# Influence of Thyme (*Thymus Vulgaris)* As Feed Additives on Growth Performance and Antifungal Activity on *Saprolegnia Spp.* In *Cyprinus carpio L.*

Ameer H. ALsafah and Jamal K. A. AL-Faragi

Department of Pathology, College of Veterinary Medicine, University of Baghdad, Baghdad, Iraq.
amee98r@gmail.com

**Abstract:** The present study was aimed to evaluate the efficiency of thyme *(Thymus vulgaris)* on growth performance and antifungal activity of common carp. A total of 100 fingerlings of *Cyprinus carpio* weight ranged between 75.18 -75.36 g were randomly distributed into five treatment groups. Fish were fed different thyme concentration: 0. 5% (T2), 1% (T3), 1.5% (T4) and 2% (T5). The first group (T1) control group was fed 0% of thyme in the diet. All treatments fed twice a day at rate 3% B.W for 56 days. After 56 days of feeding, blood samples (RBC, Hb, PCV and WBC) were taken and biochemical test (Total protein, albumin and globulin). Then Challenge with Saprolegnia with a concentration of 1 Petri dish per 20 liters then did blood picture and biochemical tests and histological study. The study was conducted at College of Veterinary Medicine/ University of Baghdad, Ichthyology laboratory, Baghdad, Iraq, from 14/2/ to 24/4/2017. Results of feeding trail in treated groups with thyme supplementation showed significant increase ($P< 0.05$) in all growth, haematological and biochemical parameters compared with the control group, and the highest values were recorded in T4 supplemented with thyme 1.5%. That suggesting that the thyme improve growth rate and survival rate in common carp.
[Ameer H. ALsafah and Jamal K. A. AL-Faragi. **Influence of Thyme (*Thymus Vulgaris)* As Feed Additives on Growth Performance and Antifungal Activity on *Saprolegnia Spp.* In *Cyprinus carpio L.*** *J Am Sci* 2018;14(2):48-54]. ISSN 1545-1003 (print); ISSN 2375-7264 (online). http://www.jofamericanscience.org. 6. doi:10.7537/marsjas140218.06.

**Key words:** Common carp, thyme, saprolegnia, growth performance

## 1. Introduction

Fish production increased rapidly in the last years by several methods such as the establishment of fish farms and intensive rearing in cages, closed recirculating system and artificial breeding [1, 2], that encouraged researchers to develop fish production in different scientific ways to raise quality and quantity production and lower costs, which including special fish diets supplemented with growth promoters as a feed additives to improve and raise the production level without any negative effect on public health of the consumer [1]. Fishes are one of the important methods to provide food for human consumption in the world [2].

Chemicals and antibiotics are widely used to control or prevent infection by many diseases, but if use of high doses or long period will lead to side effects and sometime fish die, antibiotics have negative effect on immune system, in addition to accumulate residual in tissues [3], as well as development of pathogenic resistance to the drug [4]. Also, the use of chemicals and antibiotics lead to prevent growth of aquatic organisms which benefit as natural food for fish.

For these reasons scientists trended toward the use of alternative treatments such as feed additives which are added to the fish feed to improve the growth and production and to stimulate immune system and increase resistance to different diseases as well as lower side effect comparison with chemically synthesized drugs [5].

One of the new substances which affect positively body gain increasement and which decrease food consumption, include thyme, this substance share in the metabolic processes. Thyme (*Thymus vulgaris*) is a member of Lamiaceae family. Thyme has strong antimicrobial and antioxidant activity due to its very high contents of thymol (40%), carvacrol (15%), cymene, eugenol and 4-allylphenol [6]. Thyme is a bronchial antispasmodic and an expectorant. It has shown antibacterial, antifungal, antiviral, antiprotozoal, and antioxidant properties.

The *Saprolegnia* particularly are responsible for significant infections including both dead, living fish and ova in the facilities of the aquaculture [7]. Usually the Saprolegniasis in fish begins with a cotton wool like, white to gray or brownish growth over head, dorsal fin and then spread all over the body [8]. In fish eggs, the hyphae penetrating of the chorionic membrane which regulates the embryo osmosis, this is suggested to be the cause for egg mortalities [9]. the aim of this study was:

A-Evaluate the efficiency of dietary thyme on growth rate.

B-Evaluate the efficiency of dietary thyme as antifungal activity.

C-Study the effect of dietary thyme supplementation on hematological parameters in common carp.

