

## Evaluation of Propofol Anaesthesia and its Effect on Haematological and Biochemical Blood Profile of African Catfish (*Clarias gariepinus*)

*Babalola Samuel Adeola, Ndor Chisom Juliet, Jeremiah, Kelechi Theresa,  
Njoku Uchechukwu Njoku and Unamba-Oparah Chioma*

Department of Veterinary Surgery and Radiology, College of Veterinary Medicine Michael Okpara  
University of Agriculture, Umudike, Abia state, Nigeria

**Abstract:** Propofol (2,6-diisopropylphenol) effectively induced general anaesthesia in African catfish in the preliminary test with mean induction/exposure (time to anaesthesia) time recorded as  $5.3 \pm 0.74$  minutes while the mean recovery time from anaesthesia was recorded as  $3.2 \pm 0.48$  minutes. The aim of this study is to evaluate propofol anaesthesia and its effects on the haematological and biochemical blood profile of African catfish. A total of 30 African catfish from the same farm were purchased. Blood sample was collected from the caudal vessel from non-treated fish (control), treated and 24 hours post treatment. Six (6) fish were immersed one after the other in a 30 litre water bath treated with solution of 2 mg/l Propofol for 5 minutes. Haematological and biochemical indices were analysed and compared to control group. The results of the haematological and the blood chemistry analysis show no significant differences in the means when compared to the control except for that of the neutrophils and lymphocyte that shows significant difference of  $p \leq 0.01$  and  $p \leq 0.05$  respectively. Propofol is safe and effective depressant drug for different tasks related to the management of African Catfish, since it met the established criteria for use in aquaculture.

**Key words:** Propofol • African Catfish • Anaesthesia • Haematology • Blood Profile

### INTRODUCTION

In recent years, different types of anesthetics are used to aid in the capture, handling, artificial reproduction, surgery procedures and transport of fish as an anti-stress in modern aquaculture [1]. A few number of anesthetics have proved effective in fish anaesthesia with its own advantages and drawbacks [2].

Till now, only MS-222 (tricaine methanesulfonate) is registered for use on food fish in the U.S. and the United Kingdom. However, aquaculture industry needs more compounds to be evaluated experimentally [3] and introduce on ornamental and food fish. Some anaesthetics reduce or block the activation of the hypothalamic-pituitary-interrenal (HPI) axis associated with stressors and thus decrease or prevent the release of the stress hormone cortisol to the bloodstream of fish [4]. Anesthetics act on the central nervous system in such a way by placing the fish into an anaesthetic solution, the

anaesthetic is absorbed through the gills and enters the arterial blood. With the returning of the anaesthetised fish to the fresh water, the anaesthetics or their metabolites are excreted via the gills [5].

Propofol (2,6-diisopropylphenol) is a short acting anaesthetic with accelerated metabolism, rapid uncomplicated and complete recovery after administration in bolus doses or by continuous infusion, Gomułka *et al.* and Sawyer [6, 7]. In addition, it does not have cumulative effect like thiopental [8, 9]. Propofol depressant action involves a positive modulation of the gamma-aminobutyric acid neurotransmitter inhibitory function (GABA), through GABAA receptors [10, 11]. Propofol provides a reliable, rapid and smooth induction of anaesthesia, adequate hypnosis and analgesia for surgical interventions and minimal suppression of vital organ functions, it prevents peak of cortisol levels and preserves hematological, morphological and biochemical stability [12]. The efficiency and safety of any anesthetic

agent may vary according to species, stage of life and environmental conditions [9].

African Catfish; *Clarias gariepinus* and *Clarias anguillaris* remain the two catfish species most farmed in Africa, even though more than 100 species populate African waters. *Clarias gariepinus* is commonly referred to as African Catfish, Common Catfish and Mudfish [13].

## MATERIALS AND METHODS

**Fish:** African Catfish ( $n = 30$ ) (*Clarias gariepinus*) were supplied by Sungun aquaculture farm located in Aba, Abia State Nigeria. The experiment was carried out on the farm. Mean weight was  $300 \pm 0.87$  g. Water supply is from deep well with temperature of  $20 \pm 0.3^\circ\text{C}$ , pH of  $6.57 \pm 0.4$  and oxygen saturation above 75% during all the experiments.

