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TOXICITY EFFECT OF RADIOGRAPHIC DEVELOPER EFFLUENT ON GIANT AFRICAN SNAIL (*Achatina fulica*)

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ABSTRACT

Background: The decline in the population of snails, a source of protein of people living in the high forest zone due to environmental pollution and the hazard caused by the disposal of radiographic developer effluent into streams, bushes or forests and public sewer systems makes the assessment of the effect on giant African snails (*Achatina fulica*) from environmental pollution due to radiographic developer effluent very important.

Materials and Methods: Ninety 5 months old, 12 months old and 24 months old giant African snails were randomly divided into 6 groups of 15 snails for each age group based on the dose of developer effluent to be administered. One group from each age group was designated the control and the remaining, the experimental group. Range finding test was performed at effluent concentrations of 100 %, 50 %, 25 %, 12.5 %, 6.25 %, 3.125 %, 1.6 % and 0 % (control) in 150 ml of distilled water. The effluent solution was administered on the feed and soil of the experimental snails only.

Results: Behavioural changes occurred between 0.2 – 1.0 % concentration and mortality at 24 – 96 hours exposure to the effluent solutions. The percentage (%) mortality of the giant African snails increased as the effluent concentration increased from 0.2 - 1.0 % and at increased exposure time of 24 – 96 hours. The estimated 96 hours LD_{50} for the 5, 12 and 24 months old giant African snails were 0.20 0.23, 0.23 0.25 and 0.30 0.26 respectively.

Conclusion: Radiographic developer effluent is harmful to the giant African snails, with the % mortality increasing with increase in concentration and exposure time to the developer effluent. Legislation is recommended to ensure safe disposal of radiographic developer effluents into the Nigerian environment considering the importance of giant African snails (*Achatina fulica*) to the ecosystem and the economy.

Introduction

Snails are reported to be rich in protein, potassium, phosphorus, essential amino acids, vitamins B and C [1], and form a major part of the diet of people living in the high forest zone [2] . In West Africa, snails are found mostly in humid forest areas where they are gathered for consumption [3]. Recent studies show that the wild snail population is declining due to human activities like deforestation, bush or forest burning and environmental pollution [4,5].

Radiographic developer effluent, a waste developer solution generated during radiographic film processing contain toxic organic and inorganic substances [6] such as hydroquinone, quinione, chlorides, carbonate ion, acetic acid, bromide ion, sulphates, sodium acetate, boric acid and silver in the form of complex ions [7]. These effluents when disposed into streams, bushes or forests and public sewer systems by the different radiology departments/centres cause environment pollution which affects both animals and humans negatively.

Earlier studies have shown that the toxic effect of exposure to sub-lethal doses of radiographic developer effluent resulted in 20 % mortality of Doroceras reticultum species of molluscs [8]; 50 % mortality of Catfish (Heterobranchus longifilis) [9]; tissue degeneration and necrosis of the spleen of Wistar rats [10] and alterations in the histology of the heart of Wistar rats [11]. In view of the toxic effect of developer effluent on the already mentioned organisms and on aquatic biota [12], this study assesses the effect on giant African snails (*Achatina fulica*) from environmental pollution due to radiographic developer effluent.

Materials and Methods

Two hundred and seventy giant African snails (*Achatina fulica*) made up of 90 five months old, 90 twelve months old and 90 twenty-four months old were used for this study. These animals were randomly divided into 6 groups of 15 snails for each age group, for example the 5 months old were divided into 6 groups of 15 snails, same for 12 and 24months old based on dose of developer effluent to be administered. For each age group, one out of the 6 groups was designated as the control and the rest, the experimental groups.

Experimental set up

Six snaileries of triple chambers each were constructed with dimensions of 200 cm x 80 cm x 12 cm high. The base (floor) was 20 cm thick and 12 cm above the ground. One snailery housed the control snails while the others housed the experimental snails. Each chamber had a dimension of 69 x 80 cm for the different age groups and different concentrations of developer effluent to be administered. The snails were allowed to acclimatize to the environmental conditions of 29 ± 2 °C temperature, 40 - 55 % humidity and fed with pawpaw leaves [1] on wet humus soil for a period of 120 hours (5 days). This experiment was designed to be both dose and time dependent.

