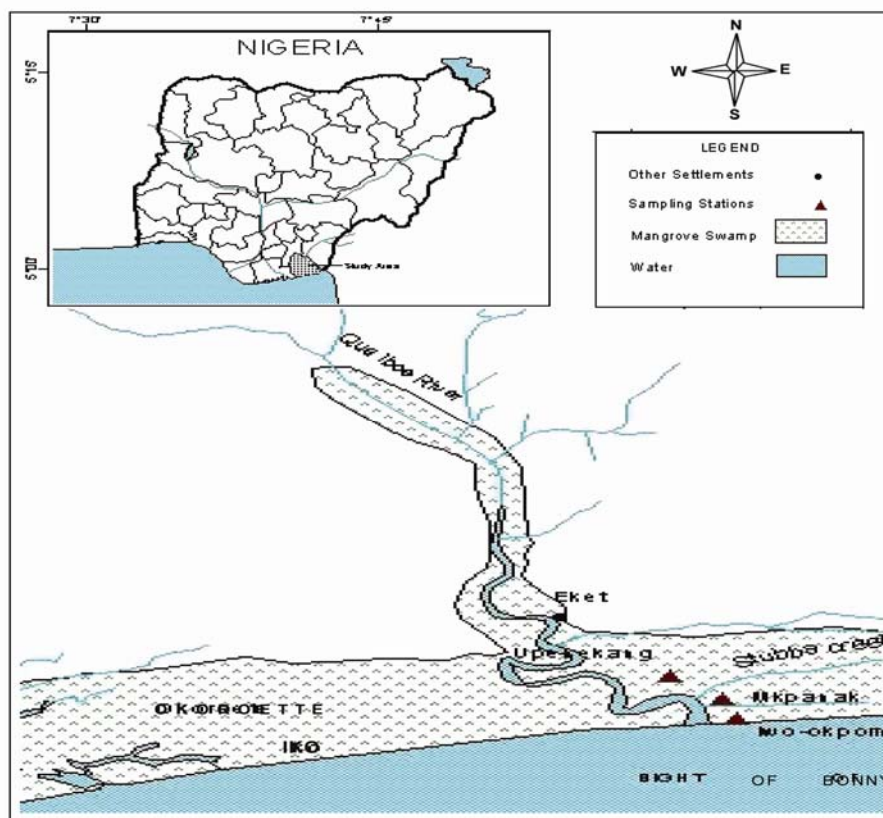

Figure 1. Qua Iboe Estuary (Niger Delta) with sampling locations at Iwo-Okpom, Mkpanak and Upenekang. (Inset) Map of Nigeria showing the study area.

operational failures. In the present study, three stations were selected within the Qua Iboe Estuary – Iwo-Okpom (IWM), Mkpanak (MPK) and Upenekang (UKG). A total of three sub-stations were selected within each station. The estuary is surrounded by sparse mangrove vegetation, with dense vegetation dominating only the MPK station. In addition, a large number of small tributary creeks flow into the MPK and UKG stations. The stations were selected due to concerns raised by the local residents, and the local and state governments over incessant oil spills, illegal dumping of untreated wastewaters from on- and offshore oil facilities, storm-water pollution, and other activities from the upstream reaches of the ecosystem. During this study, a case of spillage occurred on 22 November 2003 during which an unquantified amount of crude oil from a leakage tank farm facility spilled into the environment[6,12].

Sampling strategy

Sediments and *T. fuscatus* var. *radula* (West African mud creeper) samples were collected monthly between June and September 2003 (wet season), and November 2003 and February 2004 (dry season) from the designated stations. The sediments (intertidal and subtidal) and bio-specimens were collected at the same time from the same locations for better comparison of sediment contamination with uptake by *T. fuscatus* var. *radula*.

