

## Comparison Between Blood Haematology of Egyptian Freshwater Food Fish and Ornamental Fish

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### Abstract

The present study was carried out to compare between some freshwater food fish species and some other freshwater ornamental fish species commonly known in Egypt, concerning the haematological parameters, dressing percent, fillet (boneless meat) percentage, and chemical composition of the fish flesh on dry matter basis. Four food fish species (Nile tilapia, grass carp, catfish, and Tobará from Manzalah Fish Farm, Dakahlia governorate, General Authority For Developing Fish Wealth, Ministry of Agriculture, Cairo, Egypt) and two ornamental fish (Koi and Fan-tail (Gold fish), from local ornamental fish trade shops, Mansoura, Dakahlia governorate, Egypt) species all belonging to the freshwater fishes were sampled, three fishes from each. Body measurements of the individual fishes were recorded, blood samples were withdrawn, live body weight was recorded, fish were dressed (eviscerated) and filleted, the flesh were minced, dried and sieved for chemical analysis. From the forgoing results, it could be concluded that the worthiness comparison among fish species for physical, biochemical and haematological parameters, and fish quality must be done between similar species, sex, size, physiological status, nutritional status, rearing system. These parameters are variable and influenced by genetically and environmental aspects.

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**Keywords:** Nile tilapia, grass carp, catfish, Koi, Fan-tail, body measurements, dressing, fillet, haematology, fillet composition.

### 1. Introduction

Abdelhamid [1] cited that ornamental fish are usually mean attractive colorful fishes of various characteristics that are kept as pets in confined space of an aquarium or a garden pool for fun and fancy. They are small sized, looking beautiful, have abnormal characters (color, form, size, customs, names, etc.) than the food fish. The big sized ones are used for human table, but they often are used for decoration, hobby, training and curing. It is a worldwide great trade.

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Ornamental fish are used too in treating patient's feet, children training, decoration, hobby, besides marketing as a trade. The industrial development of freshwater ornamental fish culture has been hampered by the lack of suitable live feeds for feeding the fish at the various production stages [2].

Fitzsimmons *et al.* [3] mentioned that tilapia fish are belonging to the ancient Egyptian fish, so aquacultured intensively in Egypt, but even worldwide as "aquatic chicken", or a cheap source of animal protein for poor people as well as a replacer for red and white meats which are expensive and carriers for infectious diseases (zoonosis). Egypt is the 2<sup>nd</sup> country (followed China) in the world ranking concerning tilapia production. Tilapia is the most familiar fish species; catching, farming and consuming. The second most famous kind just after carp in the aquaculture in the world. Not only the Egyptians (poor and rich) prefer this fish species; but also, all over the world, it is the most consumed fish. So, the yearly world production of tilapia reached 4.5 million ton [1][4].

The objectives of the present investigation was to compare between some freshwater food fish species and some other freshwater ornamental fish species commonly known in Egypt, concerning the haematological parameters, dressing percent, fillet (boneless meat) percentage, and chemical composition of the fish flesh on dry matter basis.

## 2. Materials and Methods

This study was carried out in the Department of Animal Production, Faculty of Agriculture, Mansoura University, Dakahlia governorate, Egypt during the period from 15/9/2019 till 19/6/2020. Four food fish species (Nile tilapia, grass carp, catfish, and Tobarra from Manzalah Fish Farm, Dakahlia governorate, General Authority For Developing Fish Wealth, Ministry of Agriculture, Cairo, Egypt) and two ornamental fish (Koi and Fan-tail, from local ornamental fish trade shops, Mansoura, Dakahlia governorate, Egypt) species all belonging to the freshwater fishes were sampled, three fishes from each fish species. The experimented fish were transferred into the Lab. of the Mansoura Faculty of Agriculture, Department of Animal Production. The following measurements (Table 1) were taken for the experimented fishes.

Table 1 Somebody measurements taken for the experimented fish

Fish sp.	Weight, g	Length, cm	Width, cm
Nile tilapia	205 - 315	6	4
Grass carp	955 - 1245	40 - 43	9 - 13
Catfish	620 - 850	47	7
Tobarra	295 - 320	28	5
Koi	434 - 810	40 - 43	9 - 13
Fan-tail (gold Fish)	50 - 75	4	2

Clean test tubes internally coated with EDTA (ethylene diamine tetra acetic acid) as anticoagulant were used for whole blood collection (Figs. 14 - 16) from the caudal peduncle (on 15/9/2019 and 24/10/2019, via 5ml syringes) that sent immediately to the clinical Lab. (Alsafoa Lab. for analyses, Al'Asafra, Mataria, Dakahlia governorate) for the haematological analyses. Random fish samples were taken for blood collected by special syringe, adequate amount of whole blood was withdrawn in small plastic vials containing EDTA as anticoagulant. Blood hematology as concentration of hemoglobin (Hb), total count of erythrocytes (RBCs), and total leukocytes (WBC<sub>s</sub>) were determined according to Natt and Herrick [5] and hematocrit (Hct) using Auto Counter [6] in the same lab. The other hematological parameters were mathematically calculated.

Individual fishes of each fish species were weighted, eviscerated then re-weighed deboned and reweighed for calculating both dressing and boneless meat percentages. Dressing percentage = (weight of dressed fish/ weight of whole fish) x 100 [7]. Samples of the fish fillet were minced; oven dried, ground, sieved and send (on 19/6/2020) for chemical analysis in the Regional Lab. for Foods and Feeds, Agricultural Research Center, Ministry of Agriculture, Giza, Egypt using FOSS NIRS TM DA 1650 (Denmark).

Data were analyzed by one way ANOVA to determine significant differences (P<0.05) among treatment means. All statistical analyses were done using the Statistical Package for the Social Sciences [8]. Duncan's multiple range test [9] was used to separate differences between treatment means at the probability level of 5%.

### 3. Results and Discussion

#### 3.1. Dressing and fillet percentages

Table 2 show both dressing and boneless meat percentages of the four freshwater food fish species studied. It presents significant variations in values of both criteria in favor of Nile tilapia and thin-lipped mullet for dressing % but in favor of thin-lipped mullet for the fillet %. Yet, there was no significant ( $P>0.05$ ) variation between both ornamental fish species studied herein in the dressing % but Gold fish had significantly ( $P\leq 0.05$ ) higher fillet % than Koi fish (Table 3). Table 4 illustrate a comparison among all fish species studied where from food fish or ornamental fish concerning both dressing and fillet percentages. This Table shows the presence of significant ( $P\leq 0.05$ ) differences among all fish species. The significantly ( $P\leq 0.05$ ) lowest dressing % was calculated for catfish and the highest for Nile tilapia, thin-lipped mullet, and Koi fish. The significantly ( $P\leq 0.05$ ) highest boneless meat % was calculated for Gold fish and thin-lipped mullet, all other fish species studied reflected significantly ( $P\leq 0.05$ ) lower fillet %.

