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A CHANGE IN THE BEHAVIOURAL PATTERNS IN *Pangasianodon hypophthalmus*- AN EFFECT DUE TO COMBIFLAM

Pharmaceutical Science

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ABSTRACT

This paper discusses the alternations in behaviour mechanisms of *Pangasianodon hypophthalmus* exposed to combiflam, a universally taken pain relief drug. Behaviour such as swimming, surfacing, aggressiveness, grouping and feeding were observed for dose concentrations increasing from 150mg to 2250 mg. The concentration of 450 mg showed a decrease in swimming speed and reduced appetite along with instances of fish surfacing for air. Fish are not seen swimming in groups anymore. The increase in dosage resulted in a sudden decrease and then increase in the swimming speed, whereas the other behaviours intensified with an increase in dosage concentration. This study gives an insight on the harmful effects of combiflam on fish behaviour.

KEYWORDS:

Combiflam, behaviour, *Panagsionodon hypophthalmus*

INTRODUCTION

In the last few years, there is a growing concern regarding Environmental pollution caused by EDC's mainly pharmaceuticals and pesticides. A variety of pharmaceutical drugs enter the waterways via wastewater effluents from households and industries and remain biochemically active in the aquatic systems (T. Brodin, 2013). Pollution of any kind in these waters affect the terrestrial as well as the aquatic life (Wright, P. A., 1993; Thibaut, R., & Porte, C., 2004).

Combiflam is a frequently used pain relief medicine around the globe which is available without any prescription. Combiflam a NSAID (Non Steroidal Anti Inflammatory Drug) is composed of Ibuprofen and paracetamol. Ibuprofen is anti-inflammatory in action, caused by the inhibition of COX 2 enzymes in the body. On the other hand, paracetamol is analgesic and antipyretic in nature but has minimum anti-inflammatory action. Combiflam users are at a high risk of gastrointestinal problems along with renal defects. Ibuprofen especially is said to affect the hormonal system. Abuse of combiflam may cause nausea, vomiting, skin reactions, etc.

This drug enters the fresh water bodies via the sewage systems and their incomplete removal during the wastewater treatment process may be harmful to the aquatic life present there.

Most studies in the field of toxicity and pollution have only focused on the physiological death (mortality) of an organism. Very rarely the ecological death is given any importance. Ecological death is nothing but an inability to function in an ecological context when behaviours get altered due to an external factor (Gaworecki and Klaine, 2008, Mohamed Nassef, 2010). Behaviour serves as an indicator of the stress and discomfort that the animal experiences and also are used as a tool for ecological risk assessment.

The major objective of this study was to investigate the altering changes in behavioural patterns of *Pangasianodon hypophthalmus* exposed to low concentrations of combiflam. *P. hypophthalmus* is a fresh water fish along the coast of Maharashtra, cultured in ponds, lakes and rivers and is commonly eaten as it is a cheap and easily procurable fish.

MATERIALS AND METHODS

Test organism

Sixty juvenile *Pangasianodon hypophthalmus* were procured locally from freshwater areas of Mumbai region, Maharashtra with an average weight of approximately 10-15 gms. The test fish were acclimatised for 2 weeks prior to experimentation and were held in glass aquaria equipped with an aerating system. The fish were fed with comme rcially available fish food pellets.

Test chemicals

Combiflam (Ibuprofen 400mg & Paracetamol 325mg) was bought from the local chemists in Mumbai, Maharashtra.

Experimental design

Post acclimatisation, the fish were divided into two groups: control group consisting of 30 fish and an experimental group consisting of the other 30 fish. The experimental fish was exposed daily for a duration of 15 days to 150mg of chemical dissolved in 0.3ml of alcohol per 20 L of tank water, making up a concentration of 7.4ppm. The total concentration at the end of 15 days is 110.8ppm. During the experi ment, the behaviour of the control and experimental fish was monitored.

Behavioural measurement

The behaviours of experimental fish and control fish were observed daily post exposure to the chemical. Swimming, Breathing, Feeding, Aggressiveness and anxiety behaviours were monitored.

RESULTS AND OBSERVATIONS

During the course of the experimentation, the control and experimental fish were observed for any alterations in behaviour. The following behaviours were observed:

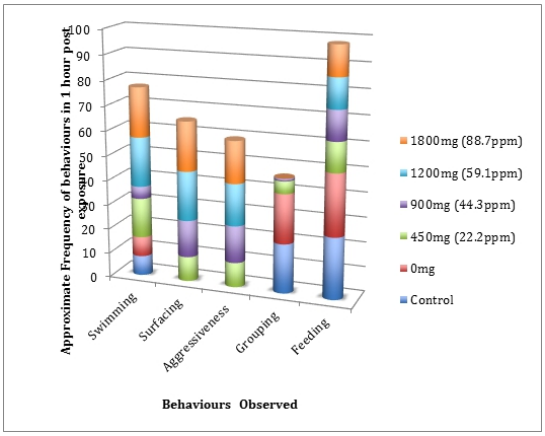
- Swimming behaviour: Swimming speed of the fish
- Surfacing behaviour: Rise to the surface of the water for air
- Aggressive behaviour: Nudging or dashing each other and the walls of the tank
- Feeding behaviour: 50% or more amount of feed consumed within an hour.
- Grouping behaviour: Swimming in groups of 3 or more

The control fish displayed regular swimming patterns, no surfacing or aggressive behaviour throughout the experiment. Fish were seen to swim in groups throughout and feed consumption was 100% at the end of 1 hour.

