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Comparative study on toxicity of nickel on hematological profile of Grass carp and Silver carp

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Abstract

The current study was aimed to investigate the impact of nickel chloride exposure on the hematological parameters of grass carp (*Ctenopharyngodon idella)* and silver carp (*Hypophthalmichthys molitrix*). Heavy metal pollution is a growing global concern affecting aquatic ecosystems and on inhabitants. In the present study the two fish species were exposed to nickel chloride for duration of 7 and 25 days. Result of Hematological profile, showed significant differences in responses between Grass carp and Silver carp. Grass carp exhibited substantial alterations in white blood cell count , hemoglobin (Hb), red blood cell count , hematocrit, mean corpuscular hemoglobin (MCH), red cell distribution width (RDW), mean platelet volume (MPV), platelet count (PLT), mean cell hemoglobin concentration (MCHC), and plateletcrit (PCT), indicating a strong immune response and physiological changes. In contrast, silver carp showed comparatively lower alterations during the seven-days of treatment in parameters: like mean corpuscular volume (MCV) was 136.7±0.90 fL, platelet distribution was (PDW) 15.83±0.64 %, while Grass carp showed alterations only in (PDW) 51.6±1.38%. For the period of 25 days, silver carp showed alterations in (MCV) and (MCH) 143.6±1.55fL and 28.70±0.11pg, and grass carp showed significant in (Hb) and (RDW-CV) 13.26±0.56gm/dL and 126.8±15.034%. Student‘t’ test was applied for the comparison of the data of control with the test samples. These species-specific responses highlight the importance of considering individual fish species' sensitivities which assessing the effects of environmental stressors like heavy metals.

Key word: *Silver carp, Grass carp, Nickel chloride, Toxicity, Hematolog*

Introduction

Environmental pollution is a huge global issue that poses significant threats to human health (Fereidoun, *et al.,* 2007). In recent decades, the escalation of environmental pollution has sparked increasing alarm for public health (Kimani., 2007). Comparatively, human populations face heightened exposure to environmental contaminants and is more intense now a days (Schell, 2006). Excessive levels of pollution from activities like municipal wastes disposal and combustion of fossil fuels cause extreme loss to wildlife, humans, and plants ecosystem (including tropical rainforests), as well as the broader natural environment (Nriagu ,1988).

There seems to be no way around the fact that polluting waterways is a worldwide issue that has a significant impact on the ability of aquatic organisms to thrive in their natural environments., Several potentially harmful xenobiotic are present in the environments where fish are active (Muniyan and Parthipanand, 2013). Fish are widely regarded as the most reliable ecological indicator of river status (Olaifa *et al*., 2004).

Metals, regarded as significant environmental contaminants, encompass metallic elements characterized by their relatively high density and toxicity, even when present in low concentrations. They are ubiquitously distributed within aquatic ecosystems and are acknowledged for their essential role in trace amounts, facilitating the normal biological functions of aquatic fauna (Khan.,2014). Heavy metals present in dissolved form in freshwater environments are readily absorbed by fish through their gills; as a result, these metals accumulate in fish tissues due to their non-degradable nature, reaching toxic levels in some cases (Malik *et al*., 2010).

The distribution of heavy metals in sediments is affected by a number of processes, which are physical,chemical and biological. Therefore, a thorough investigation of the geochemical and anthropogenic origins, as well as the amounts of metals in the sediments of aquatic systems is necessary. Heavy metal concentrations change throughout time due to the interplay of a number of causes. In freshwater ecosystems, heavy metals can be released from contaminated sediments and accumulate in microorganisms, plants, and animals, eventually causing a wide range of health problems at the lowest trophic level (Haris *et al*., 2017). In recent decades, freshwater pollution from a variety of sources has gained attention as a serious problem (Vutukuru, 2005). Pollution from heavy metals can have severe consequences for aquatic ecosystems and biodiversity

(Farombi *et al*., 2007). Fishes are the aquatic organisms that are completely helpless against the toxic effects of these contaminants (Olaifa *et al*., 2004).

Pollutants accumulate in the food chain create negative effects and mortality in aquatic systems; thus, fish are commonly employed to gauge the state of these ecosystems (Farkas *et al*., 2002). Physiological activities and biochemical markers in tissues and blood have been demonstrated to be affected by heavy metals in fish studies (Basa and Rani, 2003). Bioaccumulation of heavy metals and its harmful effects have been reviewed (Adami *et al*., 2002). In order to ward off the deleterious effects of xenobiotic such as heavy metals and oxidative stress, the organisms evolved a preventive defense mechanism (Abou EL-Naga *et al*., 2005).

Nickel is an essential element for the entire biological process (biota) as lacking this nutrient can cause enzymatic malfunction, but it can also be toxic to aquatic organism (fish) when its concentration is higher in water. Various contaminants enter water bodies via industrial, domestic, and agricultural release systems, causing stress to aquatic life including fish and other aquatic plants and animals. Stress is a generic, non-specific reaction to anything that disrupts the body's natural state of equilibrium. Physiological responses to stress include things like shifts in blood chemistry and the activation of defense mechanisms (Svoboda, 2001; Witeska, 2003).

Fish are utilized as test species in aquatic toxicology because of their place at the top in the food chain and their importance as a human food source (Farkas *et al*., 2002; Yousuf and El-Shahawi, 1999). According to the research done on many fishes (Basa and Rani, 2003; Canli, 1995; Tort and Torres, 1988), heavy metals might alter the biological activities and biochemical parameters in tissues and blood. Waqar (2006), Adami *et al*. (2002), Rasmussen and Anderson (2000), Rani (2000), and Aucoin *et al*., (1999) all provide reviews on the toxic effects of heavy metals, including bioaccumulation. The organisms had a defense against the negative effects of heavy metals, both significant and insignificant, and other xenobiotic that cause progressive changes in the body, such as oxidative stress (Abou EL-Naga *et al*., 2005).

Fish size and weight, as well as the duration and concentration of exposure in water, can affect the toxicity of heavy metals. It is well known that fish suffer from toxic heavy metal exposure, which necessitates detoxification and mending mechanisms (Vosyliene and jankaite, 2006; Zhang *et al*., 2013). The negative effects of toxicants on fish health are patented at different organizational

levels (Zarei *et al*., 2013). The hematological reactions of exposed fish must be exploited as early caution symptoms of contact with chemical toxicants (Zhang *et al*., 2013).