**Anaesthetic Agent:** Propofol 10 mg/1ml (POFOL® INJECTION I.V. Dongkook pharmaceutical, Choong cheong Book-DO, Korea).

**Experimental Design:** To avoid environmental and handling stress. The research was carried out on the same farm the fish were hatched and raised, using the same source of water for the research. Physico-chemical characteristics of the water in the establishment were within the limits required for production, temperature, pH and oxygenation remained stable throughout the course of work Eighteen (18) fish were randomly selected from the same batch with mean weight of  $300 \pm 0.872$  g and put together in a 200 litre water tank. To avoid environmental and handling stress the fish were allowed 24 hours to acclimatise during which they were fasted before commencement of the experiments.

About 2 millilitres of blood was collected from the caudal vessel from randomly selected six fish ( $n1 = 6$ ), 1 millilitre in EDTA was used for haematological analysis. Haematological indices were determined according to standard methods given in Unified methods for haematological examination of fish [14]: erythrocyte count (RBC), haemoglobin concentration (Hb), Haematocrit (PCV), mean erythrocyte volume (MCV), mean corpuscular haemoglobin concentration (MCHC), mean corpuscular haemoglobin content (MCH), leukocyte count (WBC) and the differential leukocyte count (Leukogram). The other 1 millilitre without anti-coagulant was use for blood chemistry. Analysis of biochemical parameters included: total protein (TP), albumin (ALB), ammonia ( $\text{NH}_3$ ), bilirubin (BIL), aspartate

aminotransferase (AST), alanine aminotransferase (ALT) and alkaline phosphatase (ALP). The data obtained served as control for the subsequent experiment carried out.

Using modify Gomulka [9] method, the water for immersion was kept at room temperature in a 30 litre water bath and treated with propofol at the dose rate of 2.0 ml/10L for the preliminary study.

Another set of six fish ( $n2 = 6$ ) were randomly catch out of the tank from the remaining 24 and individually immersed into the treated water with propofol and observed for induction of anaesthesia, noting the following, stages I. Loss of equilibrium; stage II. Loss of gross body movements but with continued operculum movements and stage III as in II with cessation of operculum movements. The exposure time (ET) was also noted. Following induction/exposure of anaesthesia, the fish were transferred individually into another 50 litre tank containing untreated water and the time of their recovery noted.

Following the determination of the mean ET. Another set of randomly selected six fish ( $n3 = 6$ ) was selected from the remaining 18 untreated fish. Using the method employed for  $n2$  above. The fish were individually immersed in the treated water for 5 minute, after which they were brought out and about 2 ml of blood collected from the caudal vessel for the same analysis as described earlier above. All the  $n3$  after blood collection were put together in a separate pond on the farm containing untreated water, they were left for 24 hours. Blood was collected from them after 24 hours as described above earlier for haematological and blood chemistry analysis.

**Statistical Analysis:** All the data collected were express as mean  $\pm$  standard errors of mean using statistical package for social sciences (SPSS) and the means were tested for significant differences using ANOVA were  $p = 0.05$  is declared as significant.

## RESULTS

**Anaesthetic Evaluation:** In the preliminary study, determination of the induction/exposure, all the three stages of anaesthesia were observed in all the treated fishes with all the stages overlapping each other. Stage I: Mean partial equilibrium loss (minutes)  $1.03 \pm 0.32$ ; Stage II: Mean total equilibrium loss (minutes)  $3.19 \pm 1.11$  and the mean induction/exposure (time to anesthesia) time was recorded as  $5.3 \pm 0.74$  minutes. The mean recovery time from anaesthesia was recorded as  $3.2 \pm 0.48$  minutes.

Table 1: Haematological parameters of Fish treated with 2 mg/l of propofol during and after anaesthesia.