Sediment sampling and treatment

Intertidal sediment samples were collected using a 0.065 m diameter corer to a depth of 0.1 m. Subtidal samples were obtained using a modified Van veen grab sampler (0.1 m^2). Sediment samples were stored in clean, well-labelled glass bottles at 4°C until further analyses[22]. In the laboratory, sediment samples were oven-dried in petri dishes at 80°C for two days, ground into powder and sieved using a 2 mm mesh sieve to remove coarse substances. The sieved samples were crushed and triplicates homogenized to obtain the respective composites. Precisely, 50.0 g of each homogenized sediment sample was weighed and spiked with an internal standard (C_{32}-alkane)[23].

Biospecimen sampling and treatments

Samples of *T. fuscatus* var. *radula* of variable lengths (35–98 mm) were randomly collected by local fishermen within the designated stations. Shellfish which are common in the area were collected and wrapped in sterile aluminium foil, and immediately stored in ice-packed coolers before being taken to the laboratory for pre-treatment and analysis. Fleshy tissues of each *T. fuscatus* var. *radula*

were removed from the shell with the help of a clean, sterile, steel pin. Prior to analysis, 1 g of each sample was homogenized in a mortar with 2 g Na_2SO_4 and the homogenate was transferred to a column for extraction.

Determination of total petroleum hydrocarbons

Samples of *T. fuscatus* var. *radula*, intertidal and subtidal sediments collected for TPH analyses were serially extracted with 100 ml methyl isobutyl ketone, Analar grade and the extracts were allowed to settle. Each extract was centrifuged for 5 min and decanted. The volumes of the supernatant were reduced to about 5 ml over a rotary evaporator maintained at 20°C. For the determination of total hydrocarbon content, a gas chromatograph with flame ionization detector was employed, using 1 µl aliquot of each extract. The total peaks obtained were converted to weight using hydrocarbon standard calibration[23]. Duplicates and method blanks were treated identically using the same reagents to test for the precision, accuracy and reagent purity used in the analytical procedures.

Statistical analysis

Correlation analyses, Box-and-Whiskers plots, comparative and continuous summary descriptives of data were performed using Analyse-It + General 1.73 statistical software, with level of significance maintained at 95% for each test. ANOVA and linear regression of data were performed using Statgraphics Centurion VI software, with level of significance maintained at 95% for each test.

Results and discussion

TPH concentrations in sediments and biota

The monthly concentrations of TPH in intertidal and benthic sediments, and in *T. fuscatus* are summarized in Table 1. In the intertidal (epipellic) sediment, the seasonal concentrations ranged between 18.01 ± 0.04 and 172.00 ± 0.02 mg kg^{-1} dry wt during the wet season, while a range 37.74 ± 0.20–210.23 ± 1.18 mg kg^{-1} dry wt was obtained during the dry season (Table 1). Benthic sediment samples, however, had lower values, especially during the wet season, ranging between 5.00 ± 0.82 and 28.12 ± 1.10 mg kg^{-1} dry wt (Table 1).

The Box–and–Whiskers plots for levels of TPH in epipellic and benthic sediments (Figure 2) demonstrate the distribution of the recorded values and graphically illustrate the central location and dispersion of the concentrations. Each single, horizontal box plot shows the parametric (notched lines) and non-parametric (notched box and whiskers) statistics for the respective samples. It may be noted that there was an outlier in the levels of

Table 1. Total hydrocarbon concentrations in sediment and *T. fuscatus* samples from Qua Iboe Estuary during the wet and dry seasons (data represent mean ± SD of three determinations)

