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HAEMATOLOGICAL RESPONSES OF *CLARIAS BATRACHUS* (LINN.) TO SUBLETHAL CONCENTRATIONS OF LEAD

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ABSTRACT: Blood parameters (Hb%, TLC, TLC, ESR, PCV, MCV and MCH) of the catfish, *Clarias batrachus* (Linn.) were investigated, exposed to different concentrations of lead [Group- A (control), (Group- B (0.5ppm), Group- C (1.0ppm) and Group- D (3.0 ppm)] during different time intervals (7, 14, 21 and 28 days). Haemoglobin values fell to the maximum (11.5%) from its control value (13.0%). TEC values were also reduced (2.90x106/Cumm) during the maximum exposure period (28 days) in groups C and D. On the other hand, TLC increased to the maximum (21,900/Cumm) during 28 days exposure period at 3.00 ppm of lead. ESR values also indicated significant increase (3.10mm/h) at 1.00 ppm of lead during 28 days exposure period. PCV decreased to the maximum (29%) at maximum exposure period (28 days) at intermediate concentration (1.00ppm of lead). MCH increased in all treated groups, but a maximum rise of 40.34pg was recorded at 1.00ppm of lead during 28 days exposure period. Maximum reduction in MCV (100.0µ3) was observed after 28 days exposure period at 0.5 and 1.00 ppm of lead concentration. The above research indicated that the heavy metal lead induces hematological responses in the fresh water catfish, *Clarias batrachus*.

INTRODUCTION: Heavy metals are recognized as one of the most hazardous environmental pollutants, and they are toxic to many living organisms. Environmental exposure to heavy metals has been reported to cause disease in humans and other animals [1]. In recent years there has been a growing interest in the effects of heavy metals on the health of fish [2, 3].

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Lead is one of the most widely used heavy metals that has wide applications in such products as storage batteries, electric cable sheaths, alloys, pesticides, paints, petrol, and rubber products, among other uses [4]. Lead reaches water bodies either as industrial effluents, runoff from agricultural fields or as ores from mines Lead has no recognized function biologically in the body, and thus when it enters the body, it causes serious health effects which might be permanent and lead to a fatality on the growth of fish [5].

Chronic lead poisoning has similar toxic effects in fish as in mammals [6]. Studies carried out with various fish species under laboratory conditions showed that heavy metals cause direct disorders in

morphology, osmoregulation, and secondary disorders by enzymatic and hormonal changes in sera glucose, protein, cholesterol, and blood parameters [7, 8]. These include hematological and neural disorders, clastogenic effects, growth index, proteolysis in kidney, liver, and deformation in gills were reported [9, 10, 11, 12, 13].

Blood parameters have been commonly used to observe and follow fish health since variations in blood tissue of fish are caused by environmental [14]. Many workers reported the effect of individual heavy metal on haematological aspects of fishes [15, 16, 17, 18, 19] & [20]. The objective of the present study is to provide data on the haematological parameters of *Clarias batrachus* treated with lead to work out the toxicity level of the metal under investigation

MATERIAL & METHODS: *Clarias batrachus* (40-50gms) (n=16) of each group (one control, three lead-exposed aquatic medium), collected from different fish markets of Bareilly were acclimatized for 7 days to laboratory conditions. Lead toxicants were prepared in the laboratory by using 1gram pure lead metal, which was dissolved in HNO_3 and diluted up to 1000 ml stock solution by using distilled water. Stock solution 1.0 ml has 1.0 mg of lead concentration. The groups were categorized as follows:

- **Group A:** Control
- **Group B:** Exposed to 0.5 ppm of Pb
- **Group C:** Exposed to 1.00 ppm of Pb.
- **Group D:** Exposed to 3.00 ppm of P

