https://doi.org/10.33451/florafauna.v29i1pp129-134
FLORA AND FAUNA ISSN 2456 - 9364 (Online)
2023 Vol. 29 No.1 PP 129-134 ISSN 0971 - 6920 (Print)

Haematological and Biochemical Anomalies in Catfish, *Clarias batrachus* due to Cutaneous Ulcerations

Haidar Ali and Samreen Ansari

Department of Zoology,
Government Degree College,
LENGRI GULAR, SHRAVASTI (U.P.) INDIA
*Corresponding Author:
E. mail.: haidarali.zoology@gmail.com

Received **:** 25.03.2023; ***Accepted*** : 08.05.2023

ABSTRACT

The present study was carried out to investigate the haematological and biochemical parameters in healthy and ulcerated Catfish, *Clarias batrachus.* The haemorrhagic necrotic skin lesions in ulcerated fishes showed significant reduction (P<0.01) in RBC, haemoglobin, haematocrit (PCV) eosinophils and lymphocytes. The derived erythrocyte indices, namely, the MCV, MCH and MCHC of ulcerated fishes were also significantly decreased (P < 0.05) from those of normal fishes. Whereas, the total leucocyte count (TLC), neutrophils, monocytes and ESR were significantly increased (P<0.01 and P<0.05) in infected fishes. The biochemical analysis showed a significant reduction (P<0.01) in bilirubin content, whereas the blood glucose, SGOT and SGPT were significantly increased (P<0.01).

Figure : 01 References : 23 Table : 01
KEY WORDS : *Clarias batrachus*, Cutaneous ulcerations, Haematology

Introduction

Fishes living in water bodies near human inhabitants *i.e.* canals, ponds, ditches *etc.* are prone to diseases because, such water bodies get easily polluted from unused food, sewage and decomposing organic matter, leading to rapid unhygienic conditions. Oxygen depletion in such water is also common. Abrupt fluctuation in temperature is frequent and also alteration of pH ranges between extreme limits from strong acidic conditions to strong alkaline ones. All these contribute to creating conditions favourable to the quick spread of diseases in fish.

Fish production is not only important economically but also for food security and social development in many countries, so the widespread disease resulting from an interaction between pathogen, host and environment should be handled to overcome this problem.[13]

Several haematological parameters, including leucocrit, are considered promising as indicators of the physiological status of fish in the natural environment. The enumeration of circulating blood cells, particularly lymphocytes, thrombocytes and neutrophils, would appear to be a more sensitive tool[9]. The importance of haematology in the diagnosis of fish diseases and for the assessment of the effects of pollution has been widely accepted. Certain waterborne diseases of fishes is limiting factor in the commercial exploitation of freshwater fish worldwide, including both food and ornamental species[19].

The dermal infection rates increased with the increase in environmental temperature and organic pollution. Skin lesions cause various biochemical and immunological changes in the serum[23].Cutaneous ulceration in fish is a bacterial disease caused by the bacteria, *Haemophilus piscium,* especially in salmonid fishes[15].The changes in haematological parameters occur due to epizootic ulcerative syndrome, seasonal variation and habit and habitat of fishes[17]. The present investigation aims to study the alteration in the haematology and biochemistry of Catfish, *Clarias batrachus* inhibiting in local polluted waterbody *i.e.* Suvaon Nalla.

Materials and Methods

The normal and ulcerated *Clarias batrachus* (Fig.1) were collected from Suvaon Nalla, Balrampur and kept separately in plastic aquaria. Mildly affected fish had slightly haemorrhagic areas with surface erosion of the skin but severely affected fish had hemorrhagic necrotic muscular lesions on the body. Blood samples for haematological and biochemical analyses were taken

TABLE 1: Changes in haematological and biochemical parameters of *Clarias batrachus* affected with cutaneous ulcerations