## 2. Materials and Methods

The present study was carried out at College of Veterinary Medicine/ University of Baghdad, Ichthyology laboratory, Baghdad, Iraq, from 14/2/ to 24/4/2017.

**Fish and Experimental condition:** A total of 100 fingerlings of *C. carpio* weight ranged between 75.18- 75.36 g were used in this experiment, were provided from a commercial hatchery (Al-Messayab, Babylon). Initially, fish were immersed in a salt bath of NaCl at concentration 2.5% for five minutes to get rid of external parasites and fungal infections, after that two weeks of acclimation for fish before starting the experiment and fed on commercial diet (at rate 1% of their body weight). During this time, they were stocked in three bath trough (150 × 40 × 20 cm). Then, 100 fish were randomly selected and then distributed into 10 glass aquaria filled with chlorine-free tap water and supplied with air pump at rate of 10 fish per aquarium (two replicates/ treatment) were maintained for each of the five treatments (T2, T3, T4, T5 and control (T1)). Chemo-physical parameters of water were measured by Lovibond water testing apparatus. Water temperature was adjusted at 24-26 ºC and recorded daily; pH (6.5-7.2) and dissolved oxygen (6.25-6.45 mg/L) were noted every days.

Fish were fed basic diet supplemented with different thyme concentration: 0.5% (T2), 1% (T3), 1.5% (T4). 2% (T5) and control group (T1) was fed basic diet without any addition of thyme in the diet. All treatments fed twice a day at rate about 3% of body weight for 56 days and take blood sample to hematology and biochemical testes. After 56 days challenge with *Saprolegnia spp.* Blood Samples were tanked to hematology and biochemical testes.

**Statistical analysis**

All values were statically analysis using SAS (Statistical Analysis System –version 9.1). One -way ANOVA with Least significant differences (LSD). $P < 0.05$ was considered significant values [10].

## 3. Results

### 3.1. Growth performance

Data of average body weight of experimental fish (*C. carpio)* are summarized in Table 1. The initial weight of all groups at first day of experimental period ranged between 75.18–75.36 g and there was no statistically differences among them ($P > 0.05$). The effect of thyme supplementation on growth performance was observed during the first 15 days. After 15, 30, 45 and 56 days all groups showed significantly differences ($P < 0.05$) among them. Moreover, at 56 days of the experimental period, all treatments showed the existence of a clear increase in the growth rate compared with the control group. Results showed that the body weight increased significantly ($P< 0.05$) with advanced age in all groups (Table 1). The fourth group (T4) surpassed on the other groups. The superiority of this group was detected in the second period (at 15 day) (82.21 g) and continued till the end of experiment (145.46 g).

**Table 1: Average body weight of C. *carpio* fed different level of thyme and control diet (±SE) during 56 days.**

| Weight (g) / Treatment | 1 Day | 15 Day | 30 Day | 45 Day | 56 DAY |
|---|---|---|---|---|---|
| **T1** | **75.34±0.24 a** | **77.92±0.56 b** | **85.02±0.56 e** | **98.26±1.14 d** | **115.40±0.56 d** |
| **T2** | **75.18±0.56 a** | **79.17±0.27 b** | **88.2±0.15 d** | **105.74±0.26 d** | **122.88±0.58 e** |
| **T3** | **75.24±0.20 a** | **80.97±0.28 ab** | **91.46±0.27 ab** | **114.91±0.42 b** | **138.24±1.15 b** |
| **T4** | **75.36±0.16 a** | **82.21±0.15 a** | **94.01±0.15 a** | **118.39±0.65 a** | **145.46±1.16 a** |
| **T5** | **75.26±0.14 a** | **79.82±0.14 b** | **89.77±0.14 bc** | **107.51±0.20 c** | **128.56±0.58 c** |

Means with different small letter in the same column differ significantly ($P< 0.05$).

### 3.2. Haematological parameters

#### 3.2.1. Red blood cells count (RBC)

The results revealed a significant increases ($P< 0.05$) in RBCs count of *Cyprinus carpio* at 56 days (pre challenge) in treatments T2,T3,T4 and T5 compared with control group T1. Also, similar results were revealed in RBCs count at post challenge (day 70) with slight decrease in values compared with pre challenge table (2).

