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Effect of thyme powder (*Thymus vulgaris*) on growth, feed utilization and hematological characteristics of common carp fingerling (*Cyprinus carpio).*

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ABSTRACT

The present study aimed to evaluate the efficiency of thyme powder (Thymus vulgaris) in growth performance, biochemical parameters, biological traits, and haematology of common carp fingerlings. For this purpose, (108) fingerlings of *cyprinus carpio* were randomly distributed into four treatment groups and thier weights ranged between 75.18 -75.36 g. Fish were fed with different thyme levels: 1% (T2), 1.5% (T3) and 2% (T4).The first group (T1) was a control group without any addition of thyme. The experiment was conducted at the animal science department/ college of agricultural engineering sciences/the University of Sulaimani. The experiment was continued for 8 weeks. At the end of the experiment, blood samples were collected from the caudal vein for determination of haematological and biochemical parameters. Results of the feeding trial in treated groups with thyme supplementation showed a significant increase ($P<0.05$) in the weight gain, with the highest value in treatment (T3). Also, the food conversion ratio was improved, and there was a significant difference between (T3,T4) compared to (T1, T2).For fish health indicator such liver enzymes there were no significant differeneces among treatments ($P<0.05$). Hematological parameters were analuzed and significant difference was observed for (mch) among control treatment compare to with other treatments, for rest of other hematological parametres no significant differences were noted.There were no significant differences for hepatosomatic index, meat weight index and fish weight index. For other biological traits, significant differences were seen among treatments. Highest condition factor value was observed in trreatment (2) and significant difference was seen among treatment 3 and 4. On the basis of our data, the dietary supplementation of *T. vulgaris* improved the growth rate, hematological, biochemical parameters of common carp fingerlings.

***Keywords*:** Common carp, growth, hematology, Thymus vulgaris, biochemica.

INTRODUCTION

Common carp yearly output is predicted to reach 4.2 million tons, accounting for 7.7% of aquaculture [1]. Intensive carp production is connected with a variety of risks and infectious illnesses, putting the development of sustainable carp farming at risk [2]. Unfortunately, fish producers' first line of defense against infectious pathogens is the use of antibiotics and chemical disinfectants. According to recent data, worldwide antibiotic usage in aquaculture was projected to be 10,259 tons in 2017 and is expected to climb to 13,600 tons by 2030 [3]. Plants and their derivatives have been shown to promote growth performance and boost immunological responses, resulting in antibacterial and anti-stress properties [4, 5]. Herbal medicines have been widely researched and reported to have valuable potential in the therapeutic management of fish infections during the last several decades [6, 7, 8]. Thyme (Thymus vulgaris), a fragrant plant in the Lamiaceae family, includes thymol, carvacrol, cymene, eugenol, and 4-allylphenol, and has been shown to improve growth promotion, antibacterial, immunostimulatory, and antioxidant properties [9, 10]. This plant has been shown to improve growth and immunological parameters in gibel carp (*Carassius auratus gibelio*), common carp (*Cyprinus carpio*), and rainbow trout (*Oncorhynchus mykiss*) [11, 12]. As a result, the purpose of this study was to investigate the effects of various doses of thyme powder on the growth performance and hematological features of common carp (*c.carpio*).

Materıals and Methods

Experimental fish:

The study was conducted in 60 days on 108 common carp. *C. carpio* L. is fed by local fish from the ponds of Caladze, Sillaimani, and Iraq. The fish were placed in some experimental plastic aquariums. Before the real feeding experiment, a 21-day pre-adherence laboratory and a commercial pellet feeding (as shown in Table 1) were performed.

Table 1: Chemical composition of the various diet by [13].

Ingredients	Crude Protein %	Crude Fat %	Dry Matter %	Crude Fiber %	Energy Kcal/ kg	Ingredients %
Animal protein Concentrate	40	5	92.9	2.2	2107	20 %
Yellow corn	8.9	3.6	89	2.2	3400	15 %
Soybean meal	48	1.1	89	7	2230	35 %
Barely	11	1.9	89	5.5	2640	15 %
Wheat bran	15.7	4	89	11	1300	15 %

Experimental system:

This research used twelve plastic tanks (70 litres of water) to treat four patients, each with three duplicates. With the help of the Chinese air compressor Hailea ACO-318, each tank was supplied with adequate continuous aeration. Each replica received eight fish. In order to reduce the difference between treatments, duplicates are distributed randomly. In order to eliminate any remaining feed and waste from the system, a regular siphoning procedure is used. The experiments consisted of four treatments, three of which were duplicated and eight of which were fish.

T1: Feeding meals to fish without thyme powder, T2: Feeding fish meals with 1% thyme powder, T3: Feeding fish meals containing 1.5% thyme powder, T4: Feeding fish diets containing 2% thyme powder.

