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Effect of Selenium Incorporated in Feed on the Hematological Profile of Tilapia (*Oreochromis niloticus*)

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Abstract

The present study was aimed at the assessment of potential effect of selenium, supplemented in feed, on the hematological profile of tilapia (*Oreochromis niloticus*) while maintaining certain physicochemical parameters of water. Three doses differing merely in selenium contents viz. 2, 4, and 8 mg Se/kg of fish feed were formulated bearing in mind apiece dose as an independent treatment. Four cemented rectangular tanks (triplicated) were used after proper disinfection to ensure sustainable culture environment. 15 fish per tank were stocked after appropriate health examination weights ranging 10-25 g. Variations in different hematological parameters by counting of white blood cells, red blood cells, hemoglobin estimation, granulocytes (neutrophils, eosinophils, and basophils), agranulocytes (lymphocytes and monocytes) as well as weight and length gains were recorded and analyzed using one-way ANOVA. The results revealed that WBC's counts were non-significantly (P=0.05) different among treatments 1, 2, 3 as well as in the selenium-deficient treatment. However, the counts of WBC's, neutrophils, RBC's and hemoglobin level was enhanced (P=0.05) in treatment-1 (2 mg Se/kg). On the contrary, hemoglobin level, neutrophil and RBC's counts were significantly dropped (P=0.05) low in treatment-3 (8 mg Se/kg). The WBC's counts were found lower in treatment-2 (4 mg Se/kg). Lymphocytes and monocytes were significantly higher in treatment-3 (8 mg Se/kg). This study has enlightened that the supplementation of selenium (2 mg/kg) in the feed of tilapia does not alter its inclusive hematological profile but promotes better physiological performance and productivity to enhance fish growth and paves the way towards increased supply of selenium-fortified fish meat.

Keywords: Selenium; Hematology; WBC's; RBC's; Physicochemical parameters; Tilapia; Weight gain

Introduction

Tilapia, the aquatic chicken, has recently emerged as an important aquaculture candidate species with faster growth rate besides increasing global market demands [1]. Being omnivorous in nature, efficient in food conversion and ability to cope with changes in water quality, it has emerged as an appropriate organism for the investigation of potential effects of various micronutrients like selenium when supplemented in feed. Selenium (a non-metallic element) is an essential micronutrient in standard animal nutrition plan, even though is required in trace amounts [2-4], acts as an antioxidant, compensative in metabolism, immune system, growth increments [5], plays an important role as immunostimulant as well as in normal body functions of fish. For humans, selenium is essential trace mineral with fundamental importance being a constituent of selenoproteins and catalyst for thyroid hormonal production [6]. In aquaculture, it acts as a micronutrient of live feed that stabilizes the nutritive balance in cultured fish [7-9]. Four oxidative forms of Se (-2, +2, +4, +6), selenite and selenate are dominant in aquatic environment due to higher solubility. Organic forms of Se are comestible, well bio-accumulated, biologically more active and less toxic than inorganic forms [10]. Selenium and its compounds are necessary for the development of immune system and standard function of antioxidant enzymes in the body [11-13]. Very importantly, a relatively narrow line is present among nutritive requirements of Se and its toxicity [14].

Hematological investigations are vital as an index of monitoring health conditions of fish [15] as various physiological functions are affected when trace elements are absorbed by the blood [16] or vice versa. Various factors such as species, age, health, environmental conditions, nutrition and maturation are reported to affect the blood and physiological parameters in fish [17]. Therefore, hematological parameters are indicators of fish environment and feed intakes [18] as fish can accumulate a range of elements [19] like selenium that can be even toxic if absorbed in greater concentrations. Hematological indices are increased by a reduction in pH which is resulted through osmotic imbalance as well as changes in the ion regulatory system of tissues such as sodium and potassium pump [10]. As fish health and hematological features are interlinked, therefore estimation of various parameters of blood can reveal fish health status upon deviations from normal physiological functions. Such physiological changes confirm a direct association of the external environment to the blood circulatory system. For instance, in fish, the concentration of blood glucose increases with the temperature of water and decreases with length and age of fish [20]. At a lower level of DO, no hematological changes are noticed in fish [21]. Main alterations in the blood composition are influenced by exogenous factors like disease, stress due to change of living environs and/or the concomitant variations in blood chemistry [22,23]. Levels of RBC's, hemoglobin and hematocrit were reportedly decreased in *L. rohita* when exposed to higher concentrations of sodium selenite. Transport