	Month	Station	EPS (mg/kg dry wt)	BTS (mg/kg dry wt)	TFS (mg/kg dry wt)
Wet season					
		IWM	20.21 ± 0.01	10.00 ± 3.40	10.17 ± 0.9
	Jun. '03	MPK	25.11 ± 0.01	5.00 ± 0.82	10.31 ± 1.0
		UKG	18.01 ± 0.04	28.12 ± 1.10	10.27 ± 1.0
		IWM	40.00 ± 0.01	17.00 ± 4.50	10.34 ± 1.0
	Jul. '03	MPK	35.00 ± 0.12	8.52 ± 1.62	10.13 ± 1.0
		UKG	55.00 ± 0.03	21.33 ± 2.00	11.19 ± 0.6
		IWM	41.21 ± 1.10	16.82 ± 0.78	9.40 ± 1.0
	Aug. '03	MPK	39.26 ± 0.10	17.30 ± 2.44	9.72 ± 1.0
		UKG	172.00 ± 0.02	17.24 ± 1.70	10.00 ± 1.0
		IWM	42.34 ± 0.02	16.12 ± 5.11	10.20 ± 1.0
	Sep. '03	MPK	31.24 ± 0.01	8.32 ± 1.37	9.99 ± 1.0
		UKG	51.82 ± 0.04	21.37 ± 3.52	9.26 ± 1.0
Dry season		IWM	54.71 ± 0.10	20.42 ± 1.00	10.41 ± 1.0
	Nov. '03	MPK	52.11 ± 0.33	47.01 ± 3.02	9.56 ± 0.9
		UKG	37.74 ± 0.20	43.71 ± 2.16	9.49 ± 1.0
		IWM	151.24 ± 0.04	93.24 ± 0.70	13.70 ± 1.4
	Dec. '03	MPK	163.24 ± 0.07	92.36 ± 0.60	20.02 ± 1.0
		UKG	210.23 ± 1.18	80.44 ± 1.00	13.21 ± 1.0
		IWM	104.21 ± 1.00	210.11 ± 0.04	18.66 ± 1.2
	Jan. '04	MPK	131.21 ± 0.60	180.14 ± 0.07	15.58 ± 1.0
		UKG	170.21 ± 2.20	198.21 ± 0.20	22.45 ± 1.0
		IWM	98.30 ± 0.91	232.00 ± 3.11	16.29 ± 1.0
	Feb. '04	MPK	111.02 ± 3.32	192.11 ± 1.02	23.27 ± 1.0
		UKG	98.31 ± 0.01	201.11 ± 5.70	14.80 ± 1.7

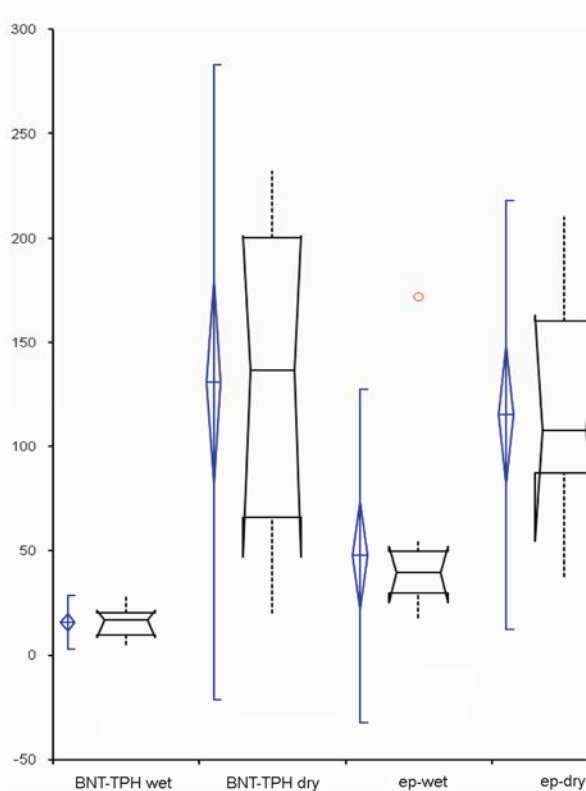EPS, Epipellic sediments; BTS, Benthic sediments; TFS, *Tympanotonus fuscatus*.

Figure 2. Multiple Box-and-Whiskers diagram for total hydrocarbons determined in sediment during the wet and dry seasons from Qua Iboe Estuary. BNT, Benthic; ep, Epipellic.