Table 2 Effect of food fish species studied on dressing and boneless meat (fillet) percentages (mean  $\pm$  standard error)

Species	Dressing %	Fillet %
<b>Thin-lipped mullet</b>	85.97 <sup>ab</sup> $\pm$ 1.25	52.67 <sup>a</sup> $\pm$ 0.87
<b>Grass carp</b>	81.77 <sup>b</sup> $\pm$ 1.85	39.57 <sup>b</sup> $\pm$ 2.25
<b>African catfish</b>	73.33 <sup>c</sup> $\pm$ 2.73	43.17 <sup>b</sup> $\pm$ 0.87
<b>Nile tilapia</b>	87.87 <sup>a</sup> $\pm$ 0.82	41.03 <sup>b</sup> $\pm$ 1.41

a-c: Means in the same column superscripted with different letters significantly ( $P\leq 0.05$ ) differ.

Table 3 Effect of ornamental fish species studied on dressing and boneless meat (fillet) percentages (mean  $\pm$  standard error)

Species	Dressing %	Fillet %
<b>Koi fish</b>	83.74 $\pm$ 1.96	33.06 <sup>b</sup> $\pm$ 1.94
<b>Gold fish</b>	81.13 $\pm$ 0.57	57.00 <sup>a</sup> $\pm$ 1.53

a-b: Means in the same column superscripted with different letters significantly ( $P\leq 0.05$ ) differ.

Table 4 Effect of variable species of food fish and ornamental fish species studied on dressing and boneless meat (fillet) percentages (mean  $\pm$  standard error)

Species	Dressing %	Fillet %
<b>Thin-lipped mullet</b>	85.97 $\pm$ 1.25 <sup>ab</sup>	52.67 $\pm$ 0.87 <sup>a</sup>
<b>Grass carp</b>	81.77 $\pm$ 1.85 <sup>b</sup>	39.57 $\pm$ 2.25 <sup>b</sup>
<b>African catfish</b>	73.33 $\pm$ 2.73 <sup>c</sup>	43.17 $\pm$ 0.87 <sup>b</sup>
<b>Nile tilapia</b>	87.87 $\pm$ 0.82 <sup>a</sup>	41.03 $\pm$ 1.41 <sup>b</sup>
<b>Koi fish</b>	83.74 $\pm$ 1.96 <sup>ab</sup>	33.06 $\pm$ 1.94 <sup>b</sup>
<b>Gold fish</b>	81.13 $\pm$ 0.57 <sup>b</sup>	57.00 $\pm$ 1.53 <sup>a</sup>

a-c: Means in the same column superscripted with different letters significantly ( $P\leq 0.05$ ) differ.

These variations in the dressing and boneless meat percentages may be attributed to the differences among the studied fish species concerning their live body weight (50 – 1245 g), length (4 – 47 cm), and width (2 – 13 cm). A comparison was made of some productive traits of Stirling Nile tilapia (wild type) (*Oreochromis niloticus*, Linnaeus) and red hybrid tilapia (Florida red tilapia\_ Stirling red *O. niloticus*) males. Fillet yield was similar for both the species, with 33.4% for the red hybrid and 32.0% for *O. niloticus*. Fresh fillet lipid content was perceptibly less in the red hybrid (0.33%) than in *O. niloticus* (2.07%) [10]. However, these differences may also due to the variations in the feeding manner and habits, digestive system and feed conversion, as well as to the skeleton of the different species. Generally, Zaher [11] calculated the lean meat percentage for Nile tilapia as 1.70 – 2.03.

### 3.2. Haematological parameters

Some haematological parameters were determined, measured or calculated in the whole blood of the studied fish species (from the freshwater food fish, Table 5, ornamental fish, Table 6, as well as the comparisons of these parameters among all fish species studied (from both food fish and ornamental fish species), Table 7. There were significant ( $P \leq 0.05$ ) differences among food fish species in all tested criteria (Table 5) where thin-lipped mullet reflected the highest red blood cells count (RBCs), haemoglobin (Hb), haematocrit (Hct). Grass carp had the highest mean corpuscular volume (MCV); mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), and platelets count (PLT). Table 6 shows significant ( $P \leq 0.05$ ) differences between both ornamental fish species concerning Hb, Hct, and PLT only, with favor values for Koi than Gold fish. However, Table 7 shows the presence of significant ( $P \leq 0.05$ ) differences among fish species in all tested haematological parameters. Thin-lipped mullet had the significantly ( $P \leq 0.05$ ) highest values of RBCs, Hb, and Hct. The MCV value was the highest ( $P \leq 0.05$ ) in grass carp comparing with all tested species. Yet, Koi reflected the highest ( $P \leq 0.05$ ) values of MCH, MCHC, and PLT among all tested fish species.

White blood cells protect the body against infection. White blood cells are bigger than red blood cells and normally fewer in number. If the RBC count is low (anemia), the body may not be getting the oxygen it needs. If the count is too high, there is a risk that the red blood cells will clump together and block blood vessels. There are three red blood cell indices: mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC). They are measured by a machine and their values are determined from other measurements. The MCV shows the size of the red blood cells. The MCH value is the amount of hemoglobin in an average red blood cell. The MCHC measures the concentration of hemoglobin in an average red blood cell. These numbers help in the diagnosis of different types of anemia. Hematocrit (HCT, packed cell volume, PCV) measures the amount of space (volume) red blood cells occupy in the blood. The value is given as a percentage of red blood cells in a volume of blood. Hemoglobin is the major substance in red blood cells. It carries oxygen and gives the blood cell its red color. Platelets (thrombocytes) are the smallest type of blood cell. They play a major role in blood clotting. If there are too few platelets, uncontrolled bleeding may be a problem ([http://www.webmd.com/hw/health\\_guide\\_atoz/hw6580.asp](http://www.webmd.com/hw/health_guide_atoz/hw6580.asp) **A-Z Health Guide from WebMD: Medical Tests**).