The excessive consumption of both essential and non-essential metals leads to their accumulation in several tissues. Higher metal concentrations can disrupt the normal biological functions of fish (Canli , 2003). When humans consume fishes contaminated with these metals, it can result in severe health complications (Kamaruzzam, 2011). Furthermore, metals can disrupt the ecological balance of aquatic ecosystem (Vinodhini, and Mastan, 2008). Fish, being positioned at the top of the aquatic food chain, have a higher tendency to accumulate metals in their bodies (Rauf and Javed, 2009). Sub-lethal absorption of contaminants, such as heavy metals in fish, can be calculated using hematological factors (Witeska, 2003). Hematological measurements such as red and white blood cell counts, hemoglobin levels, hematocrit percentages, and red blood cell counts were the most commonly taken under stress. Hematological indicators are frequently measured in fish to evaluate their overall health (Oshode *et al*., 2008).). In recent decades, public concern has grown over the widespread pollution of fresh water, which poses a threat to public water sources and can also be harmful to aquatic life. Heavy metals from household, industrial, mining, and agricultural wastes may cause severe pollution in river systems (Vander Oost *et al*., 2003). Within aquatic systems, these metals disperse radially, and fish, being apex predators, are particularly vulnerable to the adverse effects, in contrast to terrestrial vertebrates (Kousar, and Javed., 2014).

The objective of the current study was to examine and compare the toxicity effect of the of heavy metal nickel in the form of Nickel chloride on hematology of Grass carp and Silver carp exposed to varying concentrations.

Materials and Methods

Sample collection

Healthy Grass carp *(Ctenopharyngodonidell*a) and silver carp (*Hypophthalmic molitrix*), were used as experimental animals. Experiments were conducted to assess the toxic effects on Silver and Grass carp under the influence of heavy metal. Fish were collected from Sher-Abad Silver and Grass crap Hatchery, Fisheries Department Peshawar in plastic bags filled with oxygen brought to the lab. Both fish species were treated with 0.2% $KMnO_4$ solution to take off the external pollutant and infection for 2 minutes. All the fish samples were adapted for 1 week in aquarium with daily base 40% changing of tape water. After acclimatization, fish were transferred to new aquarium for

experimentation. In both aquarium they were fed with prawn powder and soya bean meal daily morning (9am) and evening (5pm). Nickel chloride was used as source of nickel.

In the present investigation experimental fishes were divided into two groups. Three (3) fish were kept in the control group and exposed to normal water and three (3) fishes were exposed to nickel added water. In both the control and experimental group fishes were exposed to a maximum of 7days and 25 days.

Blood collection

Hematological parameters were examined by collecting blood samples from metal-exposed and control fish after 7 and 25 days, Blood samples were taken by puncturing the caudal vein with a 23-gauge needle to collect the blood in a 5ml sterile syringe. The blood collected by disposable syringe from caudal vein puncture of Silver carp and Grass cap where kept in sterilized appropriate vials and processed for various hematological analyses by the method of Dacie and Lewis 1975. Blood samples were split in half and kept at 4 degrees Celsius in the fridge. Two sets of frozen blood samples were analyzed separately. Blood samples treated with anticoagulation (heparin) were centrifuged at 3,000 rpm for fifteen minutes on Fp-510 centrifuge (Labsystems OY Finland) to get clear plasma for biochemical analysis.

Hematological analysis

Blood samples from the control as well as exposed groups were analyzed for the hematological studies. Anticoagulant (heparin) preserved blood was used for the estimation of various hematological parameters like Hb, PCV, RBC, WBC, DLC, platelets, PCV, MCV and MCH by Haemocytometer method of Sharma and Singh 2000. Estimation of hemoglobin content was done according to Van-Kampan and Zijlstra (1961), packed cell volume according to micro-hematocrit method of Strumia *et al*., (1954), Red blood cell (RBC) values were utilized for calculating mean corpuscular hemoglobin (MCH), mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC) according to Dacie and Lewise (1991).

Statistical analysis

Student 't' test was applied for comparison of the data of control with the test samples. Values of P less than 0.05 were considered significance.

RESULTS

The comparative analysis of hematological parameters in Grass carp and Silver carp exposed to nickel in the form of nickel chloride for 7 days yielded valuable insights into their respective responses to environmental stressor. This study revealed that while both fish species exhibited some shared effects of exposure, there were also distinct species-specific responses. Notably, grass carp displayed more significant alterations in various hematological parameters, including WBC count, We found that Grass carp exposed to nickel for 7 days showed a significantly higher WBC count of (56.34±10.92 $x10^3$/l) than the control group (46.4±15.4 $x10^3$/l). In contrast, Silver carp exposed for 7 days had a markedly lower WBC count (29.03±19.29 $x10^3$/l) compared to their respective control group (55.16±12.75 $x10^3$/l). This suggests that Grass carp exhibited a more robust immune response to nickel chloride exposure than silver carp.

Hemoglobin (Hb) levels in both Grass carp and Silver carp did not show significant differences between the control and treated groups. While there was a slight decrease in Hb levels in grass carp.For Grass carp, the Hb concentration in the control group was 5.13±0.80(gm/dL), and after 7 days of exposure, it was (5.30±0.60(gm/dL), indicating a non-significant increase. For Silver carp, the Hb concentration in the control group was 5.13±2.12, and after exposure, it was (14.2±1.27(gm/dL)again showing a non-significant increase.

Grass carp exhibited no significant change in (RBC) count between the control (2.406±0.29/mcl) and the treated group (2.710±0.25/mcl). In contrast, Silver carp exposed for 7 days displayed a significantly lower RBC count (0.530±0.41/mcl) compared to their control group (1.156±0.16/mcl)..

Hematocrit (Hct) level significantly decreased in Grass carp after 7 days of exposure (36.8±2.43%) compared to the control group (40.76±4.78%). Similarly, Silver carp exhibited a substantial decrease in Hct (6.60±5.95%) compared to their control group (20.40±2.79%). Both species

displayed significant reductions in Hct, indicating that nickel exposure had a notable impact on this parameter in both fish.

