SHORT COMMUNICATION

HYDROCARBON ACCUMULATION AND DISCHARGE BY THE CRAYFISH IN SOME RIVERS OF LAGOS, NIGERIA

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ABSTRACT. Crayfishes exposed to ¹⁴C-naphthalene-5% of an aqueous fraction of crude oil in open non-aerated glass jars for 4 h at 25 °C showed no significant differences (P > 0.25) in the amount of naphthalene uptake. Different parts of the crayfishes were quantified for naphthalene uptake and cephalothorax containing hepatopancreas (a food absorption organ) was found to have greater uptake than the tail flesh or tail skeleton. Release of the oil from the crayfishes that were exposed for 1, 2, 3 or 4 h and later placed in open non-aerated jars containing oil-free water showed greater loss of most of the ¹⁴C activity during the first 24 h, there was little losses at 48, 72 and 96 h. There was significant volatilization (P < 0.04) of ¹⁴C-naphthalene in a 5% of crude oil placed in open, non-aerated, oil-free glass jars for 24 h at 25 °C.

KEY WORDS: Hydrocarbon accumulation and discharge, Crayfish, ¹⁴C-Naphthalene, Rivers of Lagos (Nigeria)

INTRODUCTION

Crayfish forms an important food for man and waterfowl as well, the level, retention and persistence of hydrocarbons in these invertebrates are significant.

The annual influx of petroleum hydrocarbons into the marine environment has been estimated at 5 to 10 million metric tons per year [1] of which about 30% pollute freshwater ecosystems [2-3]. The effects of crude and refined oils on marine organisms and ecosystems have been reported [4-6].

Research on petroleum hydrocarbon accumulation and discharge by marine invertebrates received considerable attention after several reports. Blummer *et al.* [7] demonstrated that the hydrocarbons contaminated Oysters (*Crassostrea virginica*) and Scallops (*Acquipecten irradians*). It was reported that different marine invertebrates accumulated hydrocarbons in proportion to the concentrations in seawater and the duration of exposure [8-10]. There were reports that the test animals to varying degrees at rather variable rates when the animals were placed in a relatively clean environment released accumulated hydrocarbons. [9, 12, 13]. Biological effects of oil pollution in freshwater ecosystems have been studied [14-16], however, little literature data are available on the accumulation of petroleum hydrocarbons by freshwater invertebrates during variable exposure periods and the subsequent release by the invertebrates in oil-free water.

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The main objective of this study was to examine the accumulation of labelled aqueous fractions (AF) of crude oil by freshwater crayfish and their release by the crayfish on exposure in oil-free, non-aerated water.

EXPERIMENTAL

Volatilization studies were carried out on the naphthalene since it was reported that more than 80% of naphthalene in some solutions after 12 h to 10 days of exposure to the atmosphere get volatilized [17-19].

A 0.1 L of the non-aerated test solution was placed in 5 different open jars and the jars were exposed to the atmosphere at 25 $^{\circ}$ C for 12 h. The solution from each jar were quantified for 14 C activity by scintillation spectrometry at 0, 1, 2 up to 24 h of exposure.

Crayfishes were collected from Lekki, Maroko, Utangan and Ijora/Ilaje of Lagos State. The crayfishes were maintained in tanks of clean, aerated water for several weeks [20]. There was no differentiation between the species and sexes; they were randomly distributed among treatments in the experiments.

The crude oil was obtained from Shell Petroleum Corporation Warri, Delta State Nigeria and the oil was placed in glass bottles and the bottles stored in a light-proof metal cabinets.

A 5% aqueous fraction (AF) of the crude oil was prepared by using Anderson *et al.* [17] mixing technique at 25 $^{\circ}$ C. The oil was decanted after separation time of 2 h. The AF was placed in 1 L Erlenmeyer flasks and capped with aluminum foil.