Blood Parameters	Normal fishes		Ulcerated fishes		Deviations (%)	t-test
	Range	Mean ± SD	Range	Mean ± SD		
RBC count ($\times 10^6/mm^3$)	1.30-1.41	1.38 ± 0.18	0.60 – 0.72	0.68 ± 0.02	-50.72%	28.15**
TLC count ($\times 10^3/mm^3$)	9.13 – 10.42	10.28 ± 0.09	13.12 – 15.01	14.15 ± 0.42	+ 37.64%	18.12**
Haemoglobin %	8.92 – 10.40	9.80 ± 0.73	5.75 – 7.0	6.75 ± 0.32	- 31.12%	14.18**
Haematocrit (PCV)%	23.0 – 26.5	25.0 ± 0.23	20.18 – 23.14	22.0 ± 0.18	-12.0%	13.25**
MCV (μmm^3)	150.2 – 162.3	158.4 ± 48.7	130.4 – 150.8	138.4 ± 12.5	-12.62%	8.18*
MCHC%	22.2 – 26.4	24.5 ± 6.2	20.8 – 23.4	22.4 ± 1.3	-8.57 %	7.25*
MCH (pg)	9.0 - 9.92	9.6 ± 2.8	7.90 – 8.50	8.30 ± 0.12	- 13.54%	8.14*
ESR (mm/hr)	1.50 - 1.62	1.58 ± 0.18	1.70 - 1.82	1.78 ± 0.11	+12.65%	11.18**
Clotting time (second)	120.0 – 124.0	122.0 ± 0.17	124.0 – 126.0	125.0 ± 0.12	+ 2.45%	1.11
Lymphocytes %	85.0 – 92.0	88.0 ± 2.28	70.0 – 73.5	72.0 ± 4.80	- 20.45%	13.25**
Monocytes %	2.75 – 3.15	3.00 ± 2.11	5.8 – 6.8	6.6 ± 1.76	+ 120.00 %	18.25**
Eosinophils %	2.80 – 3.20	3.0 ± 1.80	1.80 – 2.14	2.4 ± 1.80	- 20%	12.75 **
Neutrophils %	5.58 – 6.32	6.00 ± 0.48	18.12 – 21.4	20.00 ± 1.98	+ 233%	35.12**
Bilirubin	0.52 – 0.59	0.57 ± 0.31	0.48 – 0.55	0.51 ± 0.29	- 10.52%	8.14**
Blood Sugar (mg/ml)	35.45 – 44.12	39.03 ± 0.28	40.12 – 46.12	44.70 ± 0.21	+ 17.08%	11.14**
SGOT (unit/gm)	63.12 – 66.18	65.08 ± 0.41	66.1 – 70.08	68.03 ± 0.05	+ 4.53%	2.14
SGPT (unit/gm)	31.12 – 33.71	32.38 ± 0.41	34.14 – 38.18	36.11 ± 0.21	+ 11.51%	9.32**

* = significant ($P < 0.05$);** = significant ($P < 0.01$)

from the caudal vein and collected in a heparinized tube and then stored in polyethene cool bags until analysed.

The haematological and biochemical parameters of both ulcerated and healthy *Clarias batrachus* were determined[7]. The mean and standard deviation of various haematological and biochemical parameters were calculated with the help of standard formula[4] between experimental fishes and their respective control one P<0.01 and P<0.05.

Results

In the present study, total 16 apparently healthy fishes and 18 diseased *Clarias batrachus* were sampled. Statistical analysis showed that the haematological and biochemical profile of healthy fish was significantly different from those of the ulcerated fishes.

Comparisons of haematological parameters of normal/ healthy and ulcerated fishes are presented in (Table-1). The values of RBC count, Hb, PCV, MCV, MCH and MCHC were 1.38 × 10**6**/mm**3**, 9.8%, 25.0%, 158.4 µm**3**, 9.6 pg and 24.5%, in normal fishes but 0.68×10**6**/mm**3**, 6.75%, 22.0%, 138.4µmm**3**, 8.30pg and 22.4% in ulcerated fishes, respectively.