Mean cell volume (MCV) in Grass carp showed significantly lower values (136.4±3.72fL) compared to the control group (169.5±2.05fL). In contrast, Silver carp displayed significantly lower MCV (136.7±0.90) after 7 days of exposure compared to their control group (177.1±1.22fL). Both species exhibited a decrease in MCV.

While Grass carp displayed significantly lower mean cell hemoglobin (MCH) values (52.53±0.31(pg)) after exposure as compared to the control group (62.26±1.10(pg)), (MCHC) values remained relatively consistent. In silver carp, exposure for 7 days resulted in significantly higher MCH (47.133±0.57(pg)) and MCHC (35.06±0.60(pg)) values compared to their control group. Red cell distribution width (RDW-CV) values in both Grass carp and Silver carp did not show significant differences between the control and treated groups.

Platelet count (PLT) in both Grass carp and Silver carp control (101.3±26.23 (x10/^{3}L) and treated (597.3±215.6(x10/^{3}L) remained consistent between the control and treated groups, with no significant differences observed. Mean platelet volume (MPV) of Grass carp exposed displayed significantly higher values (7.96±0.29 (fL)) compared to the control group (7.15±0.86(fL)), while silver carp exhibited significantly higher MPV values (8.533±0.55(fL)) after exposure compared to their control group (7.63±0.14(fL)). Both species showed an increase in MPV, indicating potential platelet response to nickel chloride exposure.

Platelet distribution Width (PDW) values in Grass carp after 7 days exposure resulted in a significantly higher PDW (51.6±1.38%) compared to the control group (11.50±0.11%). In contrast, Silver carp displayed no significant difference in PDW between the control and treated groups. Plateletcrit (PCT) values in both Grass carp and Silver carp did not show significant differences between the control and treated groups, indicating that nickel exposure did not significantly impacted plateletcrit in either species.

In the current study comparisons of the hematological parameters in Grass carp showed non-significant changes except PDW (51.6±1.38%) (Table 1),while in Silver carp all parameters

showed non-significant changes except MCV(136.7±0.90 fL) and PDW (51.6±1.38%) displayed a significant change.

Discussion

The method of monitoring environmental contamination often involves bioaccumulation of heavy metals, particularly in aquatic ecosystems where organisms directly contact with polluted water. Fish, in particular, serve as reliable indicator of metal contamination in water system (Carla, *et al* .,2004, Al-Kahtani, 2009).Fish accumulate heavy metals from various sources such as water, sediments, and food supply, like algae, which is consumed by both herbivorous and omnivorous fish (Bebianno, *et al.*,2004).The amount of metal absorbed depends on the type of metal, the species of fish, and the specific chemical properties of the water. Living organisms accumulate heavy metals when they take in and store them faster than they can be broken down or expelled. These metals enter water supplies through industrial materials, consumer goods and even acidic rain that erodes soils, releasing heavy metals into streams, lakes, rivers and ground water (Rauf , 2009). Heavy metal toxicity has been linked to a variety of sources, including pollution of drinking water, high ambient air concentrations near emission sources, and the food chain. Heavy metals in fish can be determined using hematological variables (Witeska, 2003). It is common practice to evaluate the health of fish by measuring their hematological characteristics (Oshode *et al*., 2008). The intra- and inter-specific differences in fish make hematological studies challenging to interpret. Numerous factors, most significantly blood sampling and laboratory techniques, contribute to these variations (Hesser, 1960). Fish hematological measurements indicate the stress response to external or endogenous modifications more rapidly than other widely studied parameters (Cataldi E.*et al*., 1998).

The current study was carried out for scrutinizing the effect of Nickle on hematological parameters of freshwater fish Silver Carp and Grass Carp after exposing the fish to heavy metal for 7 and 25 days. The concentration of Hb, RBCs, and WBCs against Nickel have been observed and found that comparing the WBC count between Grass carp and Silver carp, it's evident that Grass carp exposed to nickel for 7 days showed a significantly higher WBC count (56.34±10.926) than the control group (46.4±15.4). In contrast, Silver carp exposed for 7 days had a markedly lower WBC

count (29.03±19.29) compared to their respective control group (55.16±12.75). These values showed that grass carp exhibited a more robust immune response to nickel chloride exposure than silver carp. The statistical analysis of data showed variances among species, durations of exposure and treatments. The contact of metal for different time durations affected significant impacts on WBCs of both experimental fish species,

While on another side Hemoglobin in both grass carp and silver carp did not show significant differences between the control and treated groups. While there was a slight decrease in Hb level in grass carp after 7 days of exposure, but this change was not statistically significant. Analysis of fish variance for information on Hb concentration revealed differences across species and treatments, whereas statistically significant differences existed between exposure times**.**There was no significant change in RBC count of Grass carp between the control (2.406±0.299 mcl) and the 7-day treated group (2.710±0.25 mcl). In contrast, silver carp exposed for 7 days displayed a significantly lower RBC count (0.530±0.41 mcl) compared to their control group (1.156±0.16 mcl). This suggests that Silver carp may be more susceptible to the effects of nickel c on RBC count than grass carp. nickel chloride exposure led to changes in the volume of red blood cells in both Grass carp and Silver carp.

Results of Moosavi and Shamushaki (2015) in the hematological part, indicated that the mean WBC, Ht, and Hb of fish in the control experiment had mean white blood cell counts (%), mean total hemoglobin concentrations of 8656± 0.13 217.15 (cells/l), and mean hemoglobin concentrations of 7.41 ±0.04 (grams per deciliter). After being exposed to nickel for 7 days, the trial fish showed a concentration-dependent decrease in these parameters., it is possible that this mechanism is responsible for the reduction in WBC count in the treatment groups (Svoboda ,2001). Depending on the species and the toxicant, researchers have found that specific blood parameters either decrease or increased. except for Grass carp treated for 7 days, all metal doses tested in the current investigation showed no statistically significant effects. According to Annune *et al.* (1994), the red blood cell count of *C.gariepinus* significantly increased after being treated with Zn. They concluded that the metal's osmotic influence on the blood was responsible for the increased red blood cell count, Another study found that the red cell count of *O.niloticus* dropped, although not significantly (Annune *et al.*, 1994).