Table 5 Effect of food fish species studied on the haematological parameters (mean  $\pm$  standard error)

Species	RBC, X $10^6/\mu\text{l}$	Hb, g/dl	Hct, %	MCV, fl	MCH, pg	MCHC, %	PLT, (X $10^3/\mu\text{l}$ )
<b>Thin-lipped mullet</b>	4.98 <sup>a</sup> $\pm 0.33$	17.23 <sup>a</sup> $\pm 1.13$	64.60 <sup>a</sup> $\pm 4.75$	129.48 <sup>b</sup> $\pm 2.79$	34.58 <sup>c</sup> $\pm 0.77$	26.70 <sup>c</sup> $\pm 0.32$	63.25 <sup>c</sup> $\pm 9.62$
<b>Grass carp</b>	2.03 <sup>b</sup> $\pm 0.18$	12.66 <sup>b</sup> $\pm 1.47$	34.72 <sup>b</sup> $\pm 3.98$	171.02 <sup>a</sup> $\pm 10.81$	63.10 <sup>a</sup> $\pm 6.39$	36.52 <sup>a</sup> $\pm 2.17$	89.80 <sup>a</sup> $\pm 19.87$
<b>African catfish</b>	2.23 <sup>b</sup> $\pm 0.14$	10.16 <sup>b</sup> $\pm 0.27$	33.46 <sup>b</sup> $\pm 3.25$	149.62 <sup>b</sup> $\pm 6.43$	46.16 <sup>b</sup> $\pm 2.48$	31.18 <sup>b</sup> $\pm 2.28$	73.00 <sup>b</sup> $\pm 15.22$
<b>Nile tilapia</b>	1.86 <sup>b</sup> $\pm 0.12$	9.84 <sup>b</sup> $\pm 0.75$	27.44 <sup>b</sup> $\pm 1.71$	147.70 <sup>b</sup> $\pm 2.37$	52.56 <sup>b</sup> $\pm 0.94$	35.90 <sup>ab</sup> $\pm 0.63$	73.40 <sup>b</sup> $\pm 15.72$

a-c: Means in the same column superscripted with different letters significantly ( $P \leq 0.05$ ) differ.

Table 6 Effect of ornamental fish species on the haematological parameters (mean  $\pm$  standard error)

Species	RBC, X $10^6/\mu\text{l}$	Hb, g/dl	Hct, %	MCV, fl	MCH, pg	MCHC, %	PLT, (X $10^3/\mu\text{l}$ )
<b>Koi fish</b>	1.68 $\pm 0.06$	13.44 <sup>a</sup> $\pm 0.48$	28.52 <sup>a</sup> $\pm 1.98$	166.66 $\pm 7.56$	81.60 $\pm 2.48$	50.92 $\pm 3.79$	94.80 <sup>a</sup> $\pm 12.49$
<b>Gold fish</b>	1.59 $\pm 0.05$	10.14 <sup>b</sup> $\pm 0.61$	23.54 <sup>b</sup> $\pm 0.42$	164.98 $\pm 3.78$	71.78 $\pm 5.40$	45.84 $\pm 3.37$	55.96 <sup>b</sup> $\pm 9.30$

a-b: Means in the same column superscripted with different letters significantly ( $P \leq 0.05$ ) differ.

Table 7 Effect of all fish species from food fish and ornamental fish studied on the haematological parameters (mean  $\pm$  standard error)

Species	RBC, X 10 <sup>6</sup> / $\mu$ l	Hb, g/dl	Hct, %	MCV, fl	MCH, pg	MCHC, %	PLT, (X 10 <sup>3</sup> / $\mu$ l
Thin-lipped mullet	4.98 <sup>a</sup>	17.23 <sup>a</sup>	64.60 <sup>a</sup>	129.48 <sup>c</sup>	34.58 <sup>d</sup>	26.70 <sup>c</sup>	63.25 <sup>d</sup>
	$\pm$ 0.33	$\pm$ 1.13	$\pm$ 4.75	$\pm$ 2.79	$\pm$ 0.77	$\pm$ 0.32	$\pm$ 9.62
Grass carp	2.03 <sup>bc</sup>	12.66 <sup>bc</sup>	34.72 <sup>b</sup>	171.02 <sup>a</sup>	63.10 <sup>c</sup>	36.52 <sup>b</sup>	89.80 <sup>b</sup>
	$\pm$ 0.18	$\pm$ 1.47	$\pm$ 3.98	$\pm$ 10.81	$\pm$ 6.39	$\pm$ 2.17	$\pm$ 19.87
African catfish	2.23 <sup>b</sup>	10.16 <sup>c</sup>	33.46 <sup>b</sup>	149.62 <sup>b</sup>	46.16 <sup>c</sup>	31.18 <sup>bc</sup>	73.00 <sup>c</sup>
	$\pm$ 0.14	$\pm$ 0.27	$\pm$ 3.25	$\pm$ 6.43	$\pm$ 2.48	$\pm$ 2.28	$\pm$ 15.22
Nile tilapia	1.86 <sup>bc</sup>	9.84 <sup>d</sup>	27.44 <sup>bc</sup>	147.70 <sup>bc</sup>	52.56 <sup>bc</sup>	35.90 <sup>b</sup>	73.40 <sup>c</sup>
	$\pm$ 0.12	$\pm$ 0.75	$\pm$ 1.71	$\pm$ 2.37	$\pm$ 0.94	$\pm$ 0.63	$\pm$ 15.72
Koi fish	1.68 <sup>c</sup>	13.44 <sup>b</sup>	28.52 <sup>bc</sup>	166.66 <sup>ab</sup>	81.60 <sup>a</sup>	50.92 <sup>a</sup>	94.80 <sup>a</sup>
	$\pm$ 0.06	$\pm$ 0.48	$\pm$ 1.98	$\pm$ 7.56	$\pm$ 2.48	$\pm$ 3.79	$\pm$ 12.49
Gold fish	1.59 <sup>c</sup>	10.14 <sup>c</sup>	23.54 <sup>c</sup>	164.98 <sup>ab</sup>	71.78 <sup>ab</sup>	45.84 <sup>a</sup>	55.96 <sup>e</sup>
	$\pm$ 0.05	$\pm$ 0.61	$\pm$ 0.42	$\pm$ 3.78	$\pm$ 5.40	$\pm$ 3.37	$\pm$ 9.30

a-e: Means in the same column superscripted with different letters significantly ( $P \leq 0.05$ ) differ.