The results showed that the primary erythrocytes indices, namely, the mean RBC counts, haematocrit levels and haemoglobin concentrations, were significantly low ($P < 0.01$) in ulcerated fishes than in the healthy/normal fish. However, derived erythrocyte indices, namely, the MCV, MCH and MCHC of ulcerated fishes were also significantly decreased ($P < 0.05$) from those of healthy/normal fishes. The ESR, clotting time and leucocyte count were 1.58 mm/hr, 122.0 seconds and 10.28 × 10**3**/mm**3** in normal fishes whereas 1.78 mm/hr, 125.0 seconds and 14.15 × 10**3**/mm**3** in ulcerated fishes. The ESR ($P < 0.01$), clotting time and total leucocyte count ($P < 0.01$) of ulcerated fishes were significantly higher than in the healthy normal fishes, changes in the distribution of individual leucocytes *i.e.* in DLC in the ulcerated fishes were detected. There was a significant increase in the percentage of neutrophils (233%; $P < 0.5$) and monocytes (120.0%; $P < 0.05$) when compared with the healthy or normal fish. The percentage of lymphocytes (20.45%; $P < 0.01$), and eosinophils (20.00%; $P < 0.01$) present in the blood of ulcerated fishes were found to be lower than in healthy/ normal fishes.

Comparison of biochemical parameters of normal/ healthy and ulcerated *Clarias batrachus* are presented (Table-1). The concentration of bilirubin, blood glucose level, Serum Glutamic Oxaloacetate Transaminase (SGOT) and Serum Glutamic Pyruvate Transaminase (SGPT) levels were 0.57 mg/ml, 39.03 mg/ml, 60.08 unit/g and 32.38unit/g in normal *Clarias batrachus* but 0.51 mg/ml, 44.70 mg/ml, 68.03 unit/g and36.11 unit/g in ulcerated fishes respectively.

Thus the bilirubin was significantly decreased (10.52%; $P < 0.01$) in ulcerated fishes. The concentration of Blood Sugar (17.08%), SGOT (4.53%) and SGPT (11.51%) were significantly increased in ulcerated fishes in comparison to healthy ones.

Discussion

The result of the present investigation indicates that cutaneous ulcerations in *Clarias batrachus* have produced alterations in the haematological and biochemical parameters. The results showed that haemorrhagic necrotic skin lesions in diseased fishes significantly reduced the primary erythrocytic indices, namely, total erythrocyte count, haematocrit and haemoglobin concentration, leading to anaemia. Anaemia could be attributed to increasing destruction or loss of erythrocytes and/or suppression of erythropoiesis. It may be due to haemodilution caused by disturbed osmoregulation. The significant reduction in RBCs count, Hb value and PCV in severely affected *Clarias batrachus* is consistent with the previous report on endoparasite-infected *Clarias garipienus*[18] and various parasites infested Bagrids[14]. If the erythrocyte destruction rate is increased without suppression of erythropoiesis, it may affect the proportion of immature and smaller erythrocytes. This should be reflected by a decline in the derived erythrocytes indices, MCV, MCH and MCHC because younger and immature cells are smaller and contain less haemoglobin. Thus significant reduction in derived erythrocytic indices of ulcerated fishes in the present study proved that reductions in the primary erythrocytes indices in ulcerated fishes is due to an increased proportion of immature erythrocytes. Another reason for the anaemic condition may be haemodilution which seems to be brought about by the loss of body fluids from advanced haemorrhagic/necrotic lesions in severely affected fish. However, suppression of erythropoiesis in the severely affected fish can not be ruled out. The decreased oxygen-carrying capacity of the blood due to anaemic conditions may be a contributing factor in producing mortalities observed in *Clarias batrachus* with severe ulcerated lesions. Anaemia also observed in severely affected *Clarias batrachus* is in accordance with the previous report on epizootic ulcerative syndrome affected, snake-headed fish, *Ophiocephlus striatus*[22], Asian cichlid fish, *Etroplus suratensis*[16], Cutaneous ulcerated catfish, *Clarias batrachus*[23], Myxosporean parasite, *Myxobolus* infected major carp, *Cirrhinus mrigala*[1], Epizootic Ulcerative Syndrome infected *Channa striatus*[17] and also in various

