ABSTRACT
This study examined the behavioral changes induced by butachlor to the juveniles of *Clarias gariepinus*. The experiment consists of five treatments (0.6, 0.7, 0.8, 0.9 and 1.0 mg/l) and a control. Ten fishes were allotted in each test tank in duplicates. Observations on behavioral responses were made at 0, 12, 24, 48, 72 and 96 hours post-exposure. The opercular movement and tail fin beat rates were observed as counts per minute using a stopwatch. The behavioral patterns of the fish in the control group were normal, whereas the exposed groups showed hyperactive movement, became hyperactive at the 48th hour. The mean values of the tail fin beat of the exposed groups were significantly higher (p<0.05) than that of the control at the 12th hour. At the 24th and 48th hour post-exposure the exposed groups showed significant (p<0.05) time-dependent decrease compared to the control. The tail fin beat became significantly higher (P<0.05) in the control group from 72nd hours onward. The decrease of tail fin beats at the 72nd and 96th hours were dose-dependent similar observations were also recorded in the opercular ventilation. At the 12th hour the opercular ventilation of the exposed fish was significantly higher (p<0.05) than the control whereas at the 24th and 48th hours, the opercular ventilation of the exposed groups showed a significant decrease (p<0.05) compared to the control.

KEYWORDS: Butachlor, Herbicide, Acute, Behavioral, *C. gariepinus*

INTRODUCTION
The use of herbicides has been recognized as part of agricultural practices throughout the world. Unfortunately, the arbitrary use of these herbicides to enhance agricultural production and yield may have negative effects on non-target terrestrial and aquatic organisms (Nwani et al., 2010). Environmental pollution by herbicides has become a serious problem in terms of global conservation, animal and human health (Katsumata et al., 2005). According to Cerejeira et al. (2003) and Flynn and Spellman (2009), herbicides are ubiquitous agricultural chemicals that can proliferate regionally and globally into many ecosystems. Over the last few decades, the use of these chemicals has considerably increased about increasingly intensive agricultural practices.

Herbicides can accumulate in bodies of water to levels that kill zooplanktons and juvenile fishes and can also kill insects on which some fish feeds, thereby causing the fish to travel a distance in search of food and this expose them to greater risk from predators (Ikpesu, 2013). Herbicides, a distinctive group of pesticides, are considered as-selective chemical weed killers; hence they have been intensively used to destroy unwanted plants, especially in agricultural lands (Dutta and Meijer, 2003). The impact of herbicides is often greater than what is intended by those who use them, surface run-off into rivers and streams can be highly lethal to aquatic life. Herbicides reduce environmental quality and influence essential ecosystem functioning by reducing species diversity and modifying food chains (Alishahi et al., 2016). Aquatic bioassays are however necessary for water pollution control to determine whether a potential toxicant is dangerous to aquatic life (Olaifa et al., 2003). The increasingly widespread use of herbicides in agriculture and their impact on non-target aquatic organisms require specific studies on their effects on economically significant species such as the African catfish (*Clarias gariepinus*). Fish can serve as bioindicators of environmental pollution and therefore can be used for the assessment of the quality of the aquatic environment (Lopes et al., 2001) since they are directly exposed to chemicals resulting from agricultural production via surface run off of water or indirectly through the food chain of the ecosystem (Ateeq et al., 2002). The present study is aimed at studying the behavioural alterations caused by butachlor to juveniles of *C. gariepinus*.

MATERIALS AND METHOD
Preparation of Test Solution
A commercially available herbicide Butatex® with Butachlor as an active ingredient was used for the study. The herbicide was purchased from an agrochemical shop in Sabon-Gari market, Zaria. The stock of the test chemical was prepared by dissolving 1 mg of Butachlor in 1 litre of test water in a conical flask following the procedures of Dede and Kagbo (2001).

A range finding test was carried out to check for the concentration of the herbicide that was used for the definitive tests. This was done by placing five nominal concentrations (0.5, 1.0, 1.5, 2.0 and 2.5 mg/l) of the herbicide in separate tanks and ten fishes were stocked in each tank, mortality of the stocked fish was observed at 0, 12, 24, 48, 72, and 96 hours. The concentrations were graded using lower ranges until 80-90% mortality was recorded in the highest concentration and 20-30% for the lowest concentration as suggested by Martins et al. (2008).

Acute Toxicity Test
A four-day static toxicity bioassay was conducted in the Fisheries Laboratory of the Department of Biological Sciences Ahmadu Bello University, Zaria, using juveniles of *C. gariepinus*.
The experiment was a completely randomized design with five treatments and a control in duplicates, (0.0, 0.6, 0.7, 0.8, 0.9 and 1.0 mg/l). Butachlor was added to each glass tank (30 cm x 30 cm x 45cm) containing 20l of de-chlorinated tap water. The addition was done for varying concentrations using a syringe and allowed to stand for 10 minutes for the toxicant to be evenly distributed. Ten fishes were allotted to each tank. Observations of the behavioral responses were made at 0, 12, 24, 48, 72 and 96hours post-exposure. The opercular movement and tail fin beat rates were observed as counts per minute using a stopwatch. The opercular movement and tail fin movement were counted for three fishes in each tank and the mean recorded. (OECD 1992).