Roberts *et al.* [12] summarized that normal hematologic parameters are not available for most pet fish species. Several reasons exist for this relative lack of data in fish. Published data exists for several significant food species but is not always applicable to pet fish. Environmental conditions such as water quality affect test results. Handling stressors, age, gender, photoperiod, and diet are other factors that can affect the results of the blood picture. They emphasized that interpretation of results in fish is difficult when compared with that in popular terrestrial species. Packed cell volume (PCV, or hematocrit) results can vary within and between species depending on age, gender, activity level, diet, stocking density, and water quality. Active species such as tuna have higher normal PCV levels, while young fish, elasmobranchs, and bottom dwellers have lower PCVs. Male fish tend to have higher PCVs when compared with female fish of the same age and sex. Anemia is typically associated with a PCV less than 20. An increased PCV ( $> 45$ ) is associated with dehydration, hypoxia, stress, and anesthesia. Elevated water temperature has been found to cause an elevated PCV in goldfish and koi. White blood cell changes occur with many diseases and adverse environmental conditions. A stress leukogram in fish reveals a leukopenia with a lymphopenia and granulocytosis. Elevated white blood cell counts can be found in freshwater fish held at higher water temperatures and those with sepsis. A decreased total count can be found during starvation. Hematology reference values for koi is packed cell volume 29.7 – 33.9 %, hemoglobin 6.3 – 8.2 g/dl, red blood cell count  $\times 10^6 / \mu$ l 1.7 – 1.9, White blood cell count  $\times 10^3 / \mu$ l 19.8 – 37, lymphocytes 74% – 93%, monocytes 0.5% – 3.4%, neutrophils 3% – 14%, and basophils 3.5% – 5.6%. Hakem [13] explained that complete blood count (CBC) meanings a complete blood picture or complete blood counts of its components. It includes the erythrocytes or red blood cells (RBCs) count, leucocytes or white blood cells (WBCs) count, platelets counts, and haemoglobin (Hgb or Hb) concentration. However, there is no standard ranges for fish haematological parameters; since, not only fish species is responsible for haematological variations [14], but also it is affected by stress factors [15], dietary supplements [16], nutritional treatments [14], immunostimulators [17], culture systems [18], and pollution [14].

Strange [19] cited that fish erythrocytes retain their nucleus. While the density of erythrocytes in fish blood varies greatly among species, a typical hematocrit in fish is about 30%, much less than the 50% usually found in mammals. Fewer RBCs are needed in fish because they have lower metabolism and thus need to move less oxygen. A twelve week feeding trial was carried out in order to assess the effect of feeding poultry hatchery waste on haematological parameters of *Clarias gariepinus* juveniles as a bio-indicator of their health status. It is concluded that using poultry hatchery waste as supplementary feed on *Clarias gariepinus* showed a slight decrease in the haematological parameters but it has no negative impact on the health status of the specie. Therefore direct use of poultry hatchery waste as sole supplementary feed should be encouraged [20]. Abdelhamid *et al.* [16] fed Nile tilapia, silver carp, common carp, and African catfish (in a poly-culture rearing system) dried sewage sludge. Sewage sludge feeding did not negatively affect the blood picture in general. Yet, Abdelhamid *et al.* [16] revealed that regardless of fish species, only RBCs, WBC and PLT reflected significant differences between both dietary treatments (dried sewage sludge versus control) with higher values for sewage sludge fed fish. However, catfish gave significantly higher values for most of the hematological parameters referring to best tolerance among the studied fish species. Except MCV, all other hematological parameters reflected significant interaction effects (diet X species), where the highest Hb and MCV values were of catfish fed the control diet, but sewage sludge fed catfish gave the highest MCHC and WBCs,

catfish fed the sewage sludge had also the highest RBCs and PCV, and control fed tilapia only had the highest MCH value. The haematological responses of *Clarias gariepinus* exposed to sub-lethal concentrations of an oilfield wastewater were investigated. PCV, Hb, WBC and ESR decreased with increasing concentration of the toxicant while platelets increased. These changes which could be used as indicators of stress in fish exposed to elevated levels of oilfield wastewater can lead to death and economic loss. Proper treatment of oilfield wastewater before discharge so as to reduce ecotoxicological problems and public health hazards is advocated [21].

However, hematological parameter of fish acts as an indicator of physiological and pathological changes in fishes. Changes in hematological parameters depend upon the aquatic biotope, fish species, age and sexual maturity and health status [22]. The physiological variables were significantly affected by dietary protein, yeast level, and their interaction. It could be concluded that the yeast supplementation as a probiotic in practical diet of Nile tilapia fry improved their challenge against *A. hydrophila* infection [23]. Abdelhamid *et al.* [18] reported significant ( $P \leq 0.05$ ) differences among treatments (stocking density of 5.0, 7.5, 10.0, 12.5, and 15 Nile tilapia fishes/m<sup>3</sup>) in all tested hematological (Hb, RBCs, Hct, MCV, MCH, MCHC, P, and WBCs), where the lower stocking densities were significantly the best ones. Haematological studies have been employed in aquaculture and are usually associated with the feed input because blood parameters have been proved to be valuable tools in determining the health status of the fish in response to the dietary manipulations. The investigated haematological indices of *Clarias gariepinus* fingerlings fed locally formulated (sinking) and commercial (floating) pelleted diets showed that the different feed forms did not significantly affect blood parameters and at such can be utilized by catfish farmers [24]. An experimental study was carried out to investigate the supplemental effects of dietary garlic (*Allium sativum*) powder on haematological parameters in fingerlings of Amur carp (*Cyprinus carpio haematopterus*) followed by challenge with *Aeromonas hydrophila*. The value of total erythrocyte count did not vary considerably ( $p > 0.05$ ). The total leucocyte count and haemoglobin resulted in significant ( $p < 0.05$ ) increment between pre challenge and post challenge in all treatment groups. It is inferred from the results of the study that dried garlic powder can be safely incorporated up to 1.5 % in carp feeds for improved haematological profile in Amur carp [25]. But the used prebiotic (POWER TOP) at the highest level (2 g / 1 kg diet) led to significantly the best Hct and lymphocytes % as well as blood glucose. The obtained blood values are within the normal ranges given for tilapia by Ayyat *et al.* [26].