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capacity of oxygen is assessed by various blood parameters that are hemoglobin, white blood cells, hematocrit and RBC's [13]. On the other hand, in hybrid tilapia, hematocrit level dropped significantly low when selenium supplementation level decreased up to zero levels [24]. Amounts of RBCs, hematocrit, lipids, hemoglobin, glucose and protein increased when the concentration of organic selenium increased in fish feed up to 0.3g/kg clearly indicating the improvement in fish health [25] because of selenium dose. Hematological variables are affected by different protein levels, the weight of fish and related indices [26] as well as due to deficiency of micronutrients like selenium. Fish exposed to probiotics showed lower blood glucose levels because the probiotics enhanced ingestion and digestion capability [27]. Total hemoglobin concentration and blood hematocrit levels were not ostentatious during the cold shock [28] but in stressful conditions like hypoxia, the stored RBC's come into circulation due to Splenic contractions [29,30]. Nile tilapia is found tolerant to lower concentrations of DO as low as 1 mg L^{-1} [21] as well as other water quality factors like temperature, salinity, electrical conductivity, carbonates etc. Uptake of oxygen is disturbed when male and female fishes are exposed to the toxic doses of Se, and its various compounds. Below the toxicity stress conditions, oxygen uptake is increased because of the rise in demand [31].

Therefore, this study was anticipated to explore the potential effect of selenium, supplemented in feed, on hematology of tilapia (*Oreochromis niloticus*) while retaining selected physicochemical parameters of water adjacent to optimal values so that individual effect of selenium is properly manifested. It also included assessing the weight and length gains by juvenile tilapia on selenium graded and selenium-deficient treatments.

Materials and Methods

Experimental site

This 90 days dietary investigation about the effect of selenium supplemented in feed on hematological parameters of Tilapia (*Oreochromis niloticus*) was conducted in concrete, fixed rectangular tanks inside the Fish Hatchery Complex at Research and Training Facilities, Department of Fisheries and Aquaculture, C-Block, Ravi Campus, University of Veterinary and Animal Sciences, Lahore, Pakistan.

Fish management and experimental plan

The uniformly sized and healthy tilapia were procured from the ponds of C block, Ravi campus and were transferred to the fish hatchery complex. First of all, the fish were properly acclimatized for two weeks duration and were fed at controlled diet followed by purging. Four types of semi-purified iso-nitrogenous feeds consisting of three treatment diets and one control feed were formulated and prepared for the whole experimental duration. The details of selected feed ingredients, thr formulated controlled and basel diets along with their chemical composition are given in Table 1. The treatment diets, considering each as independent treatment, were prepared on the basis of inclusion level of selenium i.e., 2 mg/kg (treatment-1), 4 mg/kg (treatment-2) and 8 mg/kg (treatment-3) while control diet was prepared without selenium supplementation. All the diets were formulated with similar protein and energy levels. All the formulated ingredients were mixed properly with the help of mixer followed by addition of pre-determined doses of selenium for each treatment and no selenium in the control diet. Then the feeds were separately extruded into pellets followed by packing and storage at -20°C until rationing to fish. The dimensions of concrete rectangular tanks were 2.896 × 0.762 × 0.914 m (length × width × depth) or 2.018 cubic meters. Tank 1, 2 and 3 were designated as treatment tanks whereas the 4th one as a control. There were three equal replicates (triplicates) in each of the treatment tanks as well as in control tank. 15 fish per tank were stocked having weight range 10 g to 25 g and were fed on more or less 30% CP feed @ of 3% body weight thrice each day till satiation level. The condition of the research tanks was sustained as clean, disinfected with an uninterrupted flow of