#### 3.2.2. Hemoglobin (Hb) and Packed cell volume (PCV)

In table (2), there were a significant increases ($P< 0.05$) in hemoglobin concentration and packed cell volume at 56 days (pre challenge) in groups T2, T3, T4 and T5 compared with control group (T1), and recorded highest values in T4 from all other treatments. Also, similar results were revealed in Hb concentration and PCV% at 70 days (post challenge)

with slight decreases in values compared with pre challenge values.

### 3.2.3. Total WBCs count

WBCs count showed significant increase (P ≤0.05) in thyme feeding group of fish (T2, T3,T4 and T5) in comparison with the control group (T1) at 56 days (pre challenge). The highest mean value was observed in T4 on 56th day ( 23.01x$10^3$ cell / $mm^3$) ) in comparison with other treatments. Also, in 70 days WBC numbers increased significantly (P≤ 0.05) in treatment groups.

**Table (2) Blood parameter of *C. carpio* with thyme supplement and control diet during 56 days (pre challenge) and 70 days (post challenge).**

| Parameters / Groups | RBC $10^6$/$mm^3$ | | Hb g/100ml | | PCV % | | WBC $10^3$/$mm^3$ | |
|---|---|---|---|---|---|---|---|---|
| | 56 days | 70 days | 56 days | 70 days | 56 days | 70 days | **56 days** | 70 days |
| **T1** | **1.83±0.11 b A** | **1.80±0.11 b A** | **7.57±1.15 d A** | **7.24±1.15 b A** | **22.5±1.15 b A** | **22.3±1.15 b A** | **18.3±1.15 b A** | **19.5±1.70 b A** |
| **T2** | **1.90±0.11 b A** | **1.88±0.11 b A** | **7.84±1.15 d A** | **7.83±1.15 c A** | **25±1.15 ab A** | **24.76±1.15 ab A** | **20.73±1.15 ab A** | **22.35±1.15 a A** |
| **T3** | **2.15±0.11 ab A** | **2.12±0.11 ab A** | **8.75±1.15 b A** | **8.70±1.15 a A** | **27±1.15 a A** | **26.25±1.15 a A** | **22.27±1.15 ab A** | **24.94±1.15 a A** |
| **T4** | **2.50±0.11 a A** | **2.49±0.11 a A** | **9.30±1.15 a A** | **8.750±1.15 a A** | **28±1.15 a A** | **27±1.15 a A** | **23.01±1.15 a A** | **25.56±1.15 a A** |
| **T5** | **2.05±0.11 b A** | **2.04±0.11 b A** | **8.33±1.15 c A** | **8.3±1.15 b A** | **26.3±1.15 ab A** | **26.08±1.15 a A** | **21.25±1.15 ab A** | **24.83±1.15 a A** |

Mean with different small letter in the same column significantly (P<0.05).

### 3.2.4. Differential leukocyte count

The count of Differential leukocyte revealed significant differences among treatments groups. Lymphocyte showed a significant increase (P<0.05) treated groups that supplemented by thyme in diet (T2, T3, T4 and T5 ) in comparison with the control group T1 at 56 and 70 days. A significant increase (P≤ 0.05) was recorded in T4 on 56th day (pre-challenge) in comparison with other treatment and control group.

Neutrophil showed a significant increase (P<0.05) in common carp groups that supplemented thyme in diet (T2, T3, T4 and T5) in comparison with the control diet group T1 on 56 and 70 days. A significant increase (P≤ 0.05) was observed in T4 on 56th days (before challenge) in comparison with further treatment and control group.

Monocytes treated groups significantly increased (P≤0.05) after challenged (day 70) than pre challenged groups (56 day). The maximum values were observed in T4 followed by T3, T5 and T2 respectively in comparison with control group T1.

Eosinophil showed a significant increase (P≤0.05) in comparison with the control groups.

**Table (3) Effect of thyme on differential leukocyte count in treatment groups *C. carpio* fed different concentration for 56 days (pre challenge) and 70 days post challenged with *Saprolegnia*.**