Potassium permanganate (KMnO<sub>4</sub>) is a widely used freshwater aquaculture chemotherapeutant for the treatment and prevention of waterborne parasitic and fungal diseases. *C. gariepinus* were exposed to sublethal concentrations of potassium permanganate. Hb concentrations, Hct values, mean values of the RBCs count, MCH and MCHC decreased significantly ( $P < 0.05$ ) with an increase in exposure time, but the level of the mean corpuscular volume was increased. The results suggest that potassium permanganate can negatively affect the haematology of fish, causing various disturbances in its health and wellbeing. It is hereby recommended that potassium permanganate widely used in controlling external fungal, bacterial and protozoan infections of fish should not be used indiscriminately [27]. Fingerlings of *Clarias gariepinus* were exposed to different concentrations of formalin to determine its acute toxicity. The fish exhibited changes in haematological parameters as the toxicant concentration increased. Precautions in the successful use of formalin for control of ectoparasites on fish are recommended [28].

To evaluate the haematobiochemical responses associated with fipronil (sublethal concentration) exposure in *Cyprinus carpio* fry. Significant ( $P \leq 0.05$ ) increase in white blood cell counts, blood glucose, serum complement reactive protein and serum cortisol level were noticed, whereas haemoglobin and serum total protein contents were significantly ( $P \leq 0.05$ ) decreased. Thus, haemato- parameters can be used as biomarkers for the sublethal toxicity of fipronil in the water bodies [29]. The herbic Sunsate® led to significant ( $P < 0.05$ ) changes in fingerlings African catfish concerning haematological parameters as the toxicant concentration increased. Hb, RBC, Platelet count (PLT), and PCV reduced as the concentration of toxicant increased while other parameters increased proportional with the toxicant concentration [30]. To investigate the influences of sublethal toxicity of Isoprothiolan (Fuji-one), which is one of the most commonly used fungicide, an experiment was done on adult female *Oreochromis niloticus*. The induced hazard effects were obvious in blood parameters. The insignificant decrease of the RBCs, Hb and Hct values were dose dependent [31]. To investigate the effect of the insecticide Fenvalerate on haematological parameters in freshwater fish *Channa marulius*, fish was exposed to (1/4<sup>th</sup> LC50) sub lethal concentration of Fenvalerate (0.086ppm) for 96h. The result revealed that TEC, Hb percentage, PCV and MCHC counts were significantly decreased, whereas TLC, MCV and MCH increased slightly in experimental fish [32]. Chandra Sekhara Rao *et al.* [33] reviewed the haematological toxic effects to various fish species of organophosphorus pesticide Dichlorvos

which is the most widely and commonly used insecticides worldwide. These toxic effects in various aspects of ecotoxicological perturbations in fish can be viewed as biomarkers of pesticide toxicity.

### 3.3. White blood cells

Table 8 presents the significances ( $P \leq 0.05$ ) among food fish species in white blood cells count (WBCs) in favor of the grass carp. Lymphocytes % was higher ( $P \leq 0.05$ ) in thin-lipped mullet, African catfish, and grass carp than Nile tilapia. Yet, mid- and – granulocytes' percentages were the highest in Nile tilapia blood. Concerning ornamental fish, Table 9 shows significant ( $P \leq 0.05$ ) effect of fish species only in the WBCs count in favor of Koi over the Gold fish (Fan-tail). Table 10 illustrates the effect of all fish species studied from food fish and ornamental fish on white blood cells count and its differential percentages. All the Table's components reflect significantly ( $P \leq 0.05$ ) differences among fish species. The significantly ( $P \leq 0.05$ ) highest WBCs count was recorded for grass carp (to be the most disease resistant among the studied fish species). The Gold fish have the significantly ( $P \leq 0.05$ ) highest lymphocytes %, followed by Koi then thin-lipped mullet (to be more disease resistant than the other studied fish species). While Nile tilapia have the significantly ( $P \leq 0.05$ ) highest mid.-and- granulocytes %.

The use of *Bacillus subtilis* in fish diets as a probiotic to *Labeo rohita* led to significantly higher ( $P < 0.05$ ) granulocyte numbers. The result suggests that *B. subtilis* can enhance certain innate immune responses in rohu [34]. The haematological alterations produced on exposure of the African snakehead, *Parachanna africans* to the sublethal concentration of cadmium ( $Cd^{2+}$ ) for 21 days have been studied. Cadmium exposure caused significant decreases in white blood cell (WBC) count. The primary consequence of the observed changes in total and differential leucocyte counts in stressed fish was attributed to suppression of the immune system and increased susceptibility to disease [35].

Table 8 Effect of food fish species studied on white blood cells count and its differential percentages (mean  $\pm$  standard error)

Species	WBC, $X 10^3/\mu l$	Lymphocytes, %	Mid, %	Granulocytes, %
<b>Thin-lipped mullet</b>	41.40 <sup>b</sup> $\pm 2.99$	93.10 <sup>a</sup> $\pm 0.94$	5.65 <sup>b</sup> $\pm 0.53$	1.78 <sup>b</sup> $\pm 0.32$
<b>Grass carp</b>	64.74 <sup>a</sup> $\pm 5.81$	87.36 <sup>a</sup> $\pm 1.76$	8.46 <sup>b</sup> $\pm 0.94$	4.18 <sup>b</sup> $\pm 0.95$
<b>African catfish</b>	47.04 <sup>b</sup> $\pm 2.48$	88.30 <sup>a</sup> $\pm 1.36$	8.50 <sup>b</sup> $\pm 0.76$	3.20 <sup>b</sup> $\pm 0.81$
<b>Nile tilapia</b>	52.18 <sup>b</sup> $\pm 4.62$	75.28 <sup>b</sup> $\pm 4.39$	13.56 <sup>a</sup> $\pm 1.74$	11.06 <sup>a</sup> $\pm 2.74$

a-b: Means in the same column superscripted with different letters significantly ( $P \leq 0.05$ ) differ.

Table 9 Effect of ornamental fish species studied on white blood cells count and its differential percentages (mean  $\pm$  standard error)

Species	WBC, $X 10^3/\mu l$	Lymphocytes, %	Mid, %	Granulocytes, %
<b>Koi fish</b>	53.36 <sup>a</sup> $\pm 2.13$	93.70 $\pm 0.62$	4.62 $\pm 0.44$	1.68 $\pm 0.18$
<b>Gold fish</b>	41.94 <sup>b</sup> $\pm 1.21$	95.42 $\pm 1.09$	3.78 $\pm 0.61$	1.67 $\pm 0.28$

a-b: Means in the same column superscripted with different letters significantly ( $P \leq 0.05$ ) differ.

