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Haematological Response of the African Catfish, *Clarias gariepinus*, to Transportation Stress.

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Abstract

A total of twenty-four (24) adults of the African Catfish, *Clarias gariepinus* (mean Weight 346.25±3.61g and mean total length 36.65±3.91cm) were transported from the hatchery over varying kilometers. They were grouped into six (6); each group had four (4) fish which were further subdivided into two (2) replicates. Group one (1) served as the control; while groups 2,3,4,5 and 6 were transported over 20, 40, 60, 80 and 93 kilometers respectively. Haematological indices of the fish were studied at the various kilometers of transportation. The study revealed progressive decrease in haematocrit (mean of between 16.75 ± 7.41 and $31.50 \pm 1.78\%$), haemoglobin (mean of between 7.60 ± 1.7 and 10.50 ± 0.80g/dl) and erythrocyte values (mean of between $1.43 \times 10^6 \pm 0.72$ and $2.09 \pm 0.14 \times 10^6$ cells/mm^3) as transportation progressed. Leucocyte count also decreased significantly with distance (mean of between $0.90 \pm 0.40 \times 10^6$ and $1.74 \pm 0.29 \times 10^6$ cell/mm^3). Values of these parameters in the control fish were $34.25 \pm 2.84\%$, 11.50 ± 0.80 g/dl, $2.56 \pm 0.47 \times 10^6$ cells/mm^3 and $2.06 \pm 0.13 \times 10^6$ cells/mm^3 respectively. No mortality was recorded in any of the groups on transportation. The physico-chemical parameters of the transporting water were monitored during the study. The implication of this study with respect to aquaculture management is highlighted.

Keywords: Haematology, *Clarias gariepinus*, and Transportation.

1.0 Introduction

Commercially important fish species are often exposed to various environmental stresses in their lifetime. Examples of such common stresses include: water temperature change (Mcleay, 1975), netting (Barton *et al*., 1980), transporting (Schreck, 1981), decrease in water level (Thomas and Robertson, 1991), natural and artificial reproduction (Hlavova, 1992), Fish handling (Wurts, 1995) etc.

Haematological parameters have been employed to assess effects of stressors such as chemical and heavy metal pollution (Annune and Ahuma, 1998; Mgbenka *et al.*, 2003) bacterial infection (Ezeri, 2001) and stress due to industrial effluents (Mcleay and Gordon, 1977; Casillas and Smith, 1977) on various species of fish. Environmental or physiological alterations can be determined by changes in blood parameters, and these are easy to measure, and can provide an integrated measure of the physiological status of the organism.

Clarias gariepinus, the African Catfish has witnessed an increasing interest in aquacultural science in the continent of Africa. This is as a result of its unique characteristics which include its having favourable food conversion, its resistance to diseases, its relatively low requirements for water quality, the possibility for high stocking densities and its excellent meat quality. Comparatively, *Clarias* species are much more valued in Nigeria although the expansion of the culture has suffered constraints due to scarcity of the fingerlings. The need for this fish in fish farms has warranted transport of the juveniles from the hatcheries over long distances to fish farms.

This study focuses on the effects of transportation on the fish, with special interest on the haematology of the fish. Haematological parameters are good indicators of stress and hypoxia (*Casillas* and Smith, 1977), and it has been reported to be a tool for assessing the health of fish.

2.0 Materials and Methods

2.1 Collection of specimen

Twenty- four (24) specimens (adult fish) (mean weight 346.25+ 3.61g and mean total length 33.65+ 3.91cm) were obtained from hatchery located at African Regional Aquaculture Centre (ARAC), Aluu, Port Harcourt, River State. They were divided into six (6) groups of four (4) fish per group. Each group was further subdivided into two (2) replicates of two (2) fish per replicate. The groups were transported at considerable distance of 20km, 40km, 60km 80km and 93km towards Owerri, Imo State for groups 2,3,4,5 and 6 respectively. Group one (1) which served as the control was left in the transporting water at the hatchery.

2.2 Preparation for transport

Prior to transport, the fish were kept in the storage pond at ARAC, Aluu, for twenty-four (24) hours without feeding to empty the digestive tract and to acclimatize them to the condition of transport (Kumar, 1992). They were then loaded in the plastic fish transport containers of volume 0.010898m^3.