Range finding test

Toxicity test was performed to determine the approximate range of the radiographic developer effluent. Toxicant concentration of 100 %, 50 %, 25 %, 12.5 %, 6.25 %, 3.125 %, 1.0 % and 0 % each was prepared in 150 ml of distilled water. These effluent concentrations were used to moist the snail's chamber and mixed with the feed before the snails were introduced into the chamber. The experimental snails were observed at 2, 4, 6, 12 and 24 hours for any behavioural change and mortality between 24, 48, 72 and 96 hours. The control chamber was free from any chemical. From the range finding test, the concentration that gave 100 % mortality within 96 hours was selected as the highest dose for the toxicity test. The toxicity test was conducted using concentrations of 0% (control), 0.2 %, 0.4 %, 0.6 %, 0.8 % and 1.0 % on the 6 groups of 15 snails of 5, 12 and 24 months old each.

Estimation of percentage mortality

The percentage (%) mortality was determined from Equation 1 using Abbot's method [13]:

$$\% \text{ CM} = \frac{AC \quad AT}{AC} x\ 100 \qquad 1$$

where $\% CM$ – corrected percentage mortality.
$\% AC$ – percentage alive control.
$\% AT$ – percentage alive treated.

Estimation of mean effective lethal dose

The mean effective lethal dose for 96 hours lethal dose (LD_{50}) was estimated from Equation 2 using Spearman Karber method [14]:

$$96 \text{ hours Ld50 (M)} = xk - d\ (s\text{-}½) \qquad 2$$

where: LD_{50} or M - mean effective lethal dose.
xk - largest test dose which produces 100% mortality.
d - difference between adjacent doses.
s - cumulative portions of % dead snails.
Calculating the standard deviation associated with this 96hrLD_{50} (SLD_{50} sm) from Equation 3

$$SLD_{50} = d\sqrt{2(S_2) - S_1 - S_1^2 - ½} \qquad 3$$

where:
SLD_{50} - standard deviation associated with this 96hrLD_{50}
S_1 - Sum of the relative portions of each reacting organism

S_2 - Sum of the cumulative added portions of the reacting doses

Results

The results of the toxicity tests carried out on the giant African snails (*Achatina fulica)* after 96 hours of exposure to different radiographic developer effluent concentrations are shown in Figure 1 and Tables 3 – 5.

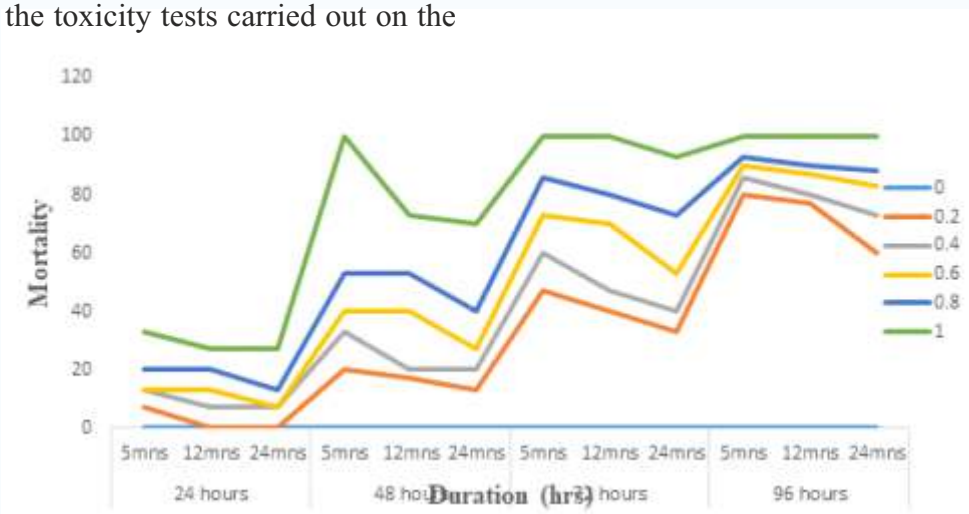

FIG. 1. Dose - response relationship of the snail population after exposure to varying concentrations of the developer effluent.

Figure 1, presents the % mortality among the snails after exposure to varying concentrations of developer effluent. The % mortality increased with increase in the concentration and exposure time to the developer effluent for all the experimental snails irrespective of age. Also, the degree of % mortality was related to the age of the snails. Increasing the concentration and exposure time to the developer effluent initially affected a greater population of the younger snails than the older ones. Again, snails of the same age group showed different responses to the same concentration of radiographic developer effluent. However, further increase in concentration and exposure time affected all the experimental snails irrespective of age.

From figure 1, below 1.0 % (highest) concentration and at 96 hours (maximum) exposure time, the % mortality was below 100 % for all the snails. At 1.0% concentration and exposure time of 24 hours, the % mortality was less than 35 % for the experimental snails irrespective of age. Increasing the exposure time to 48 hours and at 1.0 % concentration resulted in 100% mortality for all the 5 months old snails. Further increase in exposure time to 72 hours at 1.0 % concentration resulted in 100 % mortality for all the 12 months old snails. There was 100 % mortality for all the experimental snails irrespective of age when the exposure time was again increased to 96 hours at 1.0% concentration of the developer effluent.