Toxicological effects of effluents from rubber processing plant were carried out. Lethal concentration was evaluated on haematological parameters. In comparison with the control, there was no significant difference in values obtained for WBC. It was concluded that the rubber processing effluent had some negative effect on the haematology of *Clarias gariepinus*. Therefore, it is recommended that the effluent should be properly treated before discharge into the environment [36].

Table 10 Effect of all fish species studied from food fish and ornamental fish on white blood cells count and its differential percentages (mean  $\pm$  standard error)

Species	WBC, X 10 <sup>3</sup> / $\mu$ l	Lymphocytes, %	Mid, %	Granulocytes, %
<b>Thin-lipped mullet</b>	41.40 <sup>c</sup> $\pm$ 2.99	93.10 <sup>ab</sup> $\pm$ 0.94	5.65 <sup>bc</sup> $\pm$ 0.53	1.78 <sup>b</sup> $\pm$ 0.32
<b>Grass carp</b>	64.74 <sup>a</sup> $\pm$ 5.81	87.36 <sup>b</sup> $\pm$ 1.76	8.46 <sup>b</sup> $\pm$ 0.94	4.18 <sup>b</sup> $\pm$ 0.95
<b>African catfish</b>	47.04 <sup>bc</sup> $\pm$ 2.48	88.30 <sup>b</sup> $\pm$ 1.36	8.50 <sup>b</sup> $\pm$ 0.76	3.20 <sup>b</sup> $\pm$ 0.81
<b>Nile tilapia</b>	52.18 <sup>bc</sup> $\pm$ 4.62	75.28 <sup>c</sup> $\pm$ 4.39	13.56 <sup>a</sup> $\pm$ 1.74	11.06 <sup>a</sup> $\pm$ 2.74
<b>Koi fish</b>	53.36 <sup>b</sup> $\pm$ 2.13	93.70 <sup>ab</sup> $\pm$ 0.62	4.62 <sup>c</sup> $\pm$ 0.44	1.68 <sup>b</sup> $\pm$ 0.18
<b>Gold fish</b>	41.94 <sup>c</sup> $\pm$ 1.21	95.42 <sup>a</sup> $\pm$ 1.09	3.78 <sup>c</sup> $\pm$ 0.61	1.67 <sup>b</sup> $\pm$ 0.28

a-c: Means in the same column superscripted with different letters significantly ( $P \leq 0.05$ ) differ.

Mohapatra *et al.* [37] reviewed the effects of probiotics on haematological parameters of fish that used as indicators of their physiological state. Since the total and differential leucocyte count in the blood provides a clue about the health status of the fish. Feeding of Gram-positive and Gram-negative probiotic bacteria led to a notably increase in the number of erythrocyte. The increased white blood cell (WBC) count helps in the non-specific immunity via neutrophils and macrophages. The WBC content of the *S. cerevisiae* fermented cassava flour diet was significantly lower than that of the control. Probiotics interact with the immune cells to enhance innate immune responses. Probiotics also actively stimulate the proliferation of lymphocytes (both B and T cells) and further immunoglobulin production in fish.

Knowledge of the haematological characteristics is an important tool that can be used as an effective and sensitive index to monitor physiological and pathological changes in fishes. The exposure of chemical pollutants can induce either increase or decrease in haematological levels. To access the level of haematological alterations of *Clarias gariepinus* exposed to varying concentration of glyphosate herbicides, the haematological alterations showed significant differences with elevations in WBC counts compared to the control. The fluctuation in WBC (significant increase in neutrophils) was an indication of post juvenile *C. gariepinus* response to the toxicity of glyphosate herbicide. The results showed that the indices surveyed are relevant in examining the health of organisms in the ecosystem and such extent of alterations are important biomarkers of stressors [38].

The disease control in aquaculture has relied on the use of chemical compounds and antibiotics. The development of non-antibiotic and environmentally friendly agents is one of the key factors for health management in aquaculture. Consequently, with the emerging need for environmentally friendly aquaculture, the use of alternatives to antibiotic growth promoters in fish nutrition is now widely accepted. In recent years, probiotics have taken center stage and are being used as an unconventional approach that has numerous beneficial effects in fish culture: improved activity of gastrointestinal microbiota and enhanced immune status and, disease resistance. As natural products, probiotics have much potential to increase the efficiency and sustainability of aquaculture production [39].

To investigate the hematological and immunological changes in catfish (*Clarias gariepinus*) exposed to cadmium. The results showed marked leukocytosis. The treatment of *C. gariepinus* with cadmium had immunosuppressive and decrease diseases resistance in a dose-dependent effect [40].

The major types of white blood cells are neutrophils, lymphocytes, monocytes, eosinophils, and basophils. Each type of cell plays a different role in protecting the body. The numbers of each one of these types of white blood cells give important information about the immune system. The most common are mature lymphocytes and the main function is to produce antibodies, immunological memory, and regulatory factors as lymphokines in response of the humoral and cell specific immune. Lymphocytes B are bone marrow derived while T is thymus derived. T cells are responsible for cell mediated immunity as well as providing assist to B lymphocytes [41].



A characteristic feature of fish is the wide physiological range of blood parameters and also the large individual variations. An attempt was under taken to compare the haematological profile of four teleost fish species (*Channa striatus*, *Cyprinus carpio*, *Catla catla* and *Labeo rohita*) and to establish the similarities and differences between these species which are widely present in the environment. The blood parameters' values confirmed differences among the four species. The findings show a higher level of white blood cell and eosinophils, in *Catla catla* with respect to the other species. The differences found can be attributed to the feeding behaviour, life style and adaptation of the different fish species to the habitat in which they dwell [42].

Deltamethrin is a pyrethroid ester insecticide widely used in agricultural and nonagricultural purposes, creates a serious threat to the environment as well as target and non-target organisms. An investigation was undertaken to study the effect of lethal and sublethal deltamethrin level on selected haematological indices of *Cirrhinus mrigala*. The results showed that the WBC were increased considerably [43].