TPH in the epipellic sediment during the wet season. It is therefore suggestive that the anomalous TPH levels observed might have arisen from intermittent discharges to the estuary of crude oil and land-derived wastewaters[4]. In general, sediments are classified into three classes: low, moderate and high, with respect to their hydrocarbon content[24]. Based on this classification, the concentration of TPH in subtidal and intertidal sediments of Qua Iboe Estuary revealed a moderate to high pollution status. However, these comparatively high levels appear to be local and episodic.

Sediments of the estuary are mainly psammitic and a positive correlation was observed between the proportion of fine-grained sediment material and TPH. The dry season witnessed increased levels in TPH (20.42 ± 1.00–232.00 ± 3.23 mg kg^{-1}), which could have been influenced by tide, particle-size distribution, bioturbation and anthropogenic inputs, especially the 22 November 2003 oil spillage in the area. Generally, it was observed that the levels of TPH in the intertidal sediment were relatively higher than the values recorded for the benthic sediment. Intertidal and subtidal sediments are long-term repositories for hydrocarbons released into the environment[20,25], and expectedly, high concentrations of petroleum hydrocarbons are found in sediments of estuaries, harbours, bays and in coastal areas receiving industrial and urban discharges[4,14,15]. Mineral hydrocarbons are known to exhibit high affinity for fine-grained particles

found in sediments[25]. This explains why intertidal sediments with finer grain-sized particles generally accumulate enhanced levels of hydrocarbons than the coarse subtidal sediments.

The hydrocarbon concentrations in intertidal and subtidal sediments also exhibited marked seasonality trend with higher values observed for intertidal sediment, especially during the wet season. This may be attributed to increase in anthropogenic inputs such as surface run-offs, biogenic as well as petrogenic sources. The high levels of TPH detected in the estuarine ecosystems during the dry season coincided with the 22 November 2003 crude oil spillage from the facility of an oil company located within the vicinity of the study area. The impact characterized by a sharp increase in hydrocarbon level was immediately observed in the intertidal sediment (December 2003), but much later in the subtidal sediment (February 2004) of the estuarine mangrove ecosystem.

Elevated concentrations of TPH have been reported for sediments exposed to oil spills. However, concentrations of petroleum hydrocarbons greater than about 1 mg g^{-1} dry sediment have been reported to cause significant amphipod mortality[15], alterations in detoxification enzymes in fishes[26] and tissue abnormalities[27]. The enhanced levels of TPH obtained from sediments of Qua Iboe Estuary are comparatively and sufficiently high to attract chronic effects on aquatic organisms. It is also suggestive of the closeness of the study locations to anthropogenic sources of oil pollution and land-based activities. Similar finding has also been previously reported[25].

Hydrocarbons adsorbed in sediments can be accumulated by animals living in or on the sea-bed and thus re-enter the food chain[6,18]. Filter-feeding organisms and sediment-dwellers are capable of accumulating high concentrations of hydrocarbon residues both from oil spills[28–30], and chronic discharges to rivers, seas and oceans (e.g. of industrial discharges, sewage or run-off from roads). The TPH concentrations recorded for the biospecimen (*T. fuscatus*) ranged between 9.40 ± 1.0 and 11.19 ± 0.6 mg kg^{-1} dry wt during the wet season, while a range 9.49 ± 1.0–23.27 ± 1.0 mg kg^{-1} dry wt was obtained during the dry season. The TPH levels were remarkably low during the wet season compared to the dry season (Table 1). This variation is suggestive of increased intake of hydrocarbon residues from sediments whose levels reached a seasonal high of 22.45 ± 1.0; 23.27 ± 1.0 and 18.66 ± 1.2 mg kg^{-1} dry wt at Upenekang, Mkpanak and Iwo-Okpom locations respectively. The statistical descriptives of the THP concentrations are summarized in Table 2.