A 0.15 mg of crystalline ¹⁴C-naphthalene (Fluka AG, Chemische fabric, CH-9470 Buchs >98% (GC) F 79-8025) was dissolved in 0.003 L of ethyl acetate, this was diluted to 0.3 L with 0.1% Triton X-100 (0.1 g Triton X-100 in 99.9 mL of water). 0.01 L of the resulting solution was mixed with 0.09 L of the 5% AF of crude oil in oil-free glass container. This forms the test solution.

Sixteen crayfishes each weighing 1.46 ± 1.34 g were placed in an open, non-aerated, oil-free glass containers each containing 0.1 L of the test solution (¹⁴C-naphthalene + 5% AF crude oil). The exposure time 1, 2, 3 or 4 h (accumulation time) was employed after which 8 of the crayfishes were randomly picked and immediately frozen at -40 °C in order to assess accumulation. The other 8 were placed in non-aerated plastic containers (10 L) filled with 1 L of oil-free water for 24, 48 72 or 96 h (discharge time) after which they were frozen at -40 °C. Later the crayfishes were thawed, weighed and ashed in a biological materials oxidizer (Model No 621-FSE Gallenkamp Instrument Corporation). The combustion products were determined for ¹⁴C activity by scintillation spectrometry (Packard Tricarb, Model 3385, Liquid scintillation spectrometer) and corrected for differential of quenching to disintegration per minute (dpm).

Also, into another portion of the test solution were placed 16 crayfishes each weighing 1.98 ± 1.72 g for 4 h after which 8 of them were randomly picked and immediately frozen at -40 $^{\circ}$ C to asses accumulation and the remaining 8 were placed in non-aerated plastic containers (of the same size as above) containing 1 L of freshwater for 24, 48, 72 or 96 h and are later frozen. The crayfishes were dissected into tail flesh, tail skeleton and cephalothorax (head and thorax fused together) and processed as in the first experiment.

Statistical analysis. One-way analysis of variance (ANOVA) [21] was used to analyze the volatilization data. Two-way ANOVA [21] was used to analyze the experimental data. In the first experiment: 4 exposure time of accumulation by 5 exposure time of discharge, *i.e.* 0, 24, 48, 72 or 96 h and in the second experiment: 3 dissected parts by 5 exposure time in freshwater and later by Duncan's multiple range test [21] were used.

RESULTS AND DISCUSSION

The disintegration per minute (dpm) of ¹⁴C-naphthalene in the exposure solution during volatilization experiment was significantly altered (P < 0.04) following 0-24 h of exposure at 25 $^{\circ}$ C. This follows the findings of Anderson *et al.* [17] who recorded 80 to 90% loss of aqueous hydrocarbons in 24 h with gentle aeration of oil-in-water dispersions; also, Soto *et al.* [18] found that 80-90% naphthalene evaporated readily after 1 day and 99.5% loss was recorded after 10 days from naphthalene-saturated Bold's Basal medium held in non-aerated cotton plugged flasks; less than 10% of the original naphthalene escaped from the media in non-aerated glass-stopper containers; it was found that more than 80% of naphthalene lost during the first 12 h exposure of aqueous fractions of crude, fuel and used crankcase oils to the air and negligible concentrations remained after 24 h [19]. Sanborn and Malins [22] reported that hydrocarbons of low molecular weight, such as naphthalene, volatilized under static condition, also, the same thing happened with a static bioassay technique; Cradolock [23] recommended that artificial aeration should not be used with volatile toxicants, but suggested that if the dissolved oxygen levels are below saturation, aeration may be used.

In this work, high volatilization in the exposed solutions was not recorded because of the added emulsifying agent, Triton X-100, which suspended the ¹⁴C-naphthalene, the naphthalene in the aqueous fraction and the non-soluble ethyl acetate; it reduced the rate of volatilization, in the absence of which the non-soluble chemicals would have volatilized rapidly.

Figure 1a shows that the uptake of naphthalene in the first experiment did not significantly altered (P > 0.25) with exposure time (1, 2, 3 or 4 h); accumulation was approximately 0.125 mg of naphthalene.