| Parameters / Groups | **Lymphocyte %** | | **Neutrophil %** | | **Monocyte %** | | **Eosinophil %** | |
|---|---|---|---|---|---|---|---|---|
| | 56 days | 70 days | 56 days | 70 days | 56 days | 70 days | 56 days | 70 days |
| **T1** | **54.6±0.05 e A** | **49.5±0.06 d A** | **46.6±0.05 e A** | **50.3±0.06 e A** | **2.30±0.03 e A** | **2.77±0.06 e A** | **2.0±0.02 e A** | **2.10±0.02 e A** |
| **T2** | **54.1±0.06 d A** | **50±0.06 dc A** | **49.8±0.06 d A** | **55.1±0.06 d A** | **2.80±0.06 d A** | **3.0±0.03 d A** | **2.04±0.02 d A** | **2.60±0.06 d A** |
| **T3** | **58.6±0.06 b A** | **53±0.06 b A** | **52.5±0.06 b A** | **61.8±0.06 b A** | **4.20±0.03 b A** | **4.50±0.06 b A** | **2.15±0.06 b A** | **2.83±0.06 bc A** |
| **T4** | **60±0.06 a A** | **55±0.06 a A** | **53.6±0.06 a A** | **65.1±0.03 a A** | **4.75±0.03 a A** | **4.90±0.06 a A** | **2.17±0.03 a A** | **3.10±0.06 a A** |
| **T5** | **55.6±0.06 c A** | **50.5±0.06 c A** | **50.5±0.06 c A** | **60.6±0.06 c A** | **3.60±0.06 c A** | **3.80±0.03 c A** | **2.05±0.03 c A** | **2.73±0.02 c A** |

Mean with different small letter in the same column significantly (P<0.05).

### 3.3. Results of total protein, albumin, globulin, AG ratio

Total protein level significantly increased (P > 0.05) at 56 days (pre challenge) in all groups compared with control group T1, the high level were recorded in T4. Also, there were significant increases (P< 0.05) at 70 days (post challenge) in all groups compared with control group, the high level was recorded in T4. While albumin content in 56 day and in 70 day (post challenge) table (4) revealed slightly

increase insignificant difference in all treatment compared with control group. The post challenge values in T2,T3,T4 and T5 were (1.26, 1.27, 1.29 and 1.27 g/dl respectively) compared to the control treatment T1 (1.23g/dl) table (4).

While globulin content increased significantly (P< 0.05) in T2, T3, T4 and T5 compared with control group T1 at 56 days (pre challenge). Also, globulin increased in T2, T3, T4 and T5 ( 2.93, 3.40, 3.45 and 3.20 g/dl respectively) compared with control group T1 (2.50 g/dl ) at 70 days post challenge table (4).

Albumin globulin ratio in table (4) slightly decreased significantly (P< 0.05) in treated groups (T2,T3,T4 and T5) compared with control group T1 at 56 days, the best value show in T4 (0.39) from other groups,Also in post challenge (70 days) decreased significantly (P< 0.05) in treated groups (T2,T3,T4 and T5) compared with control group T1. In T2,T3,T4 and T5 show no differ significantly in all treatments pre and post challenge.

**Table (4) Biochemical parameters of *C.carpio* at 56 days (pre challenge) and 70 days (post challenge).**

| Parameters / Groups | Total protein g/dcl | | Albumin mg/dcl | | Globulin mg/dcl | | A/G % | |
|---|---|---|---|---|---|---|---|---|
| | 56 days | 70 days | 56 days | 70 days | 56 days | 70 days | 56 days | 70 days |
| T1 | 3.61±0.06 c A | 3.80±1.37 c A | 1.20±0.021 a A | 1.23±1.34 a A | 2.24±0.02 e A | 2.50±1.44 e A | 0.53±1.44 a A | 0.53±0.02 a A |
| T2 | 4.44±0.05 cd A | 4.52±1.81 cd A | 1.22±0.028 a A | 1.26±1.75 a A | 2.66±0.19 d A | 2.93±0.01 d A | 0.45±0.01 b A | 0.43±0.19 b A |
| T3 | 4.50±0.08 bc A | 4.70±1.89 b A | 1.24±0.021 a A | 1.27±1.83 a A | 3.09±0.06 bc A | 3.40±2.78 ab A | 0.40±6.55 b A | 0.40±0.06 b A |
| T4 | 4.55±0.02 a A | 4.84±2.00 a A | 1.25±0.021 a A | 1.29±2.02 a A | 3.15±0.02 a A | 3.45±0.02 a A | 0.39±2.78 b A | 0.39±0.02 b A |
| T5 | 4.45±0.08 c A | 4.59±1.98 cd A | 1.24±0.028 a A | 1.27±1.94 a A | 3.06±0.02 c A | 3.20±0.02 c A | 0.42±0.07 b A | 0.41±0.06 b A |

Mean with different small letter in the same column significantly (P<0.05).