	Control	Anaesthesia	Post anaesthesia
PCV (%)	22.17±1.3	24.8±0.98	26.00±1.53
Hb (g/dl)	8.38±1.10	9.1±1.06	8.90±1.77
RBC (x10 <sup>6</sup> /μl)	1.64±0.36	1.8±0.15	2.00±0.32
WBC (x10 <sup>9</sup> /l)	91.0±21.04	89.6±24.00	124.40±33.93
MCV (fl)	133.15±18.12	139.1±14.16	132.30±7.00
MCH (pg)	50.33±8.41	51.2±6.71	45.30±6.95
MCHC (g/dl)	38.35±1.82	36.7±2.97	34.13±3.84
NEUT (%)	24.67±1.03	26.3±2.07	31.50±3.89**
LYM (%)	69.67±1.75	68.2±2.79	64.33±3.67*
EOSINO (%)	2.17±1.72	1.5±1.22	1.50±1.22
MONO (%)	2.67±0.52	3.2±2.23	2.33±1.03
BASO (%)	0.0±0.00	0.0±0.00	0.00±0.00

\*Show mean difference is significant with anaesthesia and post anaesthesia values compared to the control at  $P < 0.05$ .

\*\* Show mean difference is significant with anaesthesia and post anaesthesia values compared to the control at  $P < 0.01$

Table 2: Blood chemistry parameters of fish treated with 2 mg/l of propofol during and after anaesthesia

Group	Control	Anaesthesia	Post anaesthesia
AST (μ/l)	31.50±8.76	37.67±11.31	18.33±6.61
ALT (μ/l)	35.83±16.46	34.83±27.97	28.33±12.24
BIL (μ/l)	4.15±0.58	4.52±1.83	3.75±0.46
ALB (g/dl)	3.07±0.93	2.85±1.03	3.18±0.94
TP (g/dl)	3.12±1.03	3.80±1.35	2.38±0.75
UREA (mmol/l)	10.00±1.67	14.83±6.05	9.83±1.83
ALP (μ/l)	26.72±24.69	25.58±24.78	43.10±11.89

There was no significant ( $P > 0.05$ ) in mean differences in the fish during and after anaesthesia compared to the control

### Haematological and Blood Chemistry Analysis:

The results of the haematological and the blood chemistry analysis (Tables 1 and 2) show no significant differences in the means when compared to the control except for that of the neutrophils and lymphocyte that shows significant difference of  $p \leq 0.01$  and  $p \leq 0.05$  respectively (Table 1).

At the end of the experiment fish were left in a 50 litre water tanks for 72 hours with no changes in behaviour or mortality.

### DISCUSSION

An ideal anesthetic for aquacultures should have the following qualities; rapid induction without hyperactivity, gradual recovery, absence of residues and toxicity, rapid metabolism and excretion of the anaesthetic [5, 15], thus it must be effective, safe and economical, the ability to produce a state of anesthesia in a period less than or equal to three (3) minutes and recovery of normal swimming excitation in less than ten (10) minutes [5].

In this study, anesthesia was achieved in 5.3±0.74 minutes at 2 mg/l of propofol, higher than the recommended but similar to that reported for Rainbow Trout (*Oncorhynchus mykiss*) 4.99±1.07 by Guillermo [16] at 2.5 mg/l of propofol.

Regarding recovery time, in this study at 2 mg/l of propofol, it was recorded as 3.2±0.48 minutes which was significantly lower than the recommended and similar to that reported for Rainbow Trout (*Oncorhynchus mykiss*) 3.59±1.44 by Guillermo [16] at 2.5 mg/l of propofol.

Both haematological and biochemical blood indices are proved to be valid measure of fish health status and also as indicators of physiological stress resulting from endogenous or exogenous alterations in fish [9]. However, reports regarding effect of propofol on African cat fish blood indices are scarce.

From this study, the results of the haematology indices, serum biochemistry and serum enzymes, shows that propofol does not have significant effect on the means of the blood profile during anaesthesia and 24 hours post anaesthesia when compared to the control, except for the neutrophil with significant increased percentage and absolute counts ( $p \leq 0.01$ ) and leukocyte with significant decreased percentage and absolute counts ( $p \leq 0.05$ ). This result agrees with that reported by Peyghan [12] that propofol prevents peak of cortisol levels and preserves hematological, morphological and biochemical stability in Grass carp.

## CONCLUSION

From this study, it can be said that propofol has the properties of an ideal anesthetic for aquaculture as the induction and recovery times were kept within that required time and besides being safe without adverse effects after 72 hours post administration.

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