Transportation was done using the open system method in which live fish are transported in open containers without facilities for mechanical aeration (Kumar, 1992).

2.3 Stocking density of fish

The stocking density of fish in the containers was as follows: Group 1 (The control) - Replicates a and b had the stocking density of 79.83kg/m^3 and 77.08kg/m^3 respectively. Group 2 had stocking density of 72.49kg/m^3 and 72.49kg/m^3 respectively for the two replicates. Group 3 had stocking density of 61.48kg/m^3 and 62.52kg/m^3 respectively while Group 4 had stocking density of 43.13kg/m^3 and 43.53kg/m^3; Group 5 had stocking density of 45.23kg/m^3 and 48.63kg/m^3. Finally, Group 6 had stocking density of 80.58kg/m^3 and 78.70kg/m^3 respectively. Groups 2, 3, 4, 5 and 6 were transported over 20km, 40km, 60km, 80km and 93km respectively from the pond site towards Owerri, Imo State. At each stipulated distance blood was collected from the various replicates for analysis.

2.4 Blood sampling and analysis

Blood was collected from the fish when desired by the cardiac puncture method described by Svobodova *et al*., (1999).

Blood samples were analyzed thus for the various parameters: Haematocrit was determined by the microhaematocrit method in which the capillary tubes filled with blood samples were centrifuged at 10,000 revolution per minute (r.p.m.) for 5 minutes as described by Wedemeyer and Yasutake (1977). Haemoglobin levels were determined using the cyanmethaemoglobin method which makes use of a spectrophotometer at wavelength of 540nm (Blaxhal and Dasiley, 1973). Erythrocyte (RBC) count was obtained using improved Neubauer counting chamber (haemocytometer) after diluting the blood (1:200 dilution factor) with formal citrate solution. Similarly, Leucocyte (WBC) count was obtained by diluting the Blood (1:200 dilution factor) with Turks's solution followed by enumeration using improved Neubauer counting chamber as described by Barham *et al*., (1980).

Finally, the physico-chemical parameters of the storage and transporting water were determined prior to and at the end of transportation. Water temperature was determined using the mercury-in-tube thermometer, pH by the use of LAB-TECH pH meter; dissolved oxygen (DO) was determined using a Horiba 10 DO meter. Ammonia content was determined titrimetrically.

2.5 Data analysis

All data were analysed by one-way Analysis of Variance (ANOVA) at 5% level of significance. Results were recorded as means ± standard error using students' t-distribution at 95% confidence limit.

3.0 Results

There was no mortality in any of the groups in this study. Results of the haematological indices of the fish prior to transport (the Control) revealed high values for all parameters.

Haematocrit mean was 34.25 ± 2.84%, Haemoglobin mean 11.50 ± 0.80g/dl while erythrocyte count mean was 2.56 ± 0.47 x 10^6 cells/mm^3 and leucocyte count mean was 2.06 ± 0.13 x 10^6 cells/mm^3. A decrease in the haematological parameters was recorded (fig I) in fish at 20km distance. The haematocrit at this distance did not vary significantly ($P>0.05$) compared to the control (mean = 31.50 ± 1.78%). On the other hand, erythrocyte and leucocyte counts differed significantly ($P<0.05$) with the control, (means were 2.09 ± 0.14 x 10^6 cells/mm^3 and 1.74 ± 0.29 x 10^6 cells/mm^3 respectively. (fig II). At 40km and 60km distance, (Fig I), haematocrit and haemoglobin values decreased significantly ($P<0.05$), mean values were 29.25 ± 1.32% and 9.83 ± 0.68/dl respectively. At 80km distance, hac.natocrit, haemoglobin and Leucocyte values decreased very significantly (mean values were 16.75 ± 7.41%, 7.60 ± 1.70g/dl and 0.90 ± 0.40 x 10^6 cells/mm^3 respectively. An appreciable decrease in the erythrocyte count was recorded at this point (mean 1.43 ± 0.72 x 10^6 cells/mm^3).

Haematorit, haemoglobin, leucocyte and erythrocyte counts decreased significantly at 93km distance (Fig I and II); mean values were 24.25 ± 7.67%, 9.18 ± 2.10g/dl, 1.24 ± 0.30 x 10^6 cell/mm^3 respectively.