Probiotics can be used as immunostimulants in aquaculture. The percentage of serum killing, serum nitric oxide and serum lysozyme activity were significantly increased by the time of *B. amyloliquefaciens* administration independently of the probiotic dose, and the phagocytic activity percentage was significantly decreased at the end of the experiment. Dietary supplementation with *B. amyloliquefaciens* improves immune status and disease resistance in Nile tilapia [44].

Cypermethrin 25% EC is a synthetic pyrethroid pesticide widely used for pest control. Its presence in freshwater environments is very common. Contamination of aquatic ecosystems with this toxicant affects all groups of aquatic fauna including fish which are non-target biota. A static-renewal bioassay was conducted to assess the acute and sublethal toxicity of cypermethrin on some haematological parameters of white carp (*Cirrhinus mrigala*). Juveniles of experimental fish were exposed for lethal and sublethal concentrations of cypermethrin. An increasing trend in WBC count at sublethal and decreasing trend at lethal concentration. Toxicity derived alterations observed in haematological indices during this study led to a conclusion that the cypermethrin has detrimental effect on the test fish and its presence in an aquatic environment may severely threaten the health of the ecosystem and its living component [45].

The immunomodulatory properties of Lawsonia inermis (henna) in common carp, *Cyprinus carpio* was investigated. Fish were intraperitoneally injected with 6, 60 or 600 mg kg<sup>-1</sup> body weight (BW), of the methanolic soluble fractions of *L. inermis*. The extract at 60 and 600 mg kg<sup>-1</sup> BW, significantly ( $P \leq 0.05$ ) enhanced some non-specific immune parameters such as serum lysozyme and bactericidal activity, phagocytic and respiratory burst activity, total leucocyte count (TLC), lymphocyte, monocyte and neutrophil number. This preliminary study indicates the beneficial effect of *L. inermis* in improving immune status [46].

Hematological indices are valuable tools in monitoring various aspects the health of fish exposed to contaminants. To study the effects of mercuric chloride on hematological parameters by the fresh water fish *Labeo rohita*; fishes were exposed to sublethal concentration of 1/10<sup>th</sup> 96 h LC<sub>50</sub> (0.025 mg l<sup>-1</sup>) for the period of 5, 10 and 15 days. There was a significant increase in WBC count and TLC in the mercuric chloride-treated group. So, estimation of these indices could provide a useful indicator of pollution of water bodies. The present finding clearly indicating of mercuric chloride toxicity is strong influence in aquatic environment on the hematological parameters in the fresh water fish *Labeo rohita* [47].

Haematological studies of the African lungfish *Protopterus annectens* were carried out in order to establish their mean and reference values which would serve as baseline data for assessment of the health status of the fish and as a reference point for future comparative surveys. Differences in haematological parameters including WBC were compared according to sex and seasons. Results obtained indicated variations in the values of WBC in sex and seasons. The results of the present study provide useful information for monitoring changes in the health status of fish [48].

An attempt was carried out to study the toxicity of the pesticide with respect to the hematology of fish *Oreochromis mossambicus*. The chlorpyrifos pesticide enters the body tissues of the fish that affects physiological activities. Fish

exposed to sublethal concentration of chlorpyrifos showed increase in the differential count clearly indicates that the pesticide stress certainly stimulates the white blood cells to produce more at all times of exposure [49].

Fenvalerate is a synthetic pyrethroid pesticide used in agriculture to protect a variety of crops and its exposure is associated with serious health consequences in several non-target organisms, fish being one of the most prominent among these. Propolis has antioxidant properties. The main chemical classes found in propolis are flavonoids and phenolics. Bioflavonoids are antioxidant molecules that play important roles in scavenging free radicals, which are produced in neurodegenerative diseases and aging. This study was undertaken to determine the possible protective role of propolis in fresh water fish *Cirrhinus mrigala*, exposed with fenvalerate by assessing haematological analyses in the blood. Maximum reduction was seen in the levels of WBC ( $p < 0.001$ ). The results demonstrated that the adverse effects, observed as a result of fenvalerate treatment, could be reversed by adding supplementary propolis [50].

Toxicants mainly act on circulatory system and show major impact on blood parameters. Hence, the present study revealed the effect of pesticide toxicity on blood parameters. The WBC counts were decreased significantly ( $P < 0.05$ ) in toxicant exposed fish when compared to control fish [51].

Blood profile was studied as affected by different fish species, dietary crude protein, replacements, and additives, as well as diets' types, in laboratorial and field studies. It was clear that most studied treatments (food type, probiotic level, level of clover seed wastes, initial body weight, and Teen Barshomy waste and level) had been significantly affected most studied haematological and/or biochemical parameters. That means that fish blood constituents are not stable within known ranges, but widely varied according to various environmental conditions [1]. Continued studies were undertaken to complete understanding various factors affecting blood composition of fish. In the present serial studies, we gave light on effect of some environmental and genetic factors, mainly water temperature, narcosis, handling stress, poly-culture, farm conditions, starvation, fish species, sex, and size. All these factors had affected most studied parameters of the haematology of fish blood [1]. More lights are given on effects of fish species, sex and size as well as different water bodies' locations (lake, farm, Nile) and water salinity and temperature on fish blood changes [1].

### 3.4. Chemical composition of fish flesh

From Table 11, it is clear that there are variations in the chemical composition on dry matter basis between table fish from one side and the ornamental fish on the other side, particularly in the crude protein (CP) content; since food (table) fish flesh contained higher values (64.32 – 86.19 %) than the ornamental ones (58.10 – 59.50 %). Also, there are variations among different fish species, these variations may be due to the other components too, rather than CP (fat, ash and fibers).

Table 11 Chemical composition of the fish flesh (% dry matter basis)

<b>Fish species</b>	<b>Protein</b>	<b>Fat</b>	<b>Ash</b>	<b>Fibers</b>
<b>Tobara</b>	64.32	24.32	7.58	4.74
<b>Grass carp</b>	67.06	16.47	5.88	4.59
<b>Catfish</b>	86.19	19.13	9.92	1.18
<b>Nile tilapia</b>	75.12	8.14	3.49	8.49
<b>Koi</b>	59.50	11.88	5.58	4.51
<b>Gold fish</b>	58.10	13.93	2.26	4.76

Body protein content of African bagrid catfish, *Chrysichthys nigrodigitatus* (Lacepède, 1803) was not affected by dietary crude protein, but body lipid increased significantly ( $P < 0.05$ ) with increasing dietary protein levels [52]. The utilization of *Leucaena leucocephala* seed meal (LSM) for sustainable fish production was examined. Fish carcass protein was statistically the same for 0%, 20% and 40% LSM fed fish. In the present study processed leucaena seed meal can be considered as a good alternative raw material in substitution to soya bean meal for *Clarias gariepinus* fingerlings' diets at 20% inclusion level [53].