Correlation between TPH levels in sediments and *Tympanotonous fuscatus* var. *radula*

The Pearson correlation between TPH concentration in epipellic and benthic sediments of the Qua Iboe Estuary during the wet and dry seasons indicated a positively insignificant ($P > 0.05$) relation between levels of TPH in epipellic and benthic sediments ($r = 0.54$, 95%, CI = 0.18 to 0.78). The percentage distribution of TPH in sediments as reflected by their coefficients of determinations was 29.67. More so, seasonal Pearson product moment correlation between TPH levels of epipellic and benthic sediments indicated a statistically significant ($P = 0.05$) non-zero correlation ($r = 0.56$) at 95.0% confidence level, with P-value of 0.0045 (Table 3, Figure 3).

The Pearson correlation between TPH levels in tidal (epipellic) sediment and biospecimens indicated a significant ($P > 0.05$) positive relation ($r = 0.63$, $P = 0.0009$), with a corresponding coefficient of determination of 40.12%. Moreover, a linear regression between TPH levels in epipellic sediment and *T. fuscatus* revealed a fitting linear model (Figure 4 a) which described the relationship as:

$$\text{Epipellic sediment (TPH conc)} = -26.4809 + 8.3953 \times \\ T. fuscatus \text{ (TPH conc)}.$$

In order to further ascertain the fact that petroleum hydrocarbons found in *T. fuscatus* were from petrogenic sources likely derived from the tidal sediment, the P-value calculated from ANOVA was less than 0.05, thereby indicating that there is a statistically significant relationship between TPH levels in epipellic sediment and *T. fuscatus* at 95.0% confidence level. Also, the R-Squared statistic indicated that the model as fitted explains 40.12% of the variability in epipellic sediment. Similarly, a moderately strong and statistically significant correlation ($r = 0.84$; Table 3; Figure 3) was recorded for TPH concentration in *T. fuscatus* and subtidal (benthic) sediment. A linear regression between TPH levels in benthic sediment and
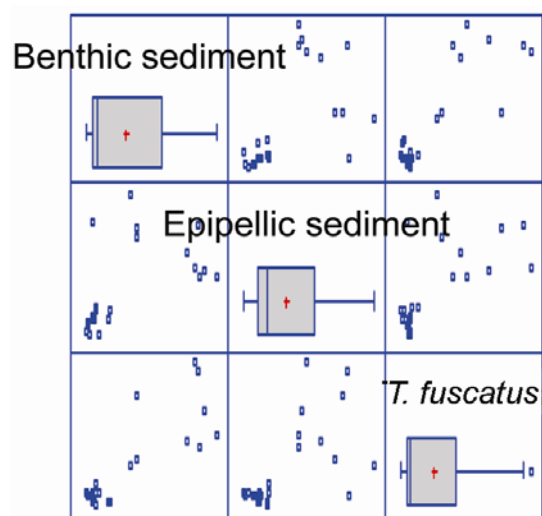
Figure 3. Matrix plots for pairs of total petroleum hydrocarbon concentrations in *T. fuscatus* and sediments.

Table 2. Seasonal continuous summary descriptives of total petroleum hydrocarbon (TPH) in epipellic and benthic sediments of Qua Iboe Estuary mangrove swamp