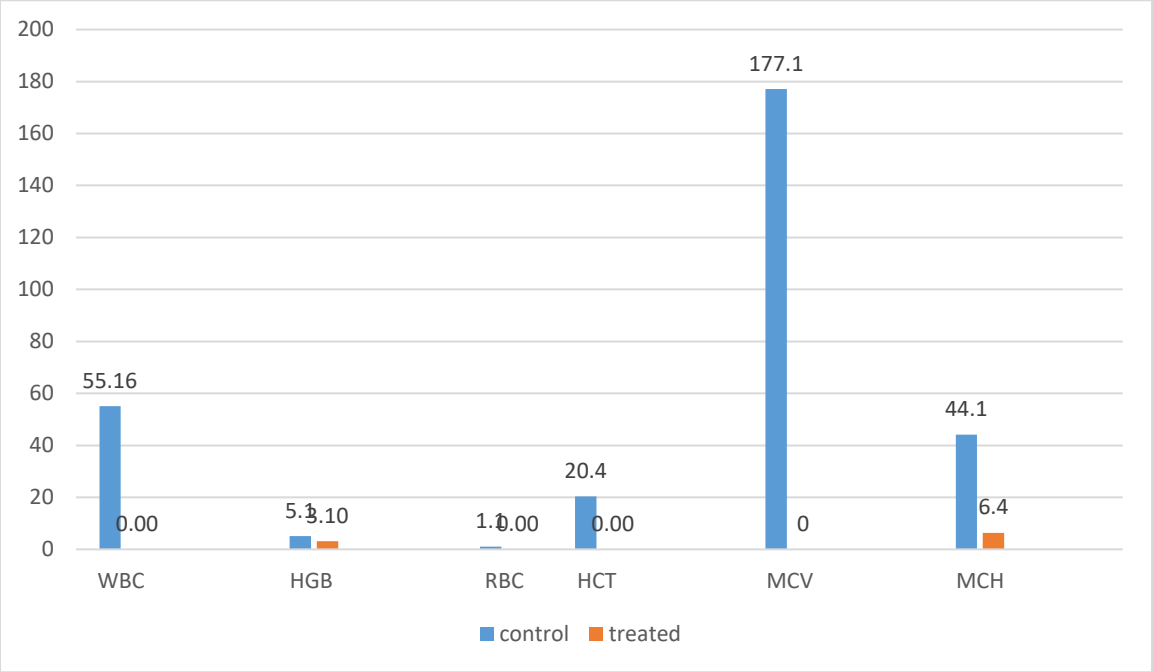
In the current study hematocrit of Grass carp showed significant changes in seven days while Flos *et al.* (1987) found that zinc treatment increased hematocrit levels in a variety of fish species. They concluded that the larger erythrocyte ware responsible for the increased hematocrit values in chromium and zinc-treated rainbow trout. According to Maheswaran *et al.,* 2008 that erythrocyte dissolution could account for decline in hemoglobin and hematocrit. A reduction in hematocrit and hemoglobin levels, as well as fewer and more abnormally shaped erythrocytes, are clear symptoms of anemia. Hemoglobin levels in *Ctenopharyn godon idell*a. Were found to rise significantly after 14 and 25 days of exposure in the current investigation. Abbas and Authman. (2009) reported some histological changes in gill structure of the Silver carp including hyperplasia, hypertrophy, and fusion of adjacent lamellae and shortening of secondary lamellae. *Cyprinus carpio* fingerlings exposed to nickel showed decreased blood parameters like erythrocyte, leucocytes, hematocrit and hemoglobin count and lowered values (MCV), (MCH) and (MCHC) when compared with the control fish. Moosavi and Shamushaki (2015) reported mean corpuscular volume and mean corpuscular hemoglobin content did not change significantly in the hematological indices reported, but mean corpuscular hemoglobin did, especially at higher concentrations (80% LC50). In contrast to the control, however, the MCV and MCHC showed some slight changes. These variations suggest that nickel chloride exposure had distinct effects on MCH and MCHC in both fish species Since the spleen is an erythropoietin organ, its discharged cells should have lower MCV values than the control. When cadmium was introduced to *Cyprinus carpio,* comparable results were seen (Koyama and Ozaki, 1984). Blood oxygen carrying capacity is decreased and erythropoiesis increases as a result of the significant difference in MCH between the trial fish and the control (Hodson, et *al* (1978).

Present study showed that the impact of nickel chloride on the distribution of red blood cell sizes in both fish species is not significant during study period. These differences suggest that grass carp experienced a significant changes in platelet distribution width, while silver carp did not respond in the same way to nickel chloride exposure respectively.

Conclusion

This comparative study underscores the distinct hematological responses of grass carp and silver carp to nickel exposure. Grass carp has shown significant changes in various blood parameters, it mean grass carp is more effected and vice-versa. While Silver carp affect exhibited comparatively

milder alterations. These findings emphasize the critical need to consider species-specific sensitivities in evaluating the impact of environmental stressors on fish populations. Understanding these unique responses is vital for devising effective conservation and management strategies for aquatic ecosystems threatened by heavy metal pollution.