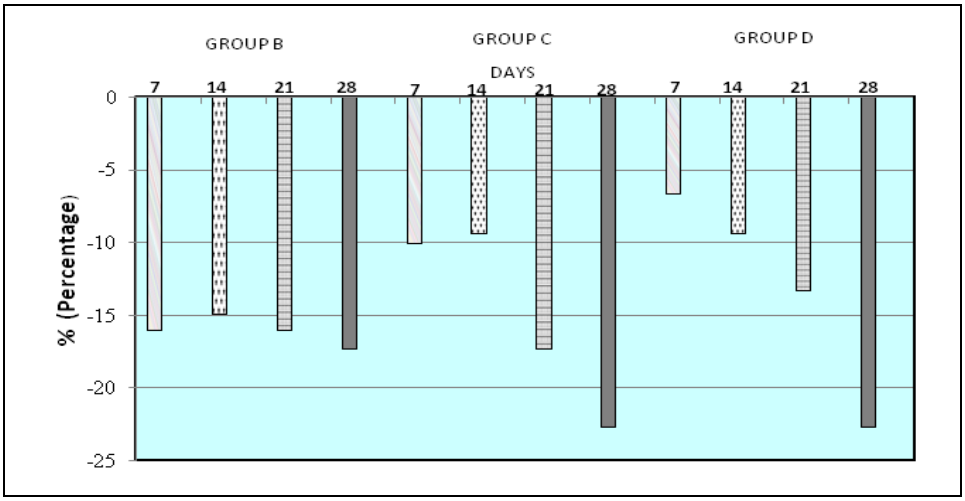
Blood was collected from the caudal vein of experimental fishes with the help of anti-coagulatedsyringe and Hb%, TEC, TLC, ESR, PCV, MCH and MCV were estimated [21].

RESULTS: *C. batrachus* exposed to lead-induced changes in the Hb value. The value fell to a maximum of 11.50% during 21 days in Group B, 28 days in Group C, and 14 days and 21 days exposure period in Group D with maximum percentage changes fall (-11.53%) **Table 1, Graph. 1**. The minimum value of TEC was 2.90x106/Cu mm. (28 days: 1.00 ppm and 3.00 ppm of Pb), and minimum fall was recorded at 3.50x106/Cu mm. (7 days: 3.00 ppm) as compared to the values of the control group with maximum percentage changes *i.e* 22.66% **Table 1**, **Graph 2**. The maximum effect of lead on TLC was 21,900 /Cumm (28 days: 3.00 ppm) as compared to the control value with a maximum percentage change of 62.22% **Table 1**, **Graph 3**. Maximum percentage changes in ESR were 63.15%, with a significant increase in the ESR values 3.10 mm/h at an intermediate concentration of lead during the highest exposure period (1.00 ppm: 28 days) **Table 1**, **Graph 4**. A significant fall in the PCV values (29.00% at 28 days: 1.00 ppm of P) and minimum reduction in the PCV value was 40.00 (7 Days: 3.00 ppm of Pb) with a maximum percentage change of 25.3%. **Table 1, Graph 5**. The effect of lead on MCH of *C. batrachus* indicated minimum value of 33.28 (Pg) (7 days: 3.00 ppm) and 40.34 maximum with maximum percentage changes 16.38% (28 days: Group D) **Table 1; Graph 6**. The maximum value of MCV was shown at 120.63 3 during 7 days post-treatment in group B; the minimum value was 100.00 (3) after 28 days exposure period in groups B and C **Table 1**, **Graph 7**.