Clarias batrachus- Normal

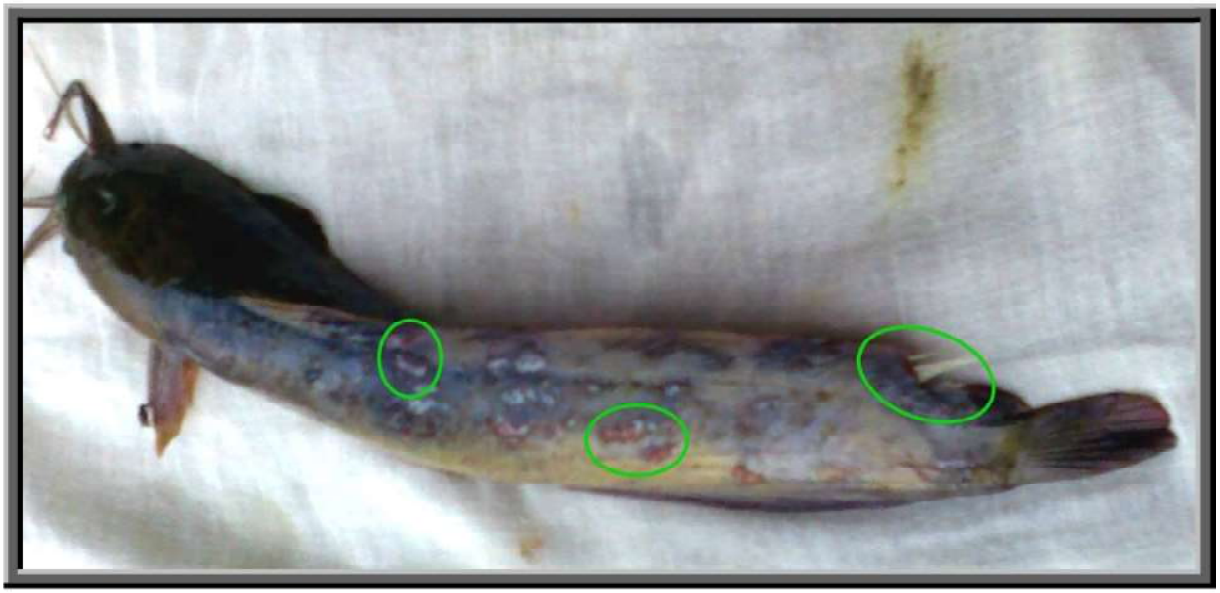

Fig. 1 : *Clarias batrachus* with cutaneous ulcerations

parasites infested Bagrids, *Bagrus bayad* and *Bagrus docmak* [14].

Total leucocyte counts of the ulcerated *Clarias batrachus* indicate leucocytosis (*i.e.* increase in TLC). The percentage distribution of different types of leucocytes (*i.e.* DLC) in ulcerated fishes showed significant changes from the normal distribution pattern. The percentage of neutrophils and monocytes was found to have increased significantly along with a marginal decrease in the percentage of lymphocytes and eosinophils. The elevation of TLC in fish is due to a defense mechanism against parasitic infection[1,12,18]. In fishes, neutrophils may be involved in the cellular immune response and lymphocytes have been regarded as immunocompetent cells involved in humoral immune responses[10]. Numerous reports have described

neutrophil migrations to injury sites, such as those resulting from bacterial infection, parasitic infection and mechanical injury in fishes[16]. Neutrophils have also been observed to be capable of phagocytic activity[11]. The increased percentage of neutrophils in the circulating blood of *Clarias batrachus* with severe ulcerated lesions may be related to their cellular immune function as a response to the local inflammation and increasing damage to the tissues of these fishes. The marginal decrease in lymphocytes in ulcerated fishes may be associated with decreased humoral immune response in fish.