S. No.	Ingredients	Inclusion level (g/100 g)			
		Basel diet (Control)	Treatment 1	Treatment 2	Treatment 3
1	Fish meal	8	8	8	8
2	Guar meal	30	29.998	29.996	29.992
3	Soya bean meal	9	9	9	9
4	Wheat bran	18	18	18	18
5	Canola meal	8	8	8	8
6	Rice polish	24	24	24	24
7	Vitamin Premix [a]	2	2	2	2
8	Selenium free mineral premix [b]	1	1	1	1
9	Selenium dose [c]	0.00	0.002	0.004	0.008
	Total	100 g			
Chemical composition					
1	Crude protein	30.2	30.2	30.2	30.1
2	Crude lipid	7.3	7.2	7.3	7.3
3	Dry matter	86.4	86.4	86.3	86.5
4	Ash	6.8	6.7	6.6	6.9

a: Vitamin premix (IU or g/kg diet): Vitamin A, 16000 IU; Vitamin D, 8000 IU; Vitamin K, 14.72; Thiamin, 17.8; Riboflavin, 48; Pyridoxine, 29.52; Cynocobalamine, 0.24, Tocopherols acetate, 160; Ascorbic acid (35%), 800; Niacinamide, 79.2; Calcium-D- Pantothenate, 73.6; Folic acid, 6.4; Biotin, 0.64; Inositol, 320; Choline chloride, 1500; L-Carnitine, 100;
b: Selenium free mineral premix, **(g /kg of diet):** Calcium, 5.5; Phosphorus, 17.5; Iron, 10; Magnesium, 2.8; Copper,1.5; Iodine, 0.15; Manganese, 9.5; Zinc, 25; Cobalt, 0.13;
c: Sodium selenite (Na_2SeO_3) in milligrams.

Table 1: Selected feed ingredients (dry weight), inclusion level and chemical composition of experimental and basal diets.

Parameter	Treatments				
	Control	Treatment 1	Treatment 2	Treatment 3	Permissible limits
pH	8.58 ± 0.020	8.56 ± 0.028	8.58 ± 0.017	8.57 ± 0.018	7-9
D.O.	6.20 ± 0.150	6.04 ± 0.167	6.14 ± 0.289	6.26 ± 0.274	>5
Temperature (°C)	30.35 ± 0.022	30.35 ± 0.026	30.33 ± 0.030	30.34 ± 0.022	15-35
TDS	396.92 ± 26.88	378.06 ± 23.378	441.81 ± 37.648	430.19 ± 32.532	500-1200
EC (µS/cm)	649.09 ± 14.776	659.27 ± 34.58	663.29 ± 30.429	697.79 ± 23.835	300-1500
Hardness	18.1 ± 0.012	18.2 ± 0.014	18.03 ± 0.018	17.9 ± 0.015	>20
Nitrates	0.83 ± 0.13	0.84 ± 0.15	0.83 ± 0.14	0.84 ± 0.20	0-100
Chlorides	6.5 ± 0.11	6.9 ± 0.19	7.0 ± 0.13	7.0 ± 0.18	4-160
Salinity	0.8 ± 0.001	0.8 ± 0.001	0.8 ± 0.001	0.8 ± 0.002	--
Ammonia	N.D.	0.011 ± 0.0034	0.012 ± 0.0051	0.010 ± 0.0032	0-0.05

D.O.: Dissolved Oxygen, TDS: Total Dissolved Solids, EC: Electrical Conductivity, T.A. Total Alkalinity, N.D.: Not Detected.
All values are mentioned in mg/L (ppm) except pH, temperature and electrical conductivity.
Statistical significance level (P<0.05).

Table 2: Monitoring records of selected physicochemical parameters in selenium graded and selenium-deficient tanks.

well-oxygenated freshwater maintained automatically by overflow pipe throughout the research trial. Further fish was daily monitored for its feed intake, erratic swimming, apparent injury or infection in order to ensure the healthy fish during culture duration. The weight and length gains were recorded from each tank on fortnightly basis for growth estimation as well as next fortnight ration adjustments.