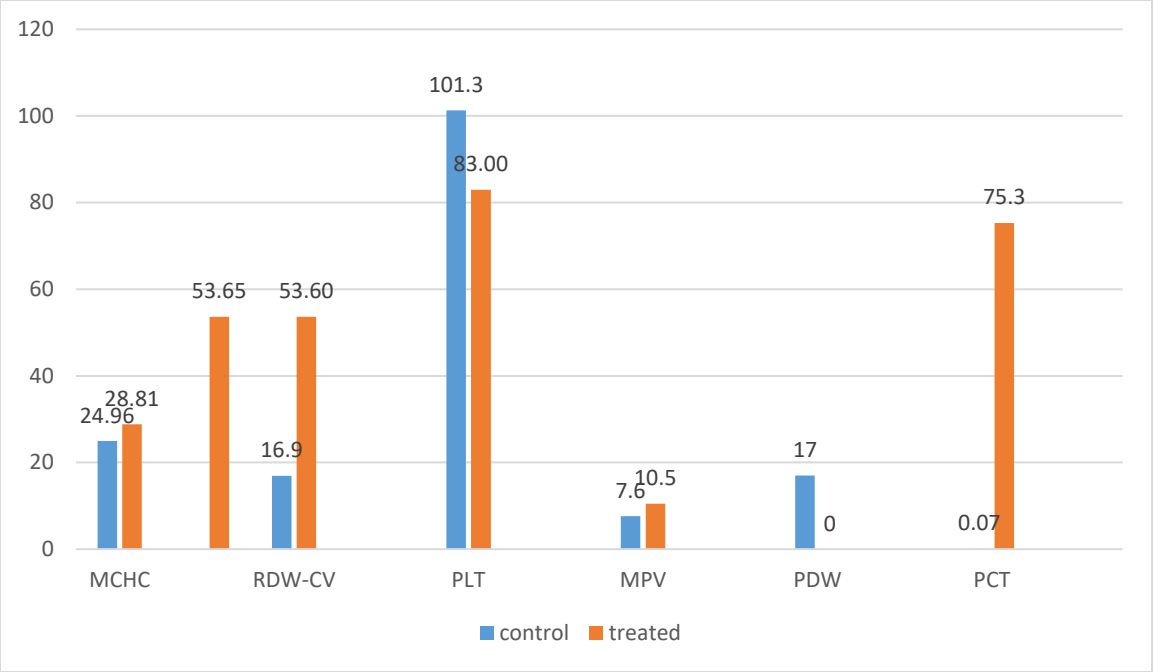
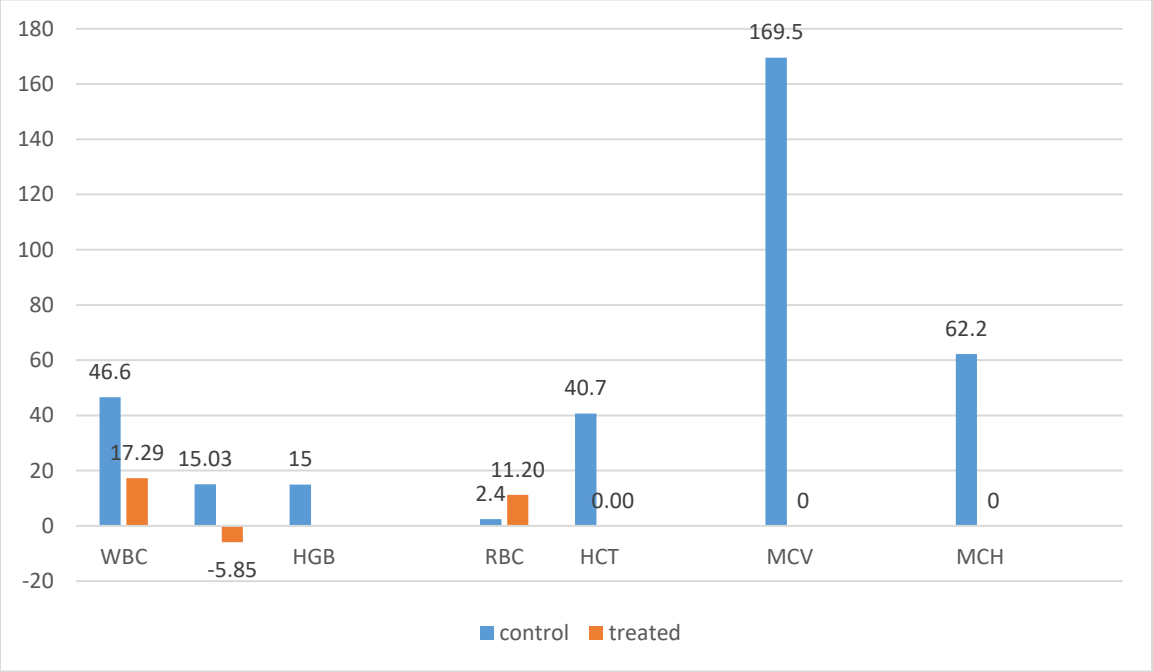


Fig I. showing % variance in hematological parameters of Silver carp after 7 days exposure to 120 mg/l of nickel).



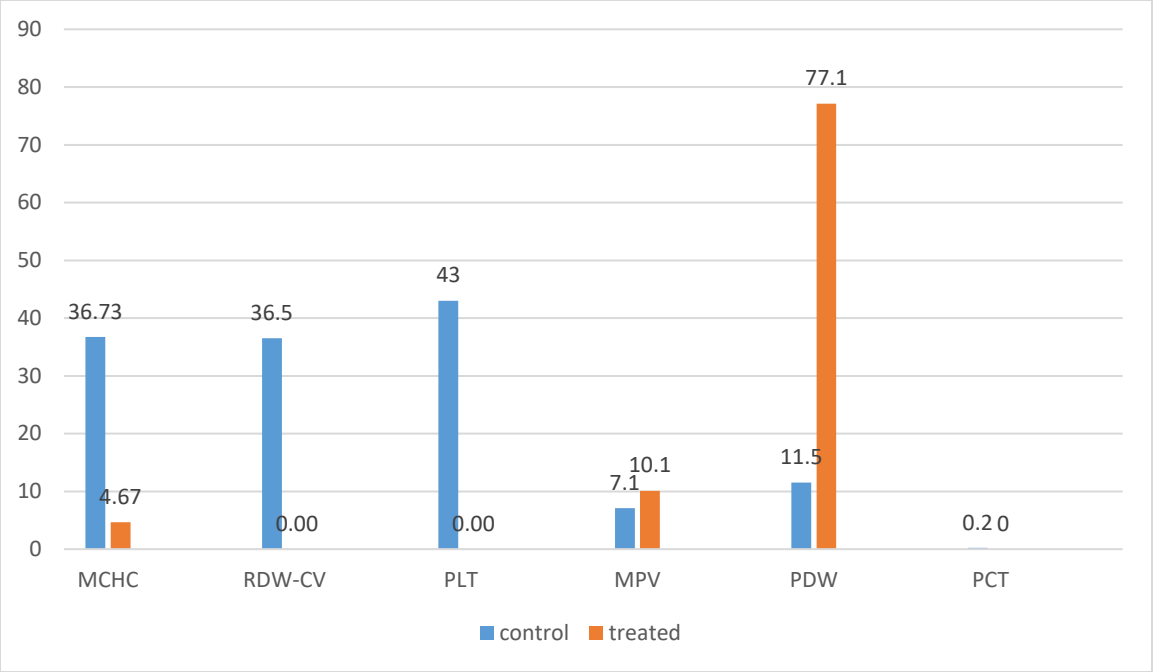


Fig II. Showing % variance for hematological parameters of grass carp after 7 days' exposure to 120 mg/l of nickel).