Bilirubin is one of the bile pigments released from the liver. It is an excretory catabolic product of haemoglobin and is excreted out in the urine. A decrease in the amount of bilirubin in ulcerated fishes points to a possible hepatodysfunction *i.e.* malfunction of the liver which causes less secretion of bilirubin into the blood and leads to hypobilirubinemia. Hypobilirubinemia was reported in several fishes inhabiting naturally as well as experimentally stressed fishes[1, 2, 5, 21].

The increase in blood sugar level in ulcerated *Clarias batrachus* may be due to an increase in the breakdown of liver glycogen or due to decreased synthesis of glycogen from glucose. Hyperglycemic conditions in naturally as well as experimentally stressed fishes may be due to impairment in the hormone level in the blood involved in carbohydrate metabolism[2,5,8,20,21]. Inhibition of cholinesterase in the adrenal medulla which stimulates the breakdown of glycogen to glucose and increases corticosteroid level which also increases the blood glucose level in experimentally stressed fishes causes hyperglycemia[3,6]. Thus hyperglycemia in ulcerated *Clarias batrachus* seems to be due to reduced insulin secretion, increased corticosteroid and also stimulation of gluconeogenesis. It is due to the metabolization of glycogen deposits in the liver to the site of their active metabolism for the liberation of energy.

The elevated levels of serum transaminases (SGOT and SGPT), which are markers of liver functions, were observed in ulcerated *Clarias batrachus.* This increased level of serum transaminases is related to disruption of normal metabolism which is due to extensive alterations in the liver cells and indicates liver damage.

Thus, it is clear from the above discussion that ulcer caused by bacteria and environmental stress exert their effect directly or indirectly on fish health by releasing toxicants at the basic level of the organisms by reacting with enzymes or metabolites in enzymatic reaction or by hormonal disorders or by binding and interaction with the membrane structures or other functional component of the cells. It is further speculated that mechanical damage caused by environmental stress, bacteria and other pathogens to the host's skin may also lead to side-tracking of iron to affected tissues which are otherwise responsible for erythropoiesis. An increased number of leucocytes may be associated with the defense mechanism and immunological responses against cutaneous ulceration. So, it can be concluded that cutaneous ulcers influence the health status of fish which was reflected by alterations in the haematological and biochemical parameters of fish.

References

1. Ali H, Ansari KK. Haematological and biochemical changes in Indian major carp, *Cirrhinus mrigala* due to myxosporean parasite, *Myxobolus. Flora and Fauna*. 2012; **18**(2): 253-258.

2. Ali H, Ansari KK, Prakash S. Haematological abnormalities in an exotic carp, *Hypophthalmicthys molitrix*, infested by a fish louse, *Argulus* sp. *Poll. Res.* 2010; **29**(4): 593-595.

3. Bhatia NP, Sandhu GS, Johal MS. Endosulfan induced changes in blood chemistry of *Heteropneustes fossilis. Poll. Res.* 2002; **21**(3): 323-327.

4. Biradar RS. Course Manual Fisheries Statistics. *CIFE,* Bombay. 1988; 92-121.

5. Chaturvedi LD, Agarwal K. Haematological changes in *Heteropneustes fossilis* following exposure to Alachlor and Rogor. *Ad. Biol.* 1993; **12:** 85-92.

6. Chowdhury AR, Rastogi VK, Arora U, Saxena C. Biochemical and morphological alterations of adrenal under exposure of orthochlorobenzlidene malononitrite. *Mikroskopie.* 1985; **42:** 151-156.