## 4. Discussion

### 4.1. Growth Performance

The enhanced growth rate in treated fish indicated by thyme supplementation in this present study may be due to improved feed consumption, improved feed utilization, which is an indication of increased nutrient digestibility and digestive tonic and antioxidant activity of thyme and it stimulating appetite of fish that stimulates protein synthesis by enzymatic system that result reported by [11]. studied the influence of some photobiotic (thyme, seabuckthorn) on growth performance of stellate sturgeon (*A. stellatus, pallas*, 1771) in an industrial recirculating aquaculture system indicate that thyme acted as a digestion tonic, stimulating the appetite of fish, which was deducted from their behavior at meal time and also from the values obtained for the final mean weight: V1 (seabuckthorn ) –219.66 g/fish; V2 (thyme ) - 228.68 g/fish and the individual growth rate: V1 98.45 g; V2–105.90 g, as a conclusion, it can be said that the two types of photobiotic (thyme, seabuckthorn), administered in a concentration of 2%/kg fodder, have influenced the growth performance of stellate.

Most probably fat was used for energy, and protein was used for growth in thyme diet. It has been shown that herbs stimulate the secretion of pancreatic enzymes, important factors in nutrient digestion and assimilation [12].

These result in line with [13] who found that the increase in specific growth rate ( 95±03), effective protein yield (13±03), percentage of weight obtained (35±46) and feed conversion efficiency (96±06) also increased significantly in thyme treatment compared to control group (49±09) (57± 1) (69± 42) (23± 9) (P<0.05) On the other hand, the presence of compounds such as saponin (which is one of the components of thyme) also stimulates the growth of rainbow trout [14].

The obtained results are in agreement with those obtained by [15] who reported that thyme supplemented diets significantly improved growth performance of stellatus sturgeons and their optimal growth was obtained at 2 % thyme /kg diet.

Also, [16] stated that *Oreochromis niloticus* fed with thyme (1% and 2%) levels had positive effect on growth performance. On the other hand, [17] showed that was no enhancement in growth performance of stellatus juvenile after feeding 1% of dietary thyme, But has led to an improvement of meat biochemical quality composition, due to a significant decrease in the percentage of water, respectively increasing the percentage of protein, fact that reveals a better meat nutritional value.

These results agreement with [18] exhibited that feeding fish with thyme result in elevated disease resistance and improved survival, which may be attributed to an improvement of immune function. Also, [19] they see the highest significant value was in

group supplemented with thyme followed by group supplemented by ginger in comparison with control group fed on basal diet ( P<0.05).

**4.2. Hematology**

The result of the RBCs, HB and PCV agreement with those obtained by [20] that studied effects of 1% of thyme supplementation at Nile tilapia and found that the significant differences (p<0.05) in case of RBCc (1.97) compare with control (1.60), in case HB ( 8.47) compare with control ( 8.45) and in case PCV (30) compare with control (25.60).

The [21] who found that the adding thymol-carvacrol to the diet had slightly effect on PCV, Hb and RBC (42.5), (4.8) and (1.24) compared with control (38), (4.5) and (1.16) whereas supplementation with thymol and carvacrol caused an increase in lymphocyte percent (P < 0.05) (95) compared with control (92.16).

Similar observations were reported by [22] who found the RBC count, Hb concentration and PCV in the treatment groups by carvacrol (3.78), (5.26) and (37.8) did not vary significantly from the values observed for the control group (3.55), (5.07) and (35.60).

The [23] who found that the PCV were increased about (36.33) compared with control (34.5) The increased value of the PCV after 1 and 2 months of feeding, indicated the safety of the probiotics used and their efficacy in improving the health status as a reduced PCV can indicate that fish are not eating or are suffering infections.

These results in line with the results of [24] who found the HB, RBC and PCV in group fed 1 % of thyme supplementation at 45 days had significantly (P < 0.05) (12), (1.90) and (34) compared with control (11), (1.40) and (30), Lymphocyte, neutrophil and monocyte percentages (62), (34) and (2.50) significantly changed in groups thyme when compared to control (P < 0.05) (45), (10) and (0.60).

The obtained results are in agreement with those obtained by [25] who found that the hematological values obtained in this study indicated that the means of total red blood cells, packed cells volume, total white blood cells and Hb were significantly higher (P≤0.05) in group 2 (chicks fed basal diet supplemented with 1% thyme (4.61 ), (33 ), (23.01 ) and (11.59 ) as compared with control group (3.90), ( 29.08), (20) and (9.03).