Tables 1 – 3 depicts values of the % dead snails, the relative portion of % dead Snails and the cumulative portion of % dead snailsincreasing with increase in the concentration of the developer effluent for the 5, 12 and 24 months old snails. The calculated values of 96 hours LD_{50} from Spearman Karber method [14] for the 5, 12 and 24 months old snails were 0.20 0.23, 0.23 0.25 and 0.30 0.26 respectively.

Table 1: Calculated 96 hours LD_{50} values for five months old *Achatina fulica*

Concentration (%)	% of dead snails	Relative portion of % dead snails	Cumulative portion of % dead snails
0	0	0	0
0.2	80.00	0.80	0.80
0.4	86.60	0.87	1.67
0.6	90.00	0.90	2.57
0.8	93.30	0.93	3.50
1.0	100	1.00	4.50
Total		**S_1= 4.59**	**S_2= 13.03**

Table 3: Calculated 96 hours LD_{50} values for twelve months old *Achatina fulica*

Concentration (%)	% of dead snails	Relative portion of % dead snails	Cumulative portions of % dead of snails
0	0	0	0
0.2	77.40	0.77	0.77
0.4	80.30	0.80	1.58
0.6	87.00	0.87	2.45
0.8	90.30	0.90	3.35
1.0	100.00	1.00	4.35
Total		**S_1 =4.35**	**S_2 =12.50**

Table 4: Calculated 96 hours LD_{50} values for twenty four months old *Achatina fulica*

Concentration %	% dead Snails	Relative portion of % dead Snails	Cumulative portion of % dead Snails
0	0	0	0
0.2	60.00	0.60	0.60
0.4	73.30	0.73	1.33
0.6	80.00	0.80	2.13
0.8	88.30	0.88	3.02
1.0	100.00	1.00	4.02
Total		**S_1 = 4.02**	**S_2 =11.10**

Discussion and Conclusion

The results obtained in this study showed that the radiographic developer effluent is harmful to the giant African snails (*Achatina fulica*). The % mortality of the experimental snail population increased with increase in the concentration and exposure time to the developer effluent implying that the intensity of the effect increased with increase in the concentration [15, 16]. The increase in the concentration and exposure time which initially affected the younger snails more maybe due to the fact that early life stage organisms are more sensitive to toxicants than the older ones [16]. The different responses to the same concentrations of radiographic developer effluent by snails of same age group indicated the genetic variation in the population of experimental snails [15]. Further increase recorded 100% mortality at 1.0 concentration and 96 hours exposure time for all the experimental snails irrespective of age thus confirming that intensity of an effect increases with increase in concentration [15,16].

Mortality of the snails may have been due to the presence of hydroquinone and boric acid in the radiographic developer effluent. Hydroquinone, a major component of the developer solution has been implicated in the 20 % mortality of Doroceras reticultum species of molluscs after exposure to developer effluent of 0.02 mg/litre for 4 days [8]. Hydroquinone has also been reported to be a haematotoxic and carcinogenic compound to both humans and other animals [17] and the most toxic dihydroxybenzene affecting soil microbial activity [18]. Boric acid on the other hand has been reported to cause histopathological degenerative changes in kidney tissue [19] and significantly inhibits the development of the spleen [20]. The already mentioned effects due to hydroquinone and boric acid may have been the cause of death of the experimental snails.

The calculated 96 hours LD_{50} values increased with increase in age, implying that 50 % mortality increased with increase in the age of the snails [15,16]. The estimated safe discharge limit of the developer effluent is reported to be one-tenth or 10% of the LD_{50} [21]. The safe discharge limit of radiographic developer effluent for 5, 12 and 24 months old giant African snails were therefore 0.20 0.23, 0.23 0.25 and 0.30 0.26 respectively. This implies that the age of the snails plays a significant role in the determination of the safe discharge limit for radiographic developer effluent.

The radiographic developer effluent is harmful to giant African snails **(*Achatina fulica*).** The harm increased with increase in the concentration and exposure to the developer effluent. In view of the

harmful effect of the exposure to sub-lethal doses of radiographic developer effluent on *Achatina fulica,* this study recommends the need for legislation to ensure adequate treatment before disposal of radiographic developer effluent into the environment and its inclusion in the toxicity database of the National Environmental Standards and Regulations Enforcement Agency (NESREA) Act.

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