	Wet season		Dry season	
	EPS	BTS	EPS	BTS
Arithmetic mean	47.59	15.60	115.22	130.90
95% CI of mean	21.68–73.51	11.42-19.78	81.89–148.55	81.55–180.25
Median	39.61	16.97	107.62	136.68
96.1% CI of median	25.11–51.82	8.52-21.33	54.71–163.24	47.01–201.11
Range	153.99	23.00	172.49	211.58
Standard deviation	40.79	6.58	52.48	77.67
Standard error	11.78	1.89	15.14	22.42
Coefficient of variation (%)	86.00	42.00	46.00	59.00
Lower quartile	29.71	9.63	87.41	66.02
Upper quartile	49.45	20.33	107.62	200.39
Interquartile range	19.74	10.70	72.83	134.36
Kurtosis	9.69	–0.27	–0.64	–1.93
Skewness	2.99	0.07	0.18	–0.11
K-SP coefficient	1.28	0.73	0.46	0.88
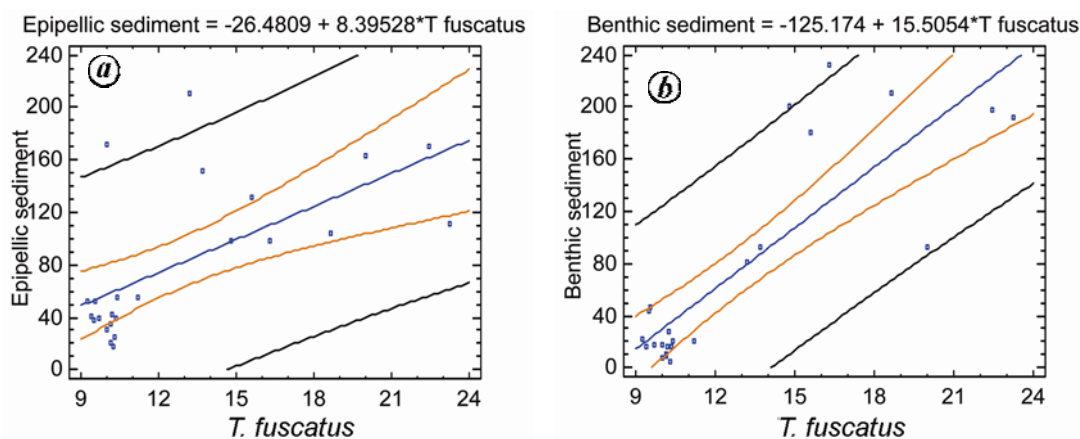
Figure 4 *a*, *b*. Plots of linear fitted model between TPH concentrations in *T. fuscatus* and sediments.

Table 3. Correlation matrix for TPH levels in *T. fuscatus* and sediment giving values of *r*-statistics for pairs of samples

	Benthic sediment	Epipellic sediment	*T. fuscatus*
Benthic sediment		0.5593 (24) 0.0045	0.8412 (24) 0.0000
Epipellic sediment	0.5593 (24) 0.0045		0.6334 (24) 0.0009
T. fuscatus	0.8412 (24) 0.0000	0.6334 (24) 0.0009	

Correlation coefficient
(Sample size)
P-value.

T. fuscatus showed a fitting linear model (Figure 4 *b*) which described the relationship as:

$$\text{Benthic sediment (TPH conc)} = -125.174 + 15.5054 \times T.\ fuscatus\ \text{(TPH conc)}.$$

The *P*-value calculated from ANOVA was less than 0.05, thereby indicating that there is a statistically significant relationship between TPH levels in benthic sediment and *T. fuscatus* at 95.0% confidence level. Moreover, the R-Squared statistic indicated that the model as fitted explains 70.76% of the variability in benthic sediment.

T. fuscatus are opportunistic epibenthic organisms that feed entirely on organic matter found in mangrove mud, scavenging whatever food is available. From the results, the strong relation between TPH levels in the mud creeper and benthic sediment revealed that the levels of hydrocarbons in *T. fuscatus* were petroleum-derived and this could pose far-reaching health effects on humans. *T. fuscatus* is widely used in the preparation of local delicacies by inhabitants of the coastal region of Nigeria, and exposure to TPH could be through consumption.

Conclusion

Pollution problems in the coastal waters of the region appear to arise from intermittent discharges to ecosystems of crude oil, petroleum-refined products, ballast water and land-based wastewaters. Major oil spill incidents in

the region represent a significant source of hydrocarbons locally and episodically. The present study presents a baseline distribution assessment of total hydrocarbons in both the epipellic and benthic sediments of the Qua Iboe Estuary, situated in the Niger Delta region, Nigeria. The overall levels of anthropogenic hydrocarbons in the estuarine sediments when compared to similar ecosystems with substantial industrial and domestic coastal activities worldwide, revealed a moderate to high level of hydrocarbon pollution. However, owing to TPH bioaccumulative potential and toxicity to both aquatic organisms (biota) and human consumers of seafood[25,31], as well as impacting the composition and diversity of infaunal communities[32], periodical monitoring and assessment of water, sediments and tissues of various biota of the estuary and Niger Delta coastal waters is necessary.