Results of the physico-chemical parameters (Table 1) revealed high values of the PH (7.62), ammonia (3.5mg/l), and dissolved oxygen (DO) (9.90mg/l) in the storage pond. The values in the control were 6.00; 1.20mg/l and 5.60mg/l respectively. Water temperature in the storage pond and the control, was 23^{o}_{c} and 27^{o}_{c} respectively. A progressive increase in water temperature was recorded as the distance increased. Minimal increase in the pH was also recorded at the sampling points (Table II). There was significant increase in the ammonia and dissolved oxygen content in the transporting water at the sampling points. Mean range was between 1.8 and 2.4mg/l for ammonia, and 5.90 and 10.80gm/l for D.O.

Table I: The physico-chemical parameters of the storage pond and transporting water before transport

PARAMETERS	STORAGE POND		TRANSPORTING WATER (CONTROL)	
	Range	Mean ± S.E.	Range	Mean ± S.E
Temperature (°C)	21 – 26	23.0 ± 4.06	25-78	27.0 ± 1.95
PH	7.57 – 7.67	7.62 ± 0.07	5.5 – 6.4	6.00 ± 0.46
Ammonia content (mg/l)	3.48 – 3.52	30.50 ± 0.03	1.00– 1.30	1.20 ± 0.19
Dissolved Oxygen (mg/l)	9.78 – 10.02	9.90 ± 0.14	5.10 – 5.60	5.40 ± 0.30

N = 4

S.E = Standard error

Table II: The physico-chemical parameters of the transporting water after transport

Parameters	20km	40km	60km	80km	93km
Temperature (^oC)	27.5	28.0	28.0	29.0	29.5
PH	6.23	6.16	6.18	6.23	6.32
Ammonia content (mg/l)	2.10	1.80	1.80	2.10	2.40
Dissolved Oxygen (mg/l)	5.90	8.80	8.70	10.80	9.90

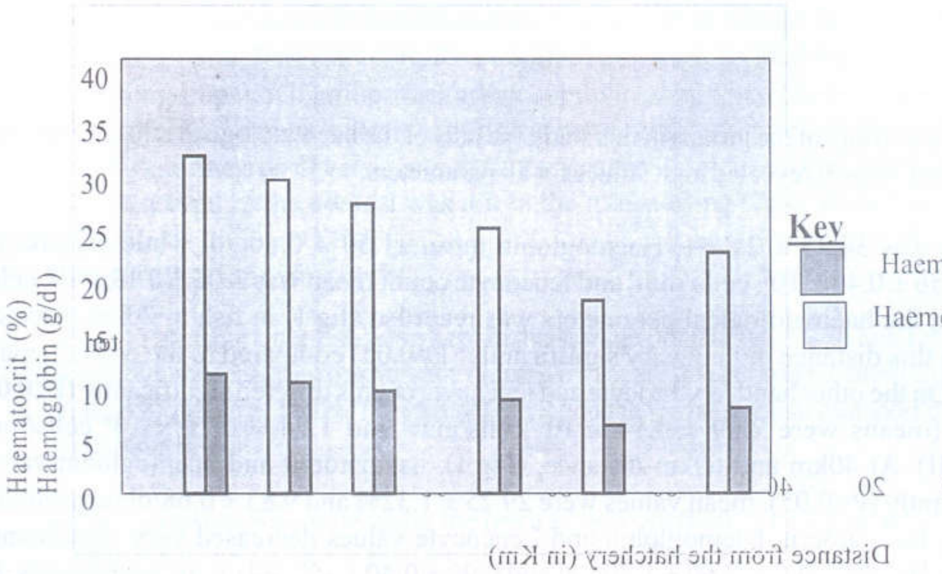

Fig I: Haematocrit and Haemoglobin value (means) in *clarias gariepinus* at various distrances from the hatchery.