On initial exposure to the drug the experimental fish displayed normal swimming speed, no surfacing or aggressive behaviour, swimming in groups of 4 or more and the feed was consumed immediately. As the concentration increased to 450 mg (22.2ppm) the experimental fish exhibited an increase in swimming speed along with slight aggressiveness, feeding and grouping behaviour remained unaffected. At a concentration of 900 mg (44.3) the experimental fish showed excessive surfacing phenomenon coupled with isolated swimming patterns with a sudden decrease in the speed. The fish also exhibit highly increased aggressiveness towards each other and a loss of appetite i.e at the end of 1 hour less than 50 % of feed is consumed. These behaviour modifications remain constant upto concentrations below 1200 mg (59.1ppm). At a concentration of 1200 mg (59.1ppm) the aggressiveness and lack of appetite continue with an increase in

surfacing mechanism. The fish swim isolated however with an increased speed. The above behaviours become more intense at 1800 mg (88.7ppm) continue to be persistent until the end of the experiment.

Table 1: approximate frequency of alterations in fish behaviour after 1 hour of increasing dose concentrations



DISCUSSION

Environmental pollutants pose a great risk to the aquatic life. The manner in which an animal acts or behaves give a well defined understanding of its health and overall well being. Behaviour links the physiological health of the fish with the ecological environment providing an insight into the environmental problems.

As seen in table 1. Exposure to a low dosage of combiflam elicited behavioural responses different from their natural behaviour.

Swimming is a mode of locomotion for the fish. As seen in the above table, control fish show a regular speed of swimming and swim in groups which are similar to the experimental fish at concentrations below 450 mg (22.2ppm). As the dose concentration reach 450 mg (22.2ppm) the fish swim with an increased speed. The fish are seen to swim isolated and this remains constant until the end of the experiment. However, with the increase in the concentration to 900 mg (44.3) the fish are observed to swim with decreased speed which again seems to increase at a concentration above 1200 mg (59.1ppm). This is comparable to work by Daniel Baganz, who demonstrated a change in locomotor activity with an increase and then decrease on motility in Zebra fish exposed to cyanobacteria toxin, Microcystin-LR. Surfacing for air and nudging and dashing each other and the tank is seen to be absent in control as well as in concentrations below 450 mg. However, as the concentration crosses 450 mg (22.2ppm), surfacing phenomenon coupled with aggressiveness is seen to appear and the frequency goes on increasing intensely with the increase in dose concentration which is similar to the studies of VineetKumar K Patil wherein the Aggressive behaviour is seen to increase in Labeo rohita on exposure to Malathion

Feeding is an innate behaviour and is important for the health of the fish. Control fish consume all the feed within 10 15 mins of its availability demonstrating a healthy behaviour. However as the concentration of the dose reached 450 mg (22.2ppm) this behaviour changes. At the end of 60 mins, less than 50 % of feed is consumed and this is persistent throughout and can be compared to studies by Daniela Baganz on feeding behaviours of Zebra fish due to the Impact of the cyanobacteria toxin, Microcystin-LR

The higher the concentration the more intense and prolonged alteration in behaviours.

These results are in accordance with the studies of Rekha Rao, which demonstrated different breathing patterns ranging from normal to hyper to hypo with respect to the toxic dosage of paracetamol and also displays schooling behaviour in *C. carpio*.

Most of the past research has been carried on ibuprofen and paracetamol individually and focuses on the mortality and reproductive process. Our research describes and confirms the effects of combiflam on the behavioural mechanisms of *P. hypophthalmus*.

CONCLUSION

Combiflam, one of the most frequently consumed drugs by human beings, when released into the waste waters via human excretory system the drug in its metabolite form enter the waters and may affect the aquatic animals present there.

From the outcome of our study, it is possible to conclude that Combiflam affects the overall behaviour patterns of the fish, *P. hypophthalmus*. The fish experiences stress due to the drug which is exhibited by the alterations in the behaviour. *P. hypophthalmus* is regularly eaten by the local people of Maharashtra and it can be safely assumed that the affected fish on consumption may also affect humans. The impact on humans may be as drastic and diverse. Further research is needed to define the effects of combiflam on the physiology and metabolism of the fish. This research was concerned with combiflam and fresh water food fish *Pangiasonodon hypophthalmus,* however, the results should be applicable also to other pain relievers and fresh water food fish.

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