1. Law, R. J. and Biscaya, J. L., Polycyclic aromatic hydrocarbons (PAH) – Problems and progress in sampling, analysis and interpretation. *Mar. Pollut. Bull.*, 1994, **29**, 235–241.
2. Medeiros, P. M., Bicego, M. C., Castelao, R. M., Rosso, C. D., Fillmann, G. and Zamboni, A. J., Natural and anthropogenic hydrocarbon inputs to sediments of Patos Lagoon Estuary, Brazil. *Environ. Int.*, 2005, **31**, 77–87.
3. NRC, *Oil in the Sea: Inputs, Fates and Effects*, National Academy Press, National Research Council, Washington DC, 2000, p. 806.
4. Abu-Hilal, A. H. and Khordagui, H. K., Petroleum hydrocarbons in the nearshore marine sediments of the United Arab Emirates. *Environ. Pollut.*, 1994, **85**, 315–319.
5. Farrington, J. W., Tripp, B. W. and Teal, J. M., Biogeochemistry of aromatic hydrocarbons in the benthos of microcosms. *Toxicol. Environ. Chem.*, 1982, **5**, 33l–346.
6. Benson, N. U., Essien, J. P., Ebong, G. A. and Williams, A. B., Total petroleum hydrocarbons in *Macura reptantia*, *Procambarus clarkia* and benthic sediments from the Qua Iboe, Estuary Nigeria. *Environmentalist*, 2008, **28**, 275–282, DOI: 10.1007/s10669-007-9140-6.
7. Chapman, P. M. and Wong, F., Assessing sediment contamination in estuaries. *Environ. Toxicol. Chem.*, 2001, **20**, 3–22.
8. ATSDR, Toxicological profile for Total petroleum hydrocarbons (TPH). Agency for Toxic Substances and Disease Registry, Public Health Service, US Department of Health and Human Services, Atlanta, GA, 1999.
9. Moles, A. and Norcross, B. L., Effects of oil-laden sediments on growth and health of juvenile flatfishes. *Can. J. Fish. Aquat. Sci.*, 1998, **55**, 605–610.
10. Wong, P. T. S. and Dixon, D. G., Bioassessment of water quality. *Environ. Toxicol. Water Qual.*, 1995, **10**, 9–17.
11. Wakeham, S. G., Aliphatic and polycyclic aromatic hydrocarbons in Black Sea sediments. *Mar. Chem.*, 1996, **53**, 189–205.
12. Essien, J. P. and Antai, S. P., Negative effects of oil spillage on beach microalgae in Nigeria. *World J. Microbiol. Biotechnol.*, 2005, **21**, 567–573.
13. Gamila, H. A. and Naglaa, F. A., Estimation of the hazard concentration of industrial wastewater using algal bioassay. *Bull. Environ. Contam. Toxicol.*, 1999, **63**, 407–414.
14. Fowler, S. W., Readman, J. W., Oregioni, B., Villeneuve, J.-P. and McKay, K., Petroleum hydrocarbons and trace metals in nearshore Gulf sediments and biota before and after the 1991 war: an assessment of temporal and spatial trends. *Mar. Pollut. Bull.*, 1993, **27**, 171–182.
15. Randolph, R. C., Hardy, J. T., Fowler, S. W., Price, A. R. G. and Pearson, W. H., Toxicity and persistence of nearshore sediment contamination following the 1991 Gulf War. *Environ. Int.*, 1998, **24**, 33–42.
16. Khan, M. A. Q., Al-Ghais, S. M. and Al-Marri, S., Petroleum hydrocarbons in fish from the Arabian gulf. *Arch. Environ. Contam. Toxicol.*, 1995, **29**, 517–522.
17. Asuquo, F. E., Ewa-Oboho, I., Asuquo, E. F. and Udo, P. J., Fish species used as biomarker for heavy metal and hydrocarbon contamination for Cross River, Nigeria. *Environmentalist*, 2004, **24**, 29–37.