Comparison between *O. niloticus* and *O. aureus* and their interspecific hybrid ( $\text{♂ } O. aureus \times \text{♀ } O. niloticus$ ) revealed that both purebred genotypes and their interspecific hybrid had similar moisture, crude protein content and crude lipid content ( $P > 0.05$ ); however, it should be noted that value of the crude lipid content was lower in the interspecific hybrid ( $\text{♂ } O. aureus \times \text{♀ } O. niloticus$ ) than in purebred genotypes. Meanwhile, the hybrid dress-out% was intermediate to the purebred parental genotypes [54]. Fry monosex Nile tilapia (*Oreochromis niloticus*) were fed in five dietary protein levels (21%, 25%, 32%, 37% and 45%) to investigate casual effects on carcass composition. There was no significant difference in protein body content of tilapia fed on five diets as compared to the initial fish. Lipid body content increased significantly with high dietary protein levels from 21.88% to 45.50%. The carcass crude lipid was recorded as higher (9.4%) in the fry fed on diet containing 45.50% protein, followed by fish fed on diet having protein 21.88% [55].

Interaction between oxygen and stocking density on juvenile tilapia performance was tested under intraperitoneally injection by pathogenic bacteria, *Aeromonas hydrophila*. Crude protein and total lipid in whole-fish body decreased significantly at low dissolved oxygen (LDO), while total lipid content decreased also at high stocking density (SD) [56]. To evaluate the effect of prebiotics and probiotics on chemical composition of *Cyprinus carpio*. *Cyprinus carpio* fry were used in experiments with Organoferum dry prebiotic and Biogreen E probiotic. The highest crude protein and lipid content ( $P < 0.05$ ) were found in the fish fed the diets with 2.5 g kg<sup>-1</sup> Organoferum dry prebiotic and 1 g kg<sup>-1</sup> Biogreen E probiotic, respectively [57]. A study was conducted to investigate the effects of dietary inulin on carcass composition of carp (*Cyprinus carpio*) fry. After acclimation, fish were fed, control diet (0 g) or diets containing 5 g and 10 g inulin kg<sup>-1</sup> for 7 weeks. Supplementation of inulin significantly increased carcass lipid content, while carcass protein content significantly decreased [58].

In a comparison between hybrid red tilapia (*Oreochromis niloticus* × *O. mossambicus*) and Nile tilapia (*O. niloticus*), condition factor was found to be significantly different among the groups ( $P < 0.01$ ). The values of biochemical constituents including fatty acids of the fish did not show prominent changes ( $P < 0.05$ ), though little variations were noted in the values of individual fatty acids [59]. Kim *et al.* [60] evaluated the effects of dietary protein levels on body composition of juvenile parrot fish *Oplegnathus fasciatus*. Five isocaloric diets (16.7 kJ/g energy) were formulated to contain crude protein levels (CP) as 35 (CP35), 40 (CP40), 45 (CP45), 50 (CP50) and 60 % (CP60). At the end of 8-week feeding trial, whole-body crude protein and lipid contents increased with the dietary protein level up to CP50 diet. Organic acids or their salts can be used as feed additives in aquaculture. This study was conducted to evaluate the use of a mixture of formic acid, propionic acid and calcium propionate compared with oxytetracycline (OTC) with *Oreochromis niloticus* fingerlings. The protein and fat contents of the whole body were the highest in the propionic group [61].

A study was conducted to evaluate the effect of fenugreek as a feed additive on the body composition of striped catfish *Pangasius hypophthalmus*. Proximate analysis revealed significant differences ( $P \leq 0.05$ ) among treatments and control for crude protein while rest of the parameters remained same. It is concluded that addition of fenugreek in fish feed can improve protein content of the fish meat [62]. Increasing the stocking density of fish significantly decreased body composition (crude protein and ash) [63].

Transport of living fish is an important operation in fish farms. This study investigated the effect of transport stress on the muscular chemical composition and flesh quality of channel catfish *Ictalurus punctatus* fingerlings. Fish were divided into two groups, the control group and the treatment group. The fish in the treatment group were exposed to the transportation process (3.5 h), while the control fish were kept under the normal aquaculture conditions (not-transport). Samples of muscle were collected for assay at several time intervals: before the experiment (basal level), and at 0, 24, 72 and 168 h after transportation process. Transportation caused significant decrease in water, fat and energy contents of muscle after 24 h compared to other time intervals and non-transported fish. It is concluded that the transportation process results in marked changes in muscular chemical composition and reduces the flesh quality of *I. punctatus* fingerlings [64].

Nile tilapia was fed diets contained yeast extract rich in nucleotides and  $\beta$ -glucan. No significant ( $P > 0.05$ ) differences were found in dry matter, protein, lipid or ash contents among the treatment groups [65]. To assess the effect of different levels of the probiotic Aqua-Max Plus® on body chemical composition of the mono-sex Nile tilapia (*Oreochromis niloticus*, L. 1758), a study was conducted. Results exhibited that the addition of Aqua-Max

Plus® at levels 2, 3, and 4 g/kg diet, it was found that by increasing the levels of Aqua-Max Plus®, ash and protein contents were significantly increased, while fat and energy content contents were decreased [66].

In 2006, the European Union banned the use of all types of therapeutic antibiotics as growth promoters in livestock farming. This led to the need for alternative growth promoters supplements to be found. Such a possibility is the development of new nutritional strategies with the participation of probiotics. To trace the meat quality and blood parameters of rainbow trout (*Oncorhynchus mykiss* W.) fed with additive of probiotic Proviotic®; meat quality parameters were measured at the end of experiment. This supplement, added to extruded pellets for feeding of rainbow trout increases the quantity of crude protein and ash in fish fillets and this way is improving the quality of fish meat [67].

**Conclusively**, from the forgoing results, it could be concluded that the worthiness comparison among fish species for physical, biochemical and haematological parameters, and fish quality must be done between similar species, sex, size, physiological status, nutritional status, rearing system. These parameters are variable and influenced by genetically and environmental aspects.

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