Physicochemical parameters

Physicochemical parameters viz. pH, dissolved oxygen (DO), temperature, total dissolved solids (TDS), electrical conductivity (EC), salinity, ammonia levels, hardness, chlorides and nitrates were recorded on daily basis in order to check any impending changes so that no harm of physicochemical parameters was inflicted upon the fish health. Electrical conductivity (E.C.), total dissolved solids (TDS) and salinity were measured by conductivity meter *(Condi 330i WTW 82362 Weilheim, Germany)*. Temperature and dissolved oxygen were measured by using D.O. meter *(YSI 55 Incorporated, Yellow Springs, Ohio, 4387, USA)*. The pH was noted by pH meter *(LT-Lutron pH-207, Taiwan)*. Ammonia by colorimetric method, hardness by titration with EDTA, chlorides by titration (*Mohr Method: Silver Nitrate*), and nitrates were measured by spectrophotometry.

Hematological parameters

Fish blood was collected by humanly sacrificing the fish using sterilized non-reusable plastic syringe having a 22-gauge needle [32]. Heparin sodium (1%) was added as an anticoagulant [23]. Diluted the blood through diluted solutions that are commonly used for numerical counting of RBC's and WBC's and were determined through using Neubauer hemocytometer [33]. Sahli's haemoglobinometer was used to calculate hemoglobin (Hb) percentage. For differential leucocytes count (DLC), slides were stained by using *Giemsa* stain [34]. At least three slides were prepared and examined for each treatment fortnightly.

Statistical analysis

The obtained results were analyzed in Statistical Analysis System (SAS 9.1) by using *one-way analysis of variance (ANOVA)*. While comparing the means, *Tukey's LSD (least significant difference)* test was used to detect a significant difference and the treatment differences were calculated with the significance level at ($P< 0.05$).

Results

There was absolutely zero mortality as well as no diseased fish individuals were observed during the whole trial as per DELT (Disease, Erosion (fin or skin), Lesion, T (tumor). The macroscopic morphological observation of fish revealed to no infection whatsoever. The regular observation of the fish activity especially right after dispensing the feed revealed that fish remain active throughout the 90 days consuming almost all feed within 30 mintes after ration was dispensed.

Physicochemical parameters

Selected water quality parameters were upheld adjacent to optimal ranges to ensure the minimal effect of environmental disturbance which could lead to stress build up and resultant compromised physiological function in fish. The means of selected physicochemical quality parameters of culture environment recorded from treatment and control tanks are given in Table 2. Treatment-1, 2, 3 and control showed the non-significant difference in pH, DO and temperature whereas the highly significant difference in TDS (430.19 ± 32.532 mg/l) was observed in treatment-3 tanks (Se 8 mg/kg) Treatment-2 and control showed non-significant TDS. A non-significant difference in EC was noted in treatment-1, 2 and control tanks whereas highly significant EC (697.79 ± 23.835 mg/l) was noted in treatment-3 tanks. Highest growth of tilapia (*Oreochromis niloticus*) was found at temperature (30.34 ± 0.022), pH (8.57 ± 0.018), DO (6.26 ± 0.274), EC (697.79 ± 23.835) and TDS (430.19 ± 32.532) in treatment 1. Ammonia was none detectable (ND) in control tank whereas it was recorded below the permissible limits in all the treatments. Hardness, chloride level, as well as salinity values, were found non-significant in all the tanks confirming the quality of culture environment was well-maintained. The best growth treatment tank was also the one which was observed near to the optimal environmental quality ranges confirming that the environmental quality changes were negligible or none-players in the obtained hematological results because the overall culture environment was sustained to support the optimal physiological functions of the fish.