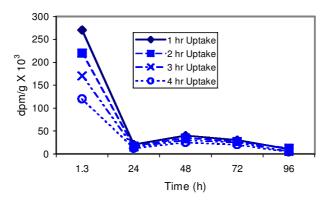


Figure 1a. Oil accumulation (dpm/g) of crayfish at time 0, 24, 48, 72, and 96 h (n = 8).

Figure 1b shows the result of the first experiment in which there was a significant reduction (P > 0.01) in ¹⁴C-dpm/g of whole crayfish after 24 h and more after 96 h in oil-free water. This pattern of loss may be as a result of discharge of ¹⁴C-naphthalene, naphthalene metabolites and most likely the components of the 5% AF of crude oil. The pattern of discharge suggested that a major component of the contaminants were discharged rapidly within 24 h, and thereafter the crayfishes gradually discharged the remaining contaminants. These findings were in line with those of Lee *et al.* [24] who found that blue marine mussels rapidly accumulated selected oil hydrocarbons from seawater solution and when placed in oil-free seawater, quickly discharge most of their accumulated petroleum hydrocarbons. Neff *et al.* [8] reported the same results for brown shrimp and brackish water clams.

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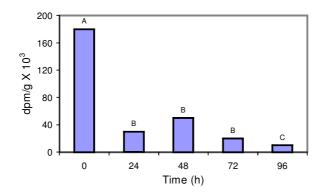


Figure 1b. Oil discharge (dpm/g) of crayfish at time 0, 24, 48, 72, and 96 h (n = 8).

However, it was reported by Boehm and Quinn [25] that not all invertebrates are capable of discharging hydrocarbons themselves once removed from a contaminated area and placed in clean water. They reported that hard shell clams (*Mercenaria mercenaria*) removed from polluted waters and placed in oil-free water only discharge 30% or less of the hydrocarbons after 120 days in the oil-free water. Such a relatively small loss indicated that the time of exposure and the chemical structures of the hydrocarbons affect the determination of their retention times in these clams.

After 24 h exposure (in the second experiment) of the crayfishes in the test solution, cephalothorax showed greatest accumulation of the naphthalene and as in the first experiment, (Figure 2a), most of the ¹⁴C activity was lost after 24 h in clean water.

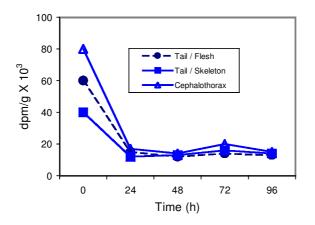


Figure 2a. Oil accumulation (dpm/g) in different body parts of crayfish at time 0, 24, 48, 72, and 96 h (n = 8).

Figure 2b shows that there was significantly more (P < 0.05) dpm in the cephalothorax than in the other body regions examined when the dpm/g was pooled across the discharge times (24, 48, 72 and 96 h).

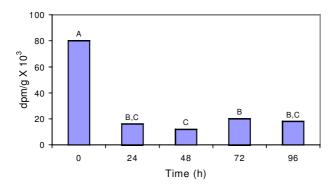


Figure 2b. Oil discharge (dpm/g) in different body parts of crayfish at time 0, 24, 48, 72, and 96 h (n = 8).

Hepatopancreas contained in the cephalothorax conferred on it the ability to have greatest accumulation of naphthalene compared to other body regions. Travis [26] in his findings reported that the hepatopancreas in the spiny lobster is a bilateral evagination of the midgut, functioning in secreting digestive enzymes and absorbing and transforming food. It also serves as a major storage depot of organic and mineral resources. Lee *et al.* [8] found on exposing blue crabs to radio-labeled hydrocarbons, that more than 50% of the radioactivity assimilated in the crab was in the hepatopancreas and after 25 days of exposure to the test solution, radioactivity could be detected only in the hepatopancreas of the cephalothorax region of the crab.

Conclusively, it was found that some crustaceans in the freshwater serving as food for man are capable of accumulating naphthalene on exposure to ¹⁴C-naphthalene + 5% AF of crude oil and are also capable of discharging naphthalene when placed in an oil-free environment.

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