18. Meador, J. P., Casilas, E., Sloan, C. A. and Varanasi, U., Comparative bioaccumulation of polycyclic aromatic hydrocarbons from sediment by two infaunal invertebrates. *Mar. Ecol. Prog. Ser.*, 1995, **123**, 107–124.
19. Asuquo, F. E., Tar balls in Ibeno-Okposo Beach of South Eastern Nigeria. *Mar. Pollut. Bull.*, 1991, **22**, 150–151.
20. Chindah, A. C., Braide, A. S. and Sibeudu, O. C., Distribution of hydrocarbons and heavy metals in sediment and a crustacean (shrimp – *Penaeus notialis*) from the Bonny/New Calabar River Estuary, Niger Delta. *AJEAM-RAGEE*, 2004, **9**, 1–17.
21. Essien, J. P., Ubom, R. M. and Antai, S. P., Productivity and distribution of epipellic microalgae along salinity gradients in mangrove swamp of the Qua Iboe Estuary (Nigeria). *Environ. Monit. Assess.*, 2006, **121**, 65–75.
22. Radojevic, M. and Bashkin, V. N., *Practical Environmental Analysis*, Royal Society of Chemistry, 1999, p. 465.
23. FEPA, Federal Environmental Protection Agency National Guidelines for spilled oil fingerprinting, Government Press, Abuja, 2001.
24. Goldberg, E. D., *The Health of the Ocean*, UNESCO Press, Paris, 1976.
25. Law, R. J. and Klungsøyr, J., The analysis of polycyclic aromatic hydrocarbons in marine samples. *Int. J. Environ. Pollut.*, 2000, **13**, 262–283.
26. Vignier, V., Vandermeulen, J. H. and Fraser, A. J., Growth and food conversion by Atlantic salmon pair during 40 days exposure to crude oil. *Trans. Am. Fish. Soc.*, 1992, **121**, 322–332.
27. McCain, B. B., Hodgins, H. O., Gronlund, W. D., Hawkes, J. W., Brown, D. W., Myers, M. S. and Vandermeulen, J. H., Bioavailability of crude oil from experimentally oiled sediments to English sole (*Paraphrys vetulus*) and pathological consequences. *J. Fish. Res. Bd Canada*, 1978, **35**, 657–664.
28. Mackie, P. R., Hardy, R., Whittle, K. J., Bruce, C. and McGill, A. S., The tissue hydrocarbon burden of mussel from various sites around the Scottish Coast. In *PAH: Chemistry and Biological Effects* (eds Bjørsth, A. and Dennis, A. J.), Fourth International Symposium, Battelle Press, Ohio, 1980, pp. 379–393.
29. ESGOSS, Ecological Steering Group on the oil spill in shetland. The environmental impact of the wreck of the Braer. The Scottish Office, Edinburgh, 1994.
30. Law, R. J., Kelly, C. A., Graham, K. L., Woodhead, R. J., Dyrynda, P. E. J. and Dyrynda, E. A., Hydrocarbons and PAH in fish and shellfish from Southwest Wales following the Sea Empress oil spill in 1999. In Proceedings of the 1997 Oilspill Conference. Fort Layderdale, Florida, 7–10 April 1997, American Petroleum Institute, Washington DC, Publication No. 4651, 1997, pp. 205–211.
31. Al-Yakoob, S., Saeed, T. and Al-Hashash, H., Polycyclic aromatic hydrocarbons in edible tissue of fish from the Gulf after the 1991 oil spill. *Mar. Pollut. Bull.*, 1993, **27**, 297–301.
32. Gyedu-Ababio, T. K., Furstenberg, J. P., Baird, D. and Vanreusel, A., Nematodes as indicators of pollution: A case study from the Swartkops River system, South Africa. *Hydrobiologia*, 1999, **397**, 155–169.