Hematological parameters

The monthly records on the effect of selenium, incorporated in the fish feed, on the hematological counts in selenium graded as well as selenium-deficient treatment tanks are given in Table 3. The mean hematological profiles of each treatment tank are mentioned in Table 4. The WBC's counts were non-significant in treatment-1, 2, 3 as well as in the controlled feed tanks. The highest WBCs count was noted in treatment 1 whereas the lowest from treatment 2 supporting enhanced level of WBCs supporting the hematological profile of

Parameters	Treatment-I			Treatment-II			Treatment-III			Control		
	1st month	2nd month	3rd month	1st month	2nd month	3rd month	1st month	2nd month	3rd month	1st month	2nd month	3rd month
WBC	1.71 ± 0.92	1.29 ± 0.87	1.86 ± 0.26	1.40 ± 0.59	1.14 ± 0.65	1.50 ± 0.44	3.95 ± 0.91	2.96 ± 0.38	3.16 ± 0.70	2.5 ± 0.32	2.6 ± 0.32	4.19 ± 3.81
RBC	6.28 ± 1.26	5.57 ± 1.55	6.20 ± 1.33	4.50 ± 0.60	4.18 ± 0.78	4.53 ± 0.55	2.44 ± 0.43	2.46 ± 0.41	2.54 ± 0.71	4.31 ± 0.71	3.58 ± 0.53	3.38 ± 0.43
Hb. %	7.62 ± 0.61	6.95 ± 0.35	6.98 ± 0.39	4.95 ± 0.38	5.87 ± 0.70	5.52 ± 0.38	3.08 ± 0.65	4.22 ± 0.96	2.87 ± 0.42	7.25 ± 0.72	6.83 ± 1.12	5.52 ± 0.45
Neut.	75.33 ± 4.59	68.50 ± 2.88	67.50 ± 2.74	66.83 ± 6.08	66.83 ± 2.40	66.83 ± 6.27	77.67 ± 2.50	73.50 ± 5.65	73.67 ± 3.01	54.33 ± 11.55	48.00 ± 20.25	43.00 ± 24.46
Eosin.	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	22.00 ± 16.17	4.858 ± 6.56	11.83 ± 2.48
Baso.	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	16.83 ± 4.12	16.17 ± 8.77	19.00 ± 5.25
Mono.	8.67 ± 1.63	8.67 ± 1.21	9.17 ± 1.47	4.17 ± 1.47	3.33 ± 1.51	4.33 ± 2.07	10.67 ± 2.42	8.67 ± 1.21	32.50 ± 5.39	9.17 ± 2.40	9.17 ± 3.97	8.67 ± 3.72
Lympho.	18.33 ± 1.86	16.00 ± 3.52	17.83 ± 2.64	23.83 ± 3.49	27.00 ± 2.90	28.50 ± 4.04	31.00 ± 3.03	39.83 ± 4.71	37.33 ± 8.04	21.00 ± 8.49	14.67 ± 6.50	35.50 ± 22.89

WBC = White Blood Cells (Thousands/mm^3); RBC = Red Blood Cells (million/mm^3); Hb = Hemoglobin in (g/dl); Neut = Neutrophils (%); Eosin = Eosinophils (%); Baso = Basophils (%); Mono = Monocytes (%); Lympho= Monocytes (%)

Table 3: Monthly means and SEM of hematological parameters of juvenile tilapia fed on selenium graded and selenium-deficient diets.

Treatments	Blood Parameters							
	WBC (Thousands/mm^3)	RBC (million/mm^3)	Hb (g/dl)	Neutrophils (%)	Eosinophils (%)	Basophiles (%)	Lymphocytes (%)	Monocytes (%)
Treatment 1	6.13 ± 1.422^a	6.90 ± 0^a	7.18 ± 0.376^a	70.44 ± 4.263^a	0^b	0^b	15.67 ± 3.055^b	8.83 ± 0.289^{ab}
Treatment 2	1.35 ± 0.183^a	4.40 ± 0.194^{ab}	5.44 ± 0.463^b	66.83 ± 0^a	0^b	0^b	26.44 ± 2.384^{ab}	3.94 ± 0.536^b
Treatment 3	4.87 ± 4.215^a	2.48 ± 0.055^b	2.48 ± 0.055^c	48.44 ± 5.678^b	0^b	0^b	36.05 ± 4.551^a	17.28 ± 13.221^a
Control	2.80 ± 2.458^a	4.47 ± 3.870^{ab}	6.53 ± 0.902^a	48.44 ± 5.678^b	16.67 ± 5.102^a	17.33 ± 1.481^a	23.72 ± 10.680^b	9.00 ± 0.289^{ab}

WBC: White Blood Cells, RBC: Red Blood Cells, Hb: Hemoglobin
Superscripted values represent the mean hematological profiles of each treatment tank that are statistically non-significant.