Similar observations were reported by [26] who found that the monocyte have significantly increase in catfish fed diet contains 1% thyme (37.75) when compared to basic diet (34.83).

The [27] who found that the Rosmary administration to *Oreochromis niloticus* were significant reduction in Hb, (6.32) compared with control (6.74).

Generally, the blood profile of any fish can change with the fish species, age, cycle of sexual maturity and health conditions [28].

Generally, fish hematocrit varies depending on physiology, health and activity of the fish and large fluctuations indicate, according to some authors [29] stressful conditions or even chronic stress appearance.

**4.3. Biochemical Profile**

Total serum protein represented the most important indicator of the nutritional

state of the fish health condition [30]. Some authors reported that the concentrations of total protein, albumin and globulin in plasma represent indicators of liver function and therefore the decrease of serum protein could be attributed to renal excretion or impaired protein synthesis, or due to liver hypofunction or disorder [31].

Albumin and globulin are two important parts of total protein (TP), and changes in these parameters affect the level of TP. Albumin and globulin concentration are commonly used for evaluating the effect of nutrients on fish immunity. Albumin in fish blood performs the transportation of lipids and helps in the general metabolism of fish [32]. Globulin considered very important to keeping of good immunity and also have all the immune globulins in blood. Another suggested that increase in albumin and globulin contents gives evidence about strong non-specific immune response in fish [33].

The results of the total protein and albumin and albumin globulin ratio are get along with [34] who found that the total protein content increased in all experimental groups compared to the control group. The highest content of total protein was determined in group OIL2 (fish with oil supplementation from thyme and fennel) (14.56 ± 1.75 g·dl-1). Generally, albumin ratio (4.04) was not significantly different from the control group (3.46) throughout the whole experiment ($P > 0.05$).

The obtained results are Similar observations were reported by [35] who found that the total serum protein content in experimental groups was significantly (P ≤ 0.05) (2.95) higher compared to control group (2.20) over the exposure duration. Similarly, there was a significantly higher albumin content in the experimental groups (1.31) compared with the control group (0.96) over all the assay period, globulin content was significantly (p ≤ 0.05) higher in group C and group D (1.66) in comparison with the control (1.23), albumin / globulin ratio was significantly (p ≤ 0.05) higher in fish fed with 1 g/kg feed on day 20 and day 60 and also in all the groups after challenge.

**5. Conclusions**

1. The results displayed positive effects of thyme supplemented on growth performance and healthy status when adding to diet of common carp in all treatments.

2. The largest value on growth rate was recorded in T4, which is supplemented with 1.5%. of thyme.

3- The obtained results showed a significant increase in RBCs count in treatments supplemented diet with thyme compared with control group. Also, WBCs count revealed a significant increase at 56 days and 70 days (pre and post challenge with *Saprolegnia* ) in all treatments as compared with control group.

4. There was positive effect of thyme on biochemical profile when supplemented to diet of common carp.

5. All the concentrations of thyme (T2, T3, T4 and T5) have proven a beneficial and protective effect against *Saprolegniasis*-infected *Cyprinus carpio*.

**6. Acknowledgment**

The first who deserve all thanks is Allah the most gracious, most merciful for granting me strength and help with which this research has been accomplished and I would like to thank Dr. Noor M Salman, Dr. Ahmed Abduljabbar Ashour and Dr. Sabah Mahmood from college of Veterinary Medicine, University of Baghdad, Iraq, for assist me in publishing this paper.