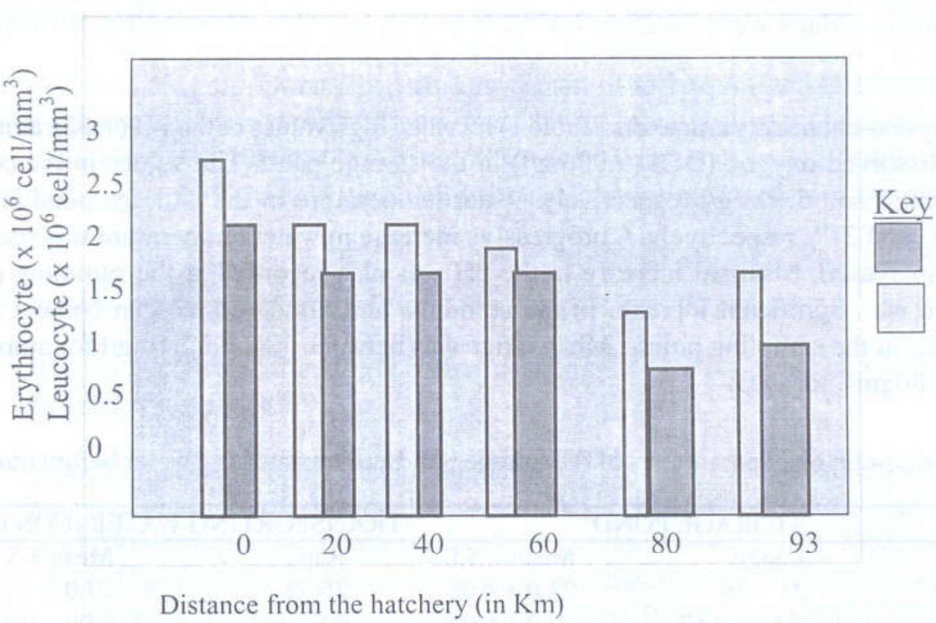

Fig II: Erythrocyte and leucocyte indices (means) in *clarias gariepinus* at various distances from the hatchery.

4.0 Discussion

Lack of mortality among the groups confirms the hardiness of *clariaś gariepinus*.

Data obtained show high values of haematocrit, haemoglobin, erythrocyte count and leucocyte count in the control fish. The haemoglobin, erythrocyte count and leucocyte count progressively decreased with distance, being lowest at kilometer 80. Decrease suggests destruction of red blood cells (haemolysis) which will result to anaemia. Changes in erythrocyte counts are considered useful indicators of haemodilution or haemoconcentration, although anaemia or stress polycythemia may at times occur (Solvio and Oikari, 1976; Ejiogu, 2006). Leucocyte count of the control fish was the highest. There was a high significant decrease ($P < 0.05$) of the number at 80km distance.

The leucocyte values at 20km, 40km and 60km showed no significant difference ($P > 0.05$) but were significantly different when compared with values at 80km and 93km. This indicated case of leucopenia due largely to lowered leucocyte number. Leucopenia is a general consequence of stress (Ellis, 1981; Wedemeyer and McLeay, 1981) and has been found to be a significant part of the physiological response to acute stressors such as crowding (McLeay and Gordon, 1977). A consequent effect of leucopenia in the fish is the breaking down of the bodily defence mechanism which makes the fish prone to infectious diseases. High risk of outbreak of diseases due to transportation stress has been affirmed (Kumar, 1992). A sick fish is also an easy prey to predators.

The physico-chemical status of the transporting water could constitute stressor to the experimental fish. Handling of fish normally causes the initial stress. Storage pond had the lowest temperature, minor change in temperature was recorded during transportation but this was within tolerant limit for *clarias gariepinus*. Temperature nevertheless plays a vital role in the health status of fish. McLeay (1975) reported a lymphopenic response in fish exposed to high temperature and crowding stress. Temperature recorded here was not high enough to expect such effect. Increase in pH recorded at the sampling points is conducive for breeding of fish. Alkalinity is preferred to acidity. Increase in dissolved oxygen and ammonia as recorded in this study favours existence of fish. The authors are inclined to state that the physico-chemical parameters of the transporting water did not constitute stress to the experimental fish.

Knowledge of the extent of these effects is particularly useful in fish farming. The normal practice is transporting juveniles from hatcheries to fish farm over long distances and these juveniles are more susceptible to stress than the adults used for this study. This probably accounts for the high mortality often encountered when fish farmers transport juveniles over long distances to their fish farms. The effects of jolting on fish during transportation need to be investigated. These findings point to the need for the Nigerian Government/Individuals/NGOs to establish hatcheries very close to fish farms they are supposed to serve, and for fish farmers to endeavour to procure juveniles from such hatcheries to reduce mortality as well as other stressors resulting from transportation.

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