Table 4: Variations in various blood parameters in Tilapia (Oreochromis niloticus) in treatment and control tanks.

tilapia. Along with WBCs, the amounts of neutrophils, RBC's and hemoglobin concentration was greater in treatment-1 i.e., 2 mg Se/kg. Perversely, the hemoglobin level, neutrophils, and RBC's count was lower in treatment-3 (8 mg Se/kg) and so were the WBCs counts from treatment-2 (4 mg Se/kg). The RBCs were recorded the highest from T-1 and the lowest in T-3. Similarly, hemoglobin level was improved in T-1 and was found to be the lowest in treatment-3. Neutrophil counts were surprisingly recorded as the similar in control and treatment-3 whereas the highest of them were noted from treatment-1. Eosinophils and basophils percentages were noted near zero in all the treatments except control. Lymphocytes and monocytes were significantly higher in treatment-3 (8 mg Se/kg). The lymphocytes percentage was the lowest from treatment-1 whereas that of monocytes was recorded as the lowest from treatment-2. However, non-significant lymphocytes and monocytes were observed in treatment-1 and control tanks. Eosinophils are statistically highly significant in control tanks. Non-significant eosinophils were observed in treatment-1, 2 and 3 tanks. Highly significant basophils were present in control tanks. Basophils were non-significant in treatment-1, 2 and 3 tanks.

Weight and length gain

The weight and length gains from selenium graded and selenium-deficient treatment tanks have shown that promising and sustained growth records were observed in treatment 1 i.e., 2 mg Se/Kg of given feed whereas lower growth records were observed in the rest of treatments. The selenium-deficient fish showed arrested growth during the trial. The linear and forecasted lines from each treatment shows that the growth and weight length gains are selenium dose-dependent. Higher growth was observed in lower selenium dose and lower growth was noticed from higher selenium doses in the present study.

Discussion

The normal physiological regime of the fish body may be subjected to change due to alterations in the physicochemical setup of the culture environment. Fish are reported to come across different environmental diseases as well as physiological *(hematological)* changes as a result of physicochemical transformations of certain parameters of water, therefore physicochemical parameters were studied and well-maintained in order to rule out their concomitant effect on the blood compositions. It has been documented that hematological variables are indicators of stress [35,36] caused by environmental changes. As per our findings and effective management of water quality, treatment-1, 2, 3 and control showed non-significant changes in the pH, DO and temperature during the culture period. Similarly, non-significant variations were noted among the treatments in the cases of salinity, hardness, nitrates as well as ammonia. Findings of the current study were in agreement with Noori et al. [37] who testified that differences in physiochemical parameters were not significantly different among all treatments of the study and were within recommended ranges for fish. Results were also in line with Gaber [38] who found no significant differences in physicochemical parameters in all treatment and control tanks. Results of the present study were also in agreement with Abdel-Tawwab et al. [39] who revealed that non-significant difference was noted in water quality parameters in all treatments and control tanks. The physicochemical results of the present study are also in line with Iqbal et al. [40] who revealed that variations in water quality parameters were non-significantly different in all treatments so is the present study. By this state of affairs, we may assume that the potential hematological change in the present study was not affected by the physicochemical quality of the culture environment.