In the 2nd set of experiment fish were exposed to nickel for 25 days

After 25 days of 80mg/l nickel treated fish showed higher WBCs than control fishes. in Silver carp control group 55.166±12.754 ($x10^3$/l) as compared treated fish 61.5±26.471($x10^3$/l), while Grass carp control group 46.4±15.450($x10^3$/l) as compared to treated group 57.46±10.64($x10^3$/l) (Table, 2).

The Hemoglobin content for 25 days significantly affected during exposure of 80mg/l nickel of both the experimental fish species. The Hb content in both exposed fish was higher than in the control.in silver carp control group 5.133±0.808 (gm/dL) as compared to treated 5.70±1.847 (gm/dL) showed non- significant, while grass carp control 15.03± 2.1262(gm/dL) as compared to exposed group 13.26±0.569(gm/dL) showed significant variation.

The low and high level of RBCs resulted from the exposure of fishes to 80 mg/l concentration of nickel. Studying RBCs levels in fish revealed highly significant (p0.05) variations between fish species, exposure times, and treatments. In Silver carp control group was (1.156±0.163/mcl) as compared to treated group (1.12±0.611/mcl,) while grass carp control group was (2.406±0.299/mcl) as compared to exposed group (2.583±0.103/mcl), both showed non-significant variation.

The data about HCT of both experimental fish exhibited maximum HCT content in treated group as compared to control group. The statistical analysis of HCT content is shown in Tables 15, 16, and 17. It showed differences among species, exposure durations and treatment.in Silver carp

control group was 20.40±2.79 and exposed 19.7±6.35 showing non-significant changing, while grass carp control group as compared to treated group 40.76± 4.784(%) 32.9±0.901(%) showed significant.

The mean cell volume showed variations in contact to 80 mg/l nickel chloride exposure. Silver carp that had a lower MCV than grass carp. In Silver carp control group had 177.1±1.222 (fL) as compared to treated 143.6±1.558 (fL) MCV showing significant variation, while grass carp control and treated MCV values were169.5±2.052 (fL) 127.5±3.926 (fL) showed non-significant.

Both fish species showed significantly higher MCH level compared to its control.In silver carp control group 44.10±0.702 (pg) as compared to treated group had 41.20±0.230 (pg), while grass carp control group 62.26±1.102 (pg) as compared to exposed group had 153.9±0.416 (pg).

Under the exposure of metal increase in MCHC was noted down for all the experimental fish. Fish treated for 25 days with 80mg/l of nickel which data analysis of the MCHC revealed statistically significant (p0.05) variations between species, treatments, and exposure durations. In Silver carp control group value for MCHC was 24.96± 0.523(g/dl) as compared to exposed group 28.70±0.115 (g/dl) ,while in Grass carp control was 36.73± 1.0713(g/dl) while treated was 40.33± 1.481(g/dl).

Silver carp and grass carp showed high level of RDW-CV as compared to control. In Silver carp control group as was 16.96±0.218 (%) and for treated was 11.33±0.145 (%), while for Grass carp control was 36.56±2.197(%) and for treated 126.8±15.034(%) .

Platelets content in Silver carp control group as compared to treated group 101.3±26.238 ($x10/^{3}L$)and for treated was 90.3±26.26($x10/^{3}L$), while for Grass carp control group was 43.0±5.196**(**$x10/^{3}L$)and for treated was13.66±1.855($x10/^{3}L$) both showed non- significant.

Both experimental fish showed value of MPV values in Silver carp control group 7.633±0.145(fL) as compared to exposed group 6.85±0.028(fL) showed non-significant, while Grass carp control group of fishes 7.15±0.866(fL) as compared to treated group 8.667±0.185(fL) showed non-significant. (Table 2).

The PDW content during exposure of 80 mg/l of nickel chloride significantly affected both the experimental fish species. Both grass and Silver carp exposed fish showed high platelet distribution width value than control .in Silver carp control with treated 17.03±0.176(%) 16.56±0.333(%) while in Grass carp control group 11.50±0.115(%) as compared to exposed group 57.30±0.519(%) both showed non- significant.

Data about both two experimental fish species exhibited maximum PCT content of 80mg/l of nickel chloride treated fishes as compared to control fishes. Analysis during exposure showed significant differences during time duration and treatmet.in silver carp control group 0.077±0.021(%) as compared to treated 0.0613±0.0181(%), while grass carp control as compared to treated 0.250±0.002(%) 0.80±0.057(%) both showed non-significant. (Table 2).

Table 1. Showing hematological indices after 7 days exposure of Grass carp and Silver carp to a dose of 120mg/l of nickel