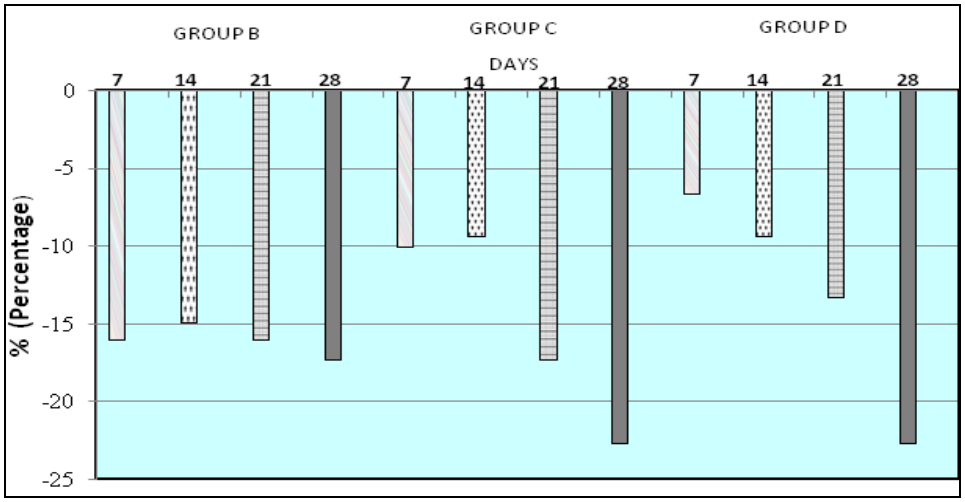
TABLE 1: HAEMATOLOGICAL PARAMETERS OF *CLARIAS BATRACHUS* EXPOSED TO VARIOUS CONCENTRATIONS OF LEAD (PB)

S. no.	Parameters	GroupA (Control	Group B (0.5 ppm Pb)				Group C (1.00 ppm Pb)				Group D (3.00 ppm Pb)			
			Days											
			7	14	21	28	7	14	21	28	7	14	21	28
1.	Hb %	13.00	12.50	12.00	11.50	11.60	12.50	12.50	11.80	11.50	12.00	11.50	11.50	11.70
2.	TECx10^6/Cumm	3.75	3.15	3.19	3.15	3.10	3.37	3.40	3.10	2.90	3.50	3.40	3.25	2.90
3.	TLC /Cumm	13500	14600	17800	17700	19800	13900	20200	21000	21200	13500	16900	19000	21900
4.	ESR (mm/h)	1.90	1.90	2.40	2.90	2.90	2.10	2.60	3.00	3.10	1.90	2.10	2.40	2.80
5.	PCV %	41.50	38.00	35.00	33.00	31.00	39.00	37.00	34.00	29.00	40.00	36.00	35.00	31.00

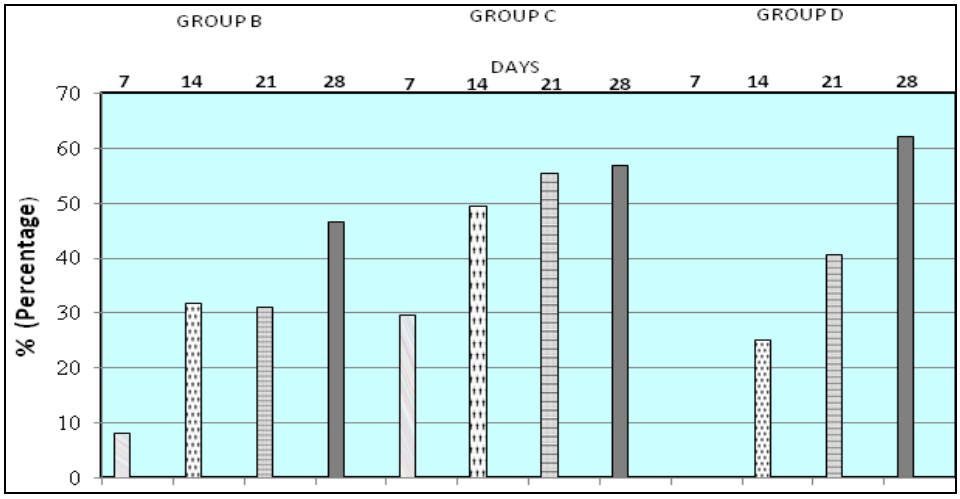
6.	MCH (Pg)	34.66	39.68	37.61	36.50	37.41	37.09	36.76	38.06	39.65	34.28	33.82	35.38	40.34
7.	MCV (μ^3)	110.60	120.63	109.71	104.76	100.00	115.72	108.82	109.82	100.00	114.28	105.88	107.69	106.89