**RESULTS**

Behavioral responses of *C. gariepinus* juveniles exposed to acute concentrations of butachlor

The behavioural symptoms observed include loss of equilibrium, erratic swimming, hyper and hypoactivity. The fish in the control tank was normal throughout the experimental period, while the exposed fish appeared to lose equilibrium and swim erratically with vigorous jerky movements. At the 12th and 24th hours, the fish in the exposed groups showed hyperactive movement until the 48th hours and onwards where the fishes in the exposed groups were slow in their activity. The exposed fishes stayed in a vertical position for a while before they start moving again (Table 1).

The results of tail fin beats and opercular ventilation are presented in figures 1 and 2 respectively. The mean values showed that the tail fin beats of the exposed groups were significantly higher (p<0.05) than that of the control by the 12th hour. At the 24th and 48th hour post-exposure, the exposed groups showed a significant (p<0.05) time-dependent decrease compared to the control. By the 72nd and 96th hours the tail fin beats of the control group were significantly higher (p<0.05) than the exposed groups, the decrease of tail fin beats at the 72nd and 96th hours were dose-dependent (Figure 1), similar observations were also recorded in the opercular ventilation (figure 2). At the 12th hour the opercular ventilation of the exposed fish was significantly higher (p<0.05) than the control, by the 24th and 48th hours, the opercular ventilation of the exposed groups showed a significant decrease (p<0.05) compared to the control. Similar observations were also made at the 96th hour.

![Figure 1: Tail fin beats (per minute) of *C. gariepinus* juveniles exposed to acute concentrations of Butachlor.](image-url)
**DISCUSSION**

Fish exposed to different concentrations of the herbicide displayed behavioural abnormalities in response to the herbicide. At the early exposure, fish stop swimming and remain stationary, this might be in response to the sudden change in the water medium, after some time the swimming of the fish in the exposed groups became erratic with vigorous jerky movements, hyperactivity, faster opercular and tail fin movement restlessness and gulping of air. Hyperactivity of fish in the exposed group could be attributed to an attempt to evade the test water (toxic environment). El-Sharkawy et al. (2011) reported that jumping to the water surface to gulp air by fish could be attributed to either oxygen depletion as a result of pollution caused by herbicides or other toxic chemicals present in the water or irritation caused by dermal contact and subsequently this irritates the gills. Mekkawy et al. (2013) also reported hyperactivity in *C. gariepinus* exposed to atrazine which was characterized by rapid and erratic swimming darting, partial loss of equilibrium, rapid pectoral fin and opercular movement, reduction in feeding activity and fin haemorrhage.

The behavioural and swimming patterns of the fish in the control group in the present study were normal but for the exposed groups abnormal swimming behaviour increased with an increase in the toxicant concentration. Similar observations were made by Ayoola (2008), Kumar et al. (2010), Bekeh et al. (2011), Banaee et al. (2011), Chang et al. (2011), Pandey et al. (2011), Altinok et al. (2012) and Nwani et al. (2013).

The early increase in opercular ventilation count in the exposed groups may be an attempt by the fish to suppress the effect of the toxicant by breathing faster. The increase in opercular ventilation rate may also be linked with the sudden response of the fish to the shock of exposure to the toxicant (Chindah et al., 2004). This hyperventilation probably an attempt by the fish to cope with the environment at the initial period but with the increase in exposure time, the fish become weak and finally die due to suffocation. The increase might also be attributed to an increase in oxygen demand needed for the increased metabolic activities. This finding is comparable to that of Nwani et al. (2013) who reported faster opercular movements in *C. gariepinus* exposed to termifos. Obiezue et al. (2014) also noted rapid opercular movement in diethyl exposed *C. gariepinus*. The authors affirmed that this as well as other abnormal behaviours observed suggested nervous disorder. Similar findings were also reported by Auta et al. (2000) and Bayero (2017). Physiologically, fish reacts to toxicants by increasing rate of respiration in an attempt to pass more water and invariably oxygen over the surface of the gills. However, the fish ends up passing more toxicant over the gills, and a decline in opercular ventilation counts with increase in the exposure period. The decline in opercular ventilation count observed in this study may be credited to a decrease in respiratory rates and suggested decreased oxygen consumption because the fish has become exhausted due to several attempts to escape from the toxic medium or frequently surfacing to facilitate oxygen intake. Auta et al. (2002) and (2005) reported a decrease in opercular ventilation count with increased exposure time. In contrast, Yaji (2012) observed an increase in the opercular ventilation count of *Oreochromis niloticus* exposed to acute concentrations of cypermethrin in the static assay at the 96th hour.

The initial increase in the tail beat frequency of fish in exposed groups observed in the present study can be linked to increased metabolic activity by the fish to swim faster and escape the polluted environment. However, as exposure time increases the tail fin beats decrease Ufodike and Omorogie (1990) reported that, increase in opercular ventilation and tail fin beats within initial periods of toxicant exposure act as an avoidance syndrome exhibited by fish. However, as this effort became...
ineffective, fish became weak and a decline was observed in the tail fin beat frequency with an increase in exposure time. Tail fin movement is a mechanical activity and requires the use of energy. This decline in tail fin movement rates can be attributed to disorder in energy synthesis possibly through blockage of the electron transport chain. Similar findings were reported by Chindah et al. (2004), Ogundele et al. (2004) and Onuoha and Ohatuonye, (2007). However, Yaji (2012) reported a different finding in O. niloticus exposed to cypermethrin for 96 hours in both static and flow-through experiments; the author recorded an increased tail movement rate in the 96th hour of exposure.

CONCLUSION

It is obvious from the findings that tachlor imposed behavioral changes in Clarias gariepinus juveniles, which might have caused by the sudden change in the water environment. The sudden change caused the fish to move faster to escape the toxic environment.

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