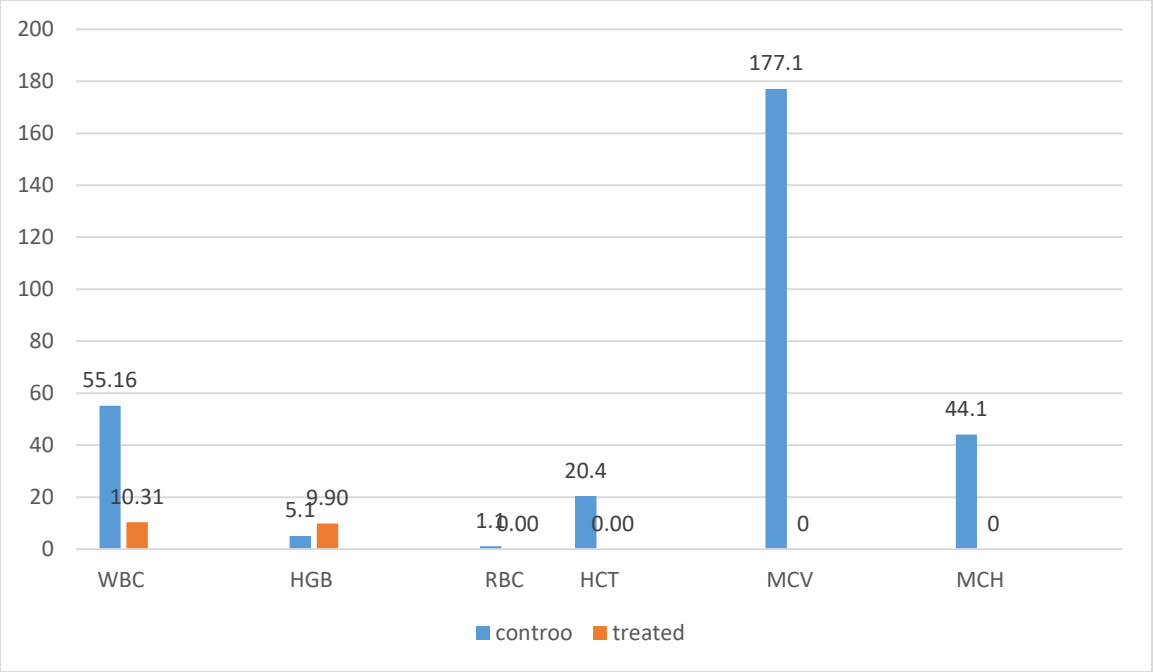
	Silver carp ***(Hypophthalmic molitrix)***		**Grass carp** ***(Ctenopharyngo donidell*****a)**	
Parameters	Control Mean ±SE	Treated Mean ±SE	Control Mean ±SE	Treated Mean ±SE
WBC ($x10^3$/l)	55.166±12.75	29.03±19.29	46.4±15.45	56.34±10.92
HGB (gm/dL)	5.133±0.80	5.30±0.60	15.03±2.12	14.2±1.27
RBC cells/mcl	1.156±0.16	0.530±0.41	2.406±0.29	2.710±0.25
HCT (%)	20.40±2.79	6.60±5.95	40.76±4.78	36.8±2.43
MCV(fL)	177.1±1.22	136.7±0.90***	169.5±2.05	136.4±3.72
MCH(pg)	44.10±0.70	47.133±0.57	62.26±1.10	52.53±0.31
MCHC(g/dl)	24.96±0.52	35.06±0.60	36.73±1.07	38.53±0.90
RDW-CV((%)	16.96±0.21	36.46±0.48	36.56±2.19	27.4±3.05
PLT($x10/^3$L)	101.3±26.23	597.3±215.6	43.0±5.19	41.0±1.52
MPV(fL)	7.633±0.14	8.533±0.55	7.15±0.86	7.96±0.29
PDW(%)	17.03±0.17	15.83±0.64*	11.50±0.11	51.6±1.383**
PCT **(%)**	0.077±0.02	0.312±0.05	0.250±-0.0	0.026±0.0

Values are presented as means ± SE (n = 3); NS = Non-significant (P>0.05); one *****for significant (P<0.05) two ** for more significant and three *** for highly significant.

Table 2. Showing hematological indices after 25 days exposure of Grass carp and Silver carp to a dose of 120mg/l of nickel.

Silver carp (*Hypophthalmic molitrix*)			**Grass carp (*Ctenopharyngodon idell*a)**	
Parameters	Control Mean ±SD	Treated Mean ±SD	Control Mean ±SD	Treated Mean ±SD
WBC ($x10^3$/l)	55.166±12.75	61.5±26.471	46.4±15.450	57.46±10.64
HGB (gm/dL)	5.133±0.808	5.70±1.847	15.03±2.126	13.26±0.569*****
RBCcells/mcl	1.1567±0.163	1.12±0.611	2.406±0.299	2.583±0.103
HCT (%)	20.40±2.797	19.7±6.350	40.76±4.784	32.9±0.901******
MCV(fL)	177.1±1.222	143.6±1.558***	169.5±2.052	127.5±3.926
MCH(pg)	44.10±0.702	41.20±0.230	62.26±1.1020	153.9±0.416
MCHC(g/dl)	24.96±0.523	28.70±0.115*****	36.73±1.071	40.33±1.481
RDW-CV(%)	16.96±0.218	11.33±0.145	36.56±2.197	126.8±15.034******
PLT($x10/^3$L)	101.3±26.23	90.3±26.26	43.0±5.196	13.66±1.855
MPV(fL)	7.633±0.145	6.85±0.028	7.15±0.866	8.667±0.185
PDW(%)	17.03±-0.176	16.56±0.333	11.50±0.115	57.30±0.519
PCT**(%)**	0.077±0.021	0.061±0.018	0.250±0.002	0.80±0.057

Values are presented as means ± SE (n = 3); NS=Non-significant (P>0.05);
one *****for significant (P<0.05) two ** for more significant and three *** for highly significant.