7. Dacie JV, Lewis SM. Practical Haematology, 5th edition. J and A Churchill. London.1975.

8. Dalela RC, Rani S, Kumar V, Verma SR. *In vivo* haematological alterations in a freshwater teleost, *Mystus vittatus* following subacute exposure to pesticides and their combination. *J. Environ Biol.* 1981; **2**(2): 79-86.

9. Dick PT, Dixon DG. Changes in circulating blood cell levels of rainbow trout, *Salmo gairdneri* Richardson,

following acute and chronic exposure to copper. *J. Fish. Biol.* 1985; **26:** 475-481.

10. Ellis AE, Munro ALS, Roberts RJ. Defence mechanisms in fish I. A study of the phagocytic system and the rate of intraperitoneally injected particulate material in the plaice (*Pleuronectes platessa* L.). *Journal of Fish Biology.* 1976; **89:** 67-78.

11. Finn JP, Nielson NO. Inflammatory response in rainbow trout. *Journal of fish Biology.* 1971; **3:** 463-478.

12. Movahed R, Khara H, Ahmadnezahad M, Sayadboorani M. Haematological characteristics associated with parasitism in pike perch, *Sander lucioperca* (Percidae) from Anzali Wetland. *J. Parasit. Dis.* 2016; **40**(4):1337-1341.

13. Muktar Y, Tesfaye S, Tesfaye B. Present status and future prospects of fish vaccination: a review. *J. Vet. Sci. Technol*. 2016; **7**(02):299 http://dx.doi.org/10.4172/2157-7579.1000299.

14. Omeji S, Alo JA, Garba, AA. Impactof parasites on haematological and biochemical parameters of selected Bagrid species from lower river Benue Nigeria. *Int. J. Vet. Sci.& Ani. Husb*. 2018; **3**(5): 23-27.

15. Pandey K, Shukla JP. Fish and Fisheries. *Rostogi Publication.,* Meerut, (U.P.) India. 2007; p. 405.

16. Pathiratne A, Rajapakshe W. Hematological changes associated with epizootic ulcerative syndrome in the Asian Cichlid fishes *Etroplus suratensis. Asian Fisheries Science.* 1998; **11:** 203-211.

17. Podeti KR, Benarjee G. Haematological changes in freshwater fish, *Channa striatus* diagnosed with Epizootic Ulcerative Syndrome. *International Journal of Advanced Biotechnology and Research*. 2015; **6**(2): 238-244.

18. Radwan M, Shehata S, Abdelhadi Y, Mohammed R, Mohamed M, Magdy M. Histopathological and biochemical indices of *Clarias gariepinus* (Burchell*,* 1822) parasitized by endoparasitic fauna in fish farm of north eastern Egypt. *Turk. J. Fish. & Aquat. Sci.* 2021; **21**(9):465-478.

19. Sayackkara N, Srivastava PP. Changes in haemato-logical parameters in *Heteropneustes fossilis* affected with cutaneous ulceration. *Proc. Nat. Acad. Sci.,* India. 1997; **67:** 117-119.

20. Shastry KV, Sunita KM. Chronic toxic effects of chromium in *Channa punctatus. J. Environ. Biol.* 1984; **5**(1): 47-57.

21. Srivastava SK, Singh D, Prakash S, Ansari KK. Effect of sublethal concentration of distillery effluent on the haematological and biochemical parameters of *Clarias batrachus* (Linn.). *Ecol. Env. & Cons.* 2007; **13**(3): 511-514.

22. Tangtrongpiros JP, Charoennetisart B, Wongsatayanont B, Tavatsin A, Chaisiri, W. Hematological changes in snakehead fish during 1984 outbreak. *Proceedings of the Technical Conference on Living Aquatic Resources, 7-8 March 1985,* Chulalongkorn Univrsity. Bangkok. 1985.

23. Tripathi I, Ansari KK, Prakash S. Changes in the haematology of a freshwater Catfish, *Clarias batrachus* affected with cutaneous ulceration. *Scitech.* 2007; **2**(1): 91-92.