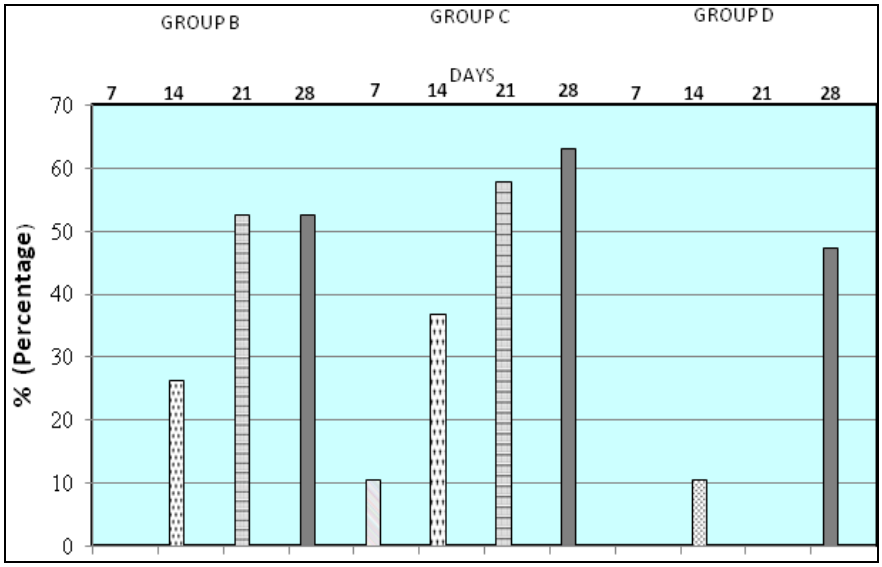
GRAPH 1: PERCENTAGE CHANGES IN HB % OF *CLARIAS BATRACHUS* TO VARIOUS CONCENTRATIONS OF LEAD



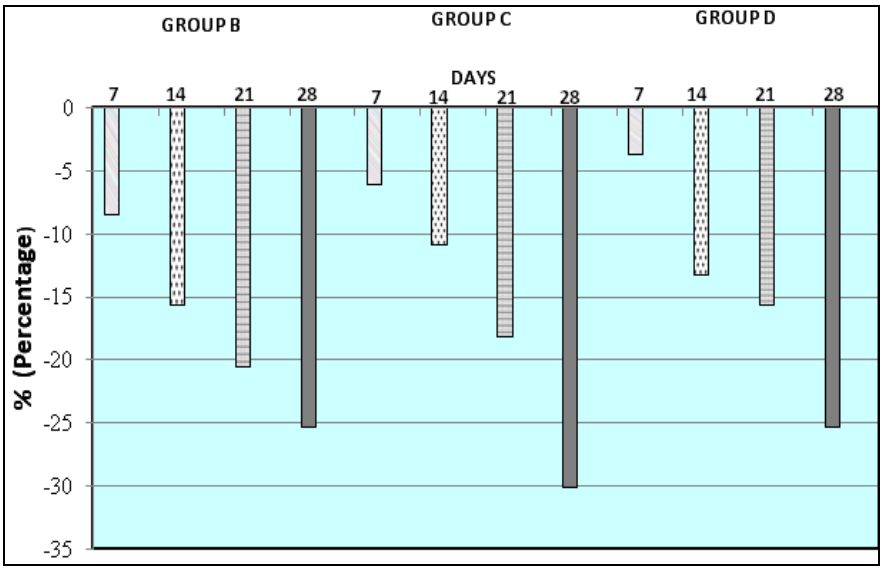
GRAPH 2: PERCENTAGE CHANGES IN TEC/ CUMM OF *CLARIAS BATRACHUS* TO VARIOUS CONCENTRATIONS OF LEAD