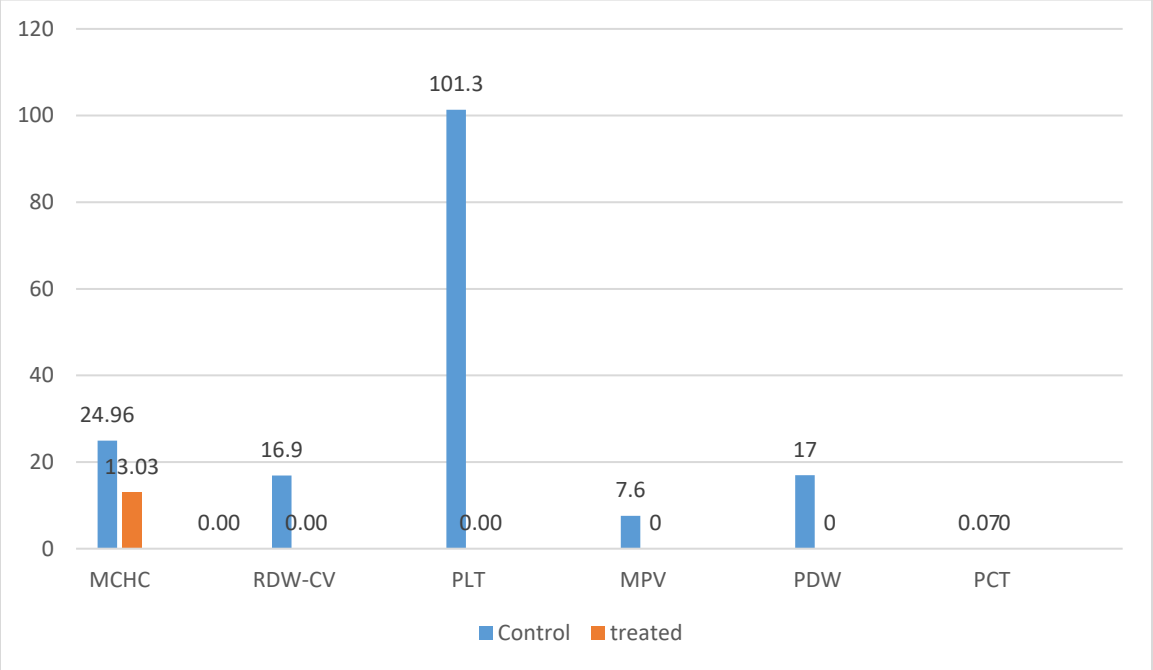
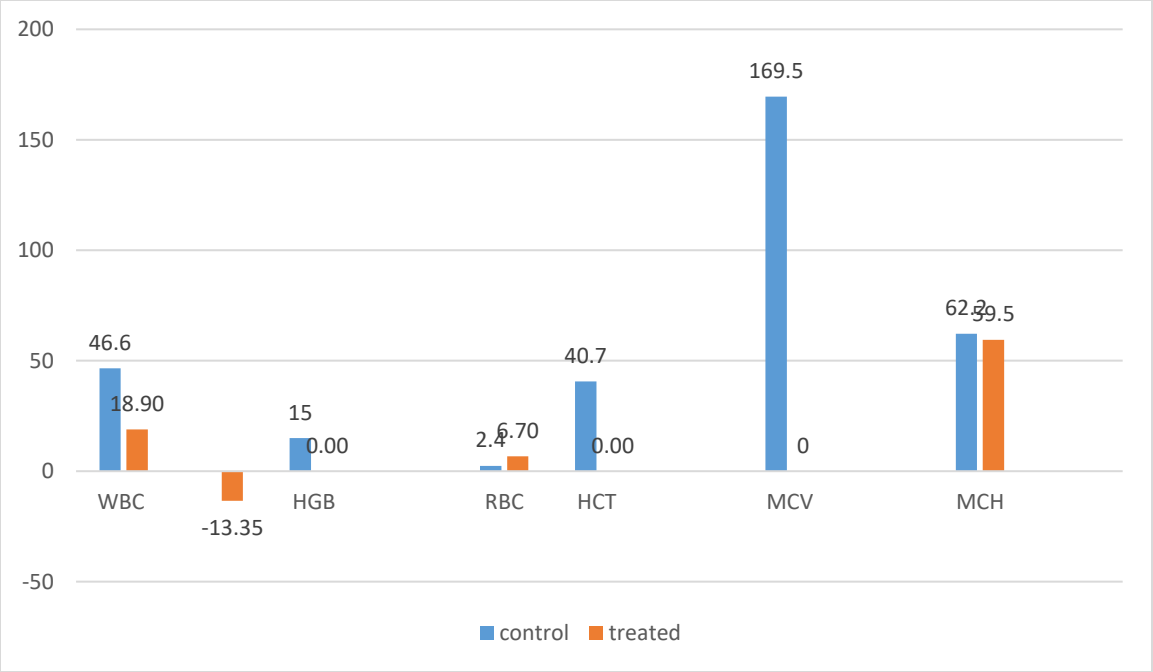


Fig III. Showing % variance for hematological parameters of Silver carp after 25 days' exposure to 80 mg/l of nickel).



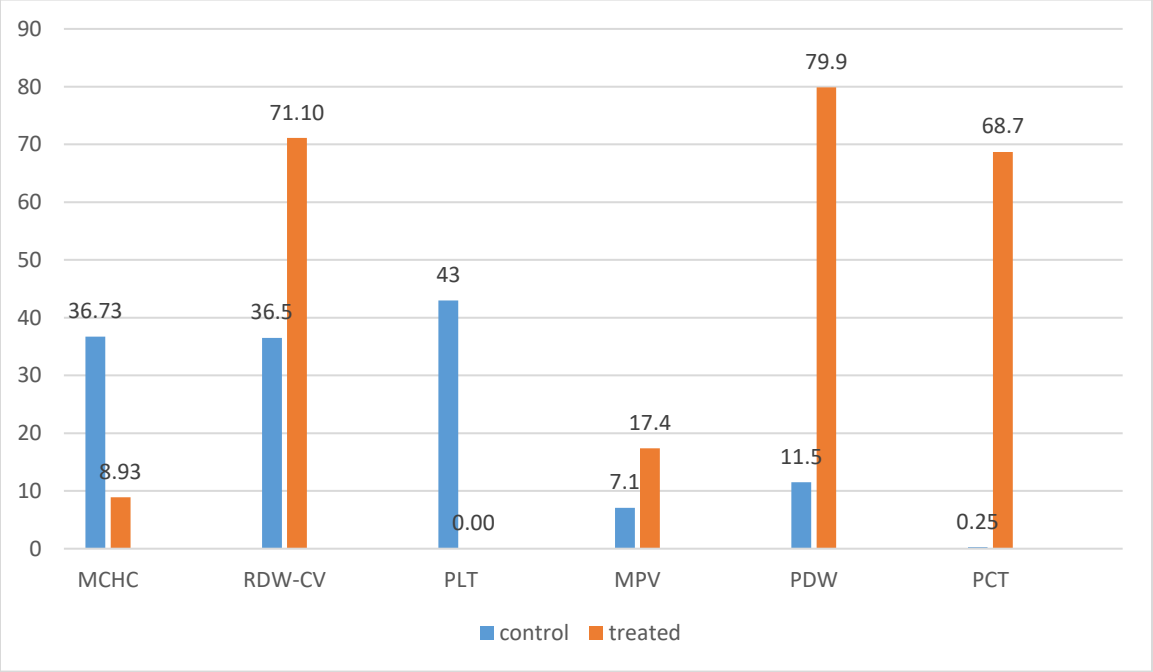


Fig IV. Showing % variance for hematological parameters in Grass carp after 25 days' exposure to 80 mg/l of nickel).

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