**References**

1. Al-Shamaa AA. Fisheries in the marshes of southern Iraq and ways to protection and development. Scientific Research Councils, Iraq, Baghdad Conference, 18 -29, 1993.
2. FAO. United Nations, the global aquaculture production has increased from about 28.3 million tonnes. 2009, p 99.
3. Harikrishnan R, Balasundaram C, Kim MC, Kim JS, Han YJ, Heo MS. Innate immune response and disease resistance in Carassius auratus by triherba L solvent extracts. Fish Shellfish Immunol. 2009; 27:508-515.
4. Smith P, Hiney MP, Samuelesen OB. Bacterial resistance to antimicrobial agent used in fish farming: a critical evaluation of method and meaning. Annu. Rev. Fish Dis. 1994; 4:273-313.
5. Akinpelu DA, Onakoya TM. Antimicrobial activities of medicinal plants used in folklore remedies in south-western. African Journal of Biochemistry. 2006; 5:1078-1081.
6. Rota, M. C.; Herrera, A.; Martinez, R. M.; Sotomayor, J. A. and Jordan, M.J. (2008). Antimicrobial activity and chemical composition of Thymus vulgaris, Thymus zygis and Thymus hyemalis essential oils. Food Control 19(7):681-687.
7. Wolinska, J.; King, K.C.; Vigneux, F. and Lively, C.M. (2008). Virulence, cultivating conditions, and phylogenetic analyses of oomycete parasites in Daphnia. Parasitology, 135:1667-1678.
8. Zaki, M. S.; Fawzi O.M. and El-Jackey J. (2008). Pathological and biochemical studies in Tilapia nilotica infected with saprolegnia parasitica and treated with potassium permanganate. American-Eurasiang. Agric. and Environ.Sci., 3(5): 677-680.
9. Liu, Y.; de Bruijn, I.; Jack, A.L.; Drynan, K. van den Berg, A.H.; Thoen, E.; Sandoval-Sierra, V.; Skaar, I.; van West, P.; Diéguez- Uribeondo, J.; van der Voort, M.; Mendes, R.; Mazzola, M. and Raaijmakers, J.M. (2014). Deciphering microbial landscapes of fish eggs to mitigate emerging diseases. The ISME Journal, 8(10):2002-2014.
10. S. SAS/STAT Users Guide for Personal Computer. Release 9.1.SAS Institute, Inc., Cary, N.C., USA. 2010.
11. Dorojan, O. V.; Cristea, V. and Petrea, Ş. M. (2014). The Influence of some Phytobiotics ( Thyme, Seabuckthorn ) on Growth Performance of Stellate Sturgeon Industrial Recirculating Aquaculture System, 47, 205–210.
12. Frankic, T.; Voljc, M.; Salobir, J. and Rezar, V. (2009). Use of herbs and spices and their extracts in animal nutrition. Acta agriculturae Slovenica, 94(2): 95-102 p.
13. Sadeghian, M.S; Mohiseni, M.; Nematdust and Haghi, D. (2016). Comparative effect of the oral prescription of Shirazi thyme ( Zataria multiflora Boiss ) and vitamin E on growth indices of juvenile common carp ( Cyprinus carpio L.). Journal of Zoology Research (Iranian Journal of Biology). Vol. 29: 195-204.
14. El-Demerdash, F.M., 2004. Antioxidant effect of vitamin E and selenium on lipid peroxidation, enzyme activities and biochemical parameters in rats exposed to aluminium. Journal of Trace Elements in Medicine and Biology. 18, PP: 113-121.
15. Dicu (Stroe), M.; Cristea, D.; Dediu, V.; Docan, L.; Grecu, I. and Vasilean, I., (2013). Effects of Stocking Density on Growth and Hematological Profile of Early Juveniles Stellate Sturgeon (A. stellatus Pallas, 1771) Reared in a Flow-Through Production System, 46(2).
16. Zaki, M. A.; Labib, E. M.; Nour, A. M.; Tonsy, H. D. and Mahmoud, S. H. (2012). Effect of some medicinal plants diet on mono sex Nile tilapia (Oreochromis niloticus), growth performance, feed utilization and physiological parameters. Asia-Pacific Chemical, Biological & Environmental Engineering Society, 220-227.