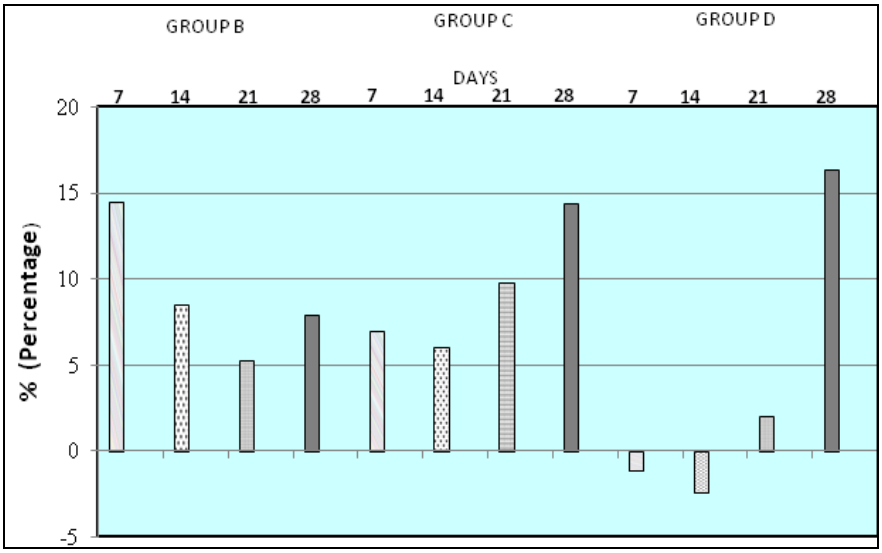
GRAPH 3: PERCENTAGE CHANGES IN TLC/ CUMM OF *CLARIAS BATRACHUS* TO VARIOUS CONCENTRATIONS OF LEAD



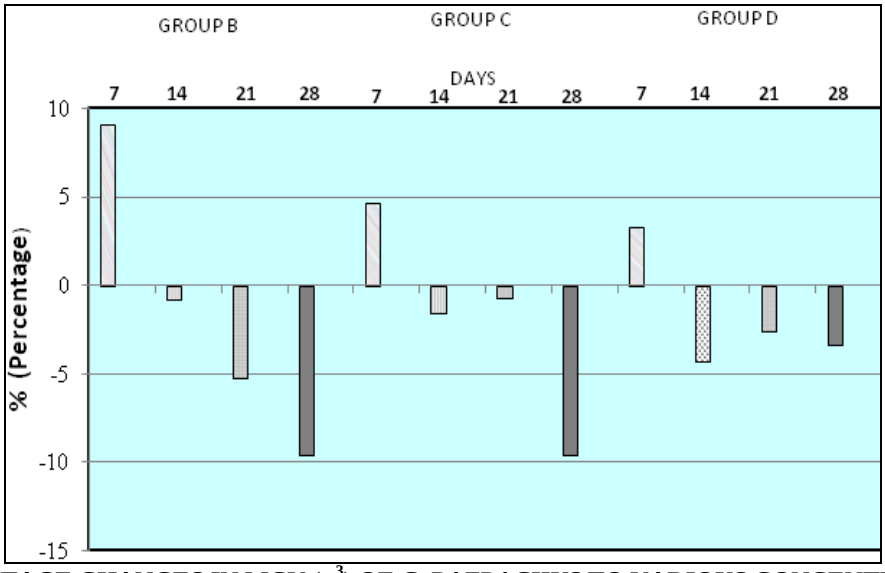
GRAPH 4: PERCENTAGE CHANGES IN ESR (MM/H) OF *CLARIAS BATRACHUS*\ TO VARIOUS CONCENTRATIONS OF LEAD