The nutritive status of fish can be linked to the health condition of an animal and potential way they deal with stress [41,42] resulting from their surrounding environment. In this study, there is a reasonable link between increased weight and length gains with the increased number of RBCs and other parameters. The primary function of WBC's is to defend the body against foreign pathogenic organisms. WBC's

were non-significantly varying among treatment-1, 2, 3 and control. It clearly denotes that the fish health was at the optimal level as well as no or minimal pathogenic activity in the fish body. In the current study, the amount of WBC's, neutrophils, RBC's and hemoglobin concentration was greater in treatment-1 (2 mg Se/kg) which confirms that this increase was the underplaying factor for the enhanced growth in treatment 1. This evidently signifies that the fishes thriving in treatment-1 were luxurious with defensive mechanism rendering them as empowered vanquishers against the impending possible pathogenic invasions or any ailments. Hemoglobin level, neutrophils, and RBC's count were lower in treatment-3 (8 mg Se/kg) and WBCs were lower in treatment-2 (4 mg Se/kg) which also showed lower growth records. Lymphocytes and monocytes were significantly higher in treatment-3 (8 mg Se/kg) which may be linked with impending ailment due to the toxicity of selenium herein higher quantity. A significant reduction in WBC's count in treatment-2 suggested that fish may have been exposed to higher risk of infection. The composition of blood is usually changed during malnutrition and/or stress conditions [43]. Notably, RBC's counts are mostly affected by dietary treatments [37]. Noteworthy declines in WBC's count may be due to stress enforced during handling activities for sampling as well as the selenium toxicity as it becomes toxic in slightly higher amounts. Results of the current study are in line with Noori et al. [37] who elucidated that RBC's were significant in all treatments and WBC's were statistically non-significant. In hybrid tilapia, hematocrit level was significantly lower when selenium supplementation level decreased up to zero [24], which is also confirmed in the present study as the selenium-deficient fish showed arrested growth increments. Similar results were stated by Bell et al. [44] with Atlantic salmon (*Salmo salar*); and Bell et al. [45] with rainbow trout (*Salmo gairdneri*). They reported that in selenium deficient fishes, packed cell volume was significantly lower than selenium supplemented fishes. Our findings are in agreement with the results of Bell et al. [46] who reported that level of hematocrit was not affected through giving selenium deficient feed but reduced when selenium was absent. No significant variations were observed in the level of hemoglobin and level of red blood cells in trout fed on different levels of selenium supplemented in diets (0.6, 6.6 and 11.4ug Se/g). Dimanov et al. [47] revealed that selenium supplemented feed (0.03 to 0.5 mg/g) improved hematocrit level in fingerlings of tilapia from 18-41%.

Ramesh et al. [13] studied *Labeo rohita* and revealed that moderate lethal concentration of sodium selenite (Na_2SeO_3) in water was 23.89 mg/L for 96 h. Various hematological parameters including hematocrit (Hct), erythrocyte (RBC) and hemoglobin (Hb) level were decreased in number when fish was exposed to sublethal (2.38 mg/l) concentration of selenium while fishes exposed to similar selenium concentrations showed increase in leucocytes (WBC) count as well as enhanced glucose level. Satheeshkumar et al. [48] reported that correlation of different blood parameters such as RBC/WBC ratio, MCV and MCHC was significant at $P<0.05$ level, therefore supporting the results coming out of the present study. The level of WBC decreased during the whole study due to increase in RBC level. Abdel-Tawwab et al. [49] observed that greater values of red blood cells (RBC's), hemoglobin (Hb), hematocrit (Hct), lipids and uric acids were attained when fish fed on selenium supplemented feed (0.3 or 0.5 g OS/kg) showing non-significant variations among the obtained values.

Conclusion

In conclusion, certain facts surface that the increments in weight and length and elevated RBCs count are interlinked as the counts are dropped in lower growth juvenile tilapia. Although, the selenium-deficient fish blood counts are near to normal but the continued selenium insufficiency may cause the dropped blood counts. Therefore, the present study has enlightened that the supplementation of selenium in tilapia feed at the rate of 2 mg/kg of feed does not alter its inclusive hematological profile as well as normal physiological activities rather promotes better performance and productivity to enhance fish growth and paves the way towards increased supply of selenium-fortified fish meat. Therefore, it may be recommended to include selenium (2 mg/kg) to enhance the fish vitality in dealing with physiological changes as well as with the magnitude of stress.

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