17. Dorojan, O. G.; Cristea, V.; Creţu, M.; Coadă, M. T.; Dediu, L.; Grecu, I. R. and Plăcintă, S. (2015). Effect of thyme (Thymus vulgaris) and vitamin E on growth performance and body composition of Acipenser stellatus juveniles. AACL Bioflux 8(2):195-202 p.
18. Ergün, S.; Yılmaz, S. and Yigit, M. (2011). Effects of thyme, rosemary and fenugreek on some hematological and immunological parameters of tilapia, Oreochromis mossambicus. Mediterranean Aquaculture 2020, Aquaculture Europe (EAS 2011); 2011 Oct 19-21; Rhodes-Greece, p. 18-21.
19. Abd El-Gawad Abd El-Halim El-Sayed, S. (2016). Evaluation of Growth Performance, IL-6 and Serum Biochemical Parameters of Rats Fed on Diets Containing Thyme and Ginger Powder. Animal and Veterinary Sciences, 4(5), 78.
20. Antache, Alina, Victor Cristea, Iulia Grecu, Lorena Dediu, Mirela Cretu, Elena Bocioc, and Stefan M Petrea. (2014). "Effects of Dietary Supplementation at Nile Tilapia with Thymus Vulgaris, Trigonela Foenum Graecum and Azadirachta Indica on Welfare Status." Bulletin UASVM Animal Science and Biotechnologies 71 (2): 1843–5262.
21. Ahmadifar, E.; Falahatkar, B. and Akrami, R. (2011). Effects of dietary thymol-carvacrol on growth performance, hematological parameters and tissue composition of juvenile rainbow trout, Oncorhynchus mykiss. Journal of Applied Ichthyology 27 (4): 1057–1060.
22. Yilmaz, E.; Sebahattin, E. and Sevdan, Y. (2015). Influence of Carvacrol on the Growth Performance, Hematological, Non-Specific Immune and Serum Biochemistry Parameters in Rainbow Trout ( Oncorhynchus Mykiss ). Food and Nutrition Sciences 6 (April): 523–31.
23. Aly, S. M.; Abdel-Galil, A.Y.; Abdel-Aziz, G.A. and Mohamed, M. F. (2008). Studies on Bacillus subtilis and Lactobacillus acidophilus, as potential probiotics, on the immune response and resistance of Tilapia nilotica (Oreochromis niloticus) to challenge infections. Fish and Shellfish Immunology, 25, 128–136.
24. Gültepe, N.; Bilen, S.; Yılmaz, S.; Güroy, D. and Seyit Aydın1, S. (2014). Effects of herbs and spice on health status of tilapia (Oreochromis mossambicus) challenged with Streptococcus iniae. Acta Vet. Brno 2014, 83: 125–131.
25. Jameel, Y. J.; Abed, A. R. And Al-Shimmary, F.O. ( 2014). Influence Of Adding Garlic And Thyme And Their Combination On Immune Response And Some Blood, 4 (2): 211–15.
26. Walaa, F.A. E. And Aya G Saad El-Deen. (2016). Immunomodulatory Effects Of Thyme And Fenugreek In Sharptooth Catfish, Clarias Gariepinus. Assiut Veterinary Medical Journal. Vol. 62: 1-7.
27. El-Barabay,M.I. and Mehrim A.I. (2009). Protective effect of antioxidant medicinal herbs, Rosemary and Parsley, on sub acute aflatoxicosis in Nile Tilapia, O. niloticus). Journal of Fisheries and Aquatic Science 4(4):178-190.
28. Yousefisn M., Sheikholeslami A., Kor Davood M., (2011). Serum biochemical parameter of male, immature and female Persian sturgeon (Acipenser persicus). Australian Journal of Basic and Applied Sciences 5(5):476.
29. Farrell A. P., (2011). Encyclopedia of fish physiology. Elsevier, 2272 pp.
30. Patriche, T.; Patriche, N.; Bocioc, E. and Coada, M.T. (2011). Serum biochemical parameter of farmed carp (C. carpio). International J. the Bioflux Society, 4(2): 131-140.
31. Kori-Siakpere O.; Ake J. E. G. and Avworo U. M. (2006). Sublethal effects of cadmium on some selected hematological parameters of Heteroclarias (a hybrid of Heterobranchus bidorsalis and Clarias gariepinus). International Journal of Zoological Research 2:77-83.
32. Andreeva A. M., (1999). Structural and functional organization of the blood albumin system in fish. Vopr Ikhtiol 39:825-832.
33. Wiegertjes, G.F.; Stet, R.J.M.; Parmentier, H.K. and Van Muiswinkel, W.B. (1996). Immunogenetics of disease resistance in fish; a comparable approach. Dev. Comp. Immunol. 20, 365–381.
34. Gulec, Azime Kucukgul, Durali Danabas, Mesut Ural, Engin Seker, Ali Arslan, and Osman Serdar. 2013. "Effect of Mixed Use of Thyme and Fennel Oils on Biochemical Properties and Electrolytes in Rainbow Trout as a Response to Yersinia Ruckeri Infection." Acta Veterinaria Brno 82 (3): 297–302.
35. Sahu, S.; Basanta, K. D.; Jyotirmayee, P.; Mohapatra, B. C.; Mishra, B. K. and Niranjan, S. (2007). "Effect of Magnifera Indica Kernel as a Feed Additive on Immunity and Resistance to Aeromonas Hydrophila in Labeo Rohita Fingerlings" 23:109-118.

2/21/2018