GRAPH 5: PERCENTAGE CHANGES IN PCV% OF *C. BATRACHUS* TO VARIOUS CONCENTRATIONS OF LEAD



GRAPH 6: PERCENTAGE CHANGES IN MCH (PG) OF *C. BATRACHUS* TO VARIOUS CONCENTRATIONS OF LEAD



GRAPH 7: PERCENTAGE CHANGES IN MCV (μ^3) OF *C. BATRACHUS* TO VARIOUS CONCENTRATIONS OF LEAD

DISCUSSION: The toxic effects of heavy metal on fish are multidirectional and manifested by numerous changes in the physiological and chemical processes of their body systems [22, 23]. In the present study, Hb%, TEC, and PCV decreased while TLC, ESR, MCH, and MCV increased significantly, corresponding to the increase in exposure time and concentration in the blood of *Clarias batrachus*. Haematological indices (RBC count, haemoglobin and haematocrit) have been reported to indicate an organism's secondary responses to irritants as indicated by the researcher who concluded that lead toxicity occurs ionregulatory disruption [24]. On the other hand, TLC counts were reduced in all treated groups due to release of epinephrine during stress causing a decrease in leukocyte counts, which suggests the weakening of the immune system. Lead-induced changes in blood parameters of *Anguilla anguilla* as a consequence of the destruction of energy resources in order to maintain homeostasis [25]. The study suggested that the presence of toxic heavy metals in aquatic environment has strong influence on the hematological parameters in the freshwater fish common carp (*Cyprinus carpio* L. [26].

The haematological changes were more significant in fish exposed to combined metals as compared to individual lead and copper. That can be used as an indicator of metal related stress in fish on exposed to an elevated level of metals in the water, which can postulate the reduction in Hb% and RBC by lead and copper occurs due to destruction of the haemopoetic system, which leads to anaemic condition while a significant increase in WBC may be due to stimulation of the immune mechanism to eliminate the effects of the pollutants in metal exposed fish [20]. The effects of short-term exposure from 2 to 16 h of common carp to a high level of lead showing fluctuation in reduction on the haematological parameters like Hb%, RBC, PCV, MCH, MCV and TLC [27]. *C. gariepinus* exposed to lead chloride, resulting in fall in Hb%, Hct, MCV and MCH and rise in RBC and WBC after 28 days exposure [28]. Decrease in haematological parameters (RBC, WBC and Hb%) at short-term exposure while WBC increased during long term exposure in *Channa punctatus* [29]. Haematological parameters of Cyprinus carpio (RBC, Hb, and Hct) decreased significantly when exposed to sublethal concentration (4.45 ppm) of lead nitrate. But the WBC count showed an increasing trend as the concentration of the metal increased in the medium [30].

The effects of lead acetate on haematological parameters of Bunny fish *Mesopotamichthyes* sharpey, indicating significant decreases in RBC, WBC, Hb%, and PCV values as compared to the control group [10]. Comparative heamatological studies on *Oreochromis niloticus* with Cu and Pb was investigated that lead was more toxic on the blood parameters of fish as compared to copper [31]. Many workers (*Rutilus rutilus* 15, *Cyprinus carpio* [30], [32], *Channa punctatus*and *Clarias gariepinus* [33] reported that lead has a negative impact on the blood parameters of fish which reduces the immune system.

Lead treated *Channa punctatus* showed reduced TEC, Hb, and PCV, probably due to cumulative response of lead toxicity towards excessive erythrocyte destruction. Further, the increase in TLC is due to adaptive response of the fish to lead stress [34, 35]. Haematoxical investigations on of heavy metals on the blood parameters of catfishes (*H. fossilis* and *C. batrachus*) revealed that lead-induced toxicological effects on all the investigated blood parameters of experimental fish [36, 37, 38]. It is thus evident that lead is responsible for inducing haematological aberrations in fish.

CONCLUSION: Exposure of C. batrachus to sub-lethal concentrations of heavy metal, lead resulted in a significant decrease in hemoglobin and TEC content, while an increase was observed in case of TLC. The observations showed that exposure of *C. batrachus* to sublethal concentrations of lead (2.25 ppm), (2.65 ppm), and (2.85 ppm) culminated in increased TLC and ESR values and decreased TEC, Hb and PCV values, whereas changes in MCH, and MCV values were insignificant. Thus, short-term exposure of lead produced a negative response to blood biochemistry of fish.

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