Diet formulation:

This study used 12 plastic tanks (70l of water) in four treatments, and each treatment used three identical tanks. With the help of Chinese air compressors, the Hailea ACO-318, each tank was fitted with proper continuous ventilation. Each replication received eight fish. To reduce the differences between treatments, duplicates will be distributed randomly. A siphoning process is regularly used to remove the remaining feed and excrement from the system. The experiment consisted of four treatments with three replications and eight fish:

Health and Biological parameters

All fish samples were dissected, the abdomen cavity opened to separate each organ's weight, and the results were calculated as follows.

Hepatosomatic index % = Liver weight (gm) / Fish weight (gm) x 100 [14]
Spleenosomatic index% = Spleen weight (gm) / Fish weight (gm) x 100
Gonadosomatic index% = gonad weight (gm) / fish weight (gm) x 100
Intestine weight index% = Intestine weight (gm) / Fish weight (gm) x 100
Intestine length index% = Intestine length (gm) / Fish length (gm) x 100
Condition factor = Fish weight (gm) / Fish length (cm)3 [15]
Gill index% = Gill weight (gm) / Fish weight (gm) x 100.
Fish weight index% = fish weight (gm) / fish weight (gm) x 100
Meat weight index% = fish weight (gm) minus viscera and head (gm) x 100

Growth standards used in the study:

Weight increase (g/fish) = mean of weight (g) after the experiment minus weight (g) at the start of the trial.
W2 - W1 = weight growth (g/fish)
Where:
W2: Fish weight (g) at the end of the experiment
W1: Fish weight (g) at the start of the trial.
Relative Growth Rate (RGR%) = Weight Gain / Initial Weight x 100
= W2 - W1 / W1 x 100

At the end of the experiment, three fish from each experiment group were selected randomly. All fish samples are measured and weighed separately. Blood samples were obtained from the caudal veins of each fish in each group. All blood samples were collected and stored in a small plastic bottle containing heparin [16].

The following parameters were measured: the number of erythrocytes (rbcs: 1012 cells/l), the average number of hemoglobins in the body (mch; pg), the average number of hemoglobins in the body (mchc; g/dl), the average number of hemoglobins (mcv; fl), the hemoglobin (hb; g/dl) and the platelets (plt; 109 cells/l), the difference.

Biochemical parameters

Alanine aminotransferase (alt), aspartate aminotransferase (ast), total proteins, globulin (g/dl), and albumin (g/dl) were measured using an Radioimmuno test (RIA) technology (Cobas e 411) supplied from Roche and produced by Hitachi High-Technologies Corporation.

The instrument uses the following procedures to measure the serum parameters: [17] for ALT and AST activities.

Statistical analysis

The study was carried out using the XLSTAT 2019 Version.02.28451 one-way ANOVA with fully randomized design (CRD) and general linear models (GLM) technique. Duncan's test was used to compare the means of different treatments [18] .

Results

There were significant differences ($p < 0.05$) in weight gain among treatments; the highest values of weight gain were observed in treatment 3. The relative growth rate, specific growth rate, food conversion ratio and food efficiency ratio of the *c. Carpio* were significantly affected by feeding fish with thyme powder (thymus vulgaris).

Table 2: Effect of thyme powder (thymus vulgaris) on growth and feed utilization parameters of young common carp (c. Carpio).

Parameters	t1 control	t2 1 % *thyme powder*	t3 1.5 % *thyme powder*	t4 2 % *thyme powder*
Weight gain	4.000±1.000 C	11.535±0.425 BC	17.640±0.405 A	15.520±3.510 AB
Relative growth rate	3.125±0.56 B	6.648±0.653 AB	11.574±1.840 AB	14.525±1.334 A
Specific growth rate	8.719±0.03 AB	9.338±0.119 A	9.145±0.101 AB	8.551±0.147 B
Food conversion ratio	1.442±0.258 A	1.384±0.163 A	0.656±0.034 B	0.685±0.084 B
Food efficiency ratio	0.488±0.087 B	0.732±0.086 B	1.565±00.115 A	0.068±0.016 C

Different letter in same rows mean significant differences ($p<0.05$).

Mean values for (rbc, hgb, mcv, mch, mchc, mcv, plt, wbc, granules, lymphocyte and monocyte) were presented in table 3 as mean± se. According to the results, there were significant differences ($p <0.05$), for (mch and lymphocyte). However, there were no significant differences for (rbc, mchc, plt, mcv, granules, hgb and monocyte).

Ttable 3: Effect of thyme powder (thymus vulgaris) on some haematological indices of young common carp (c. Carpio).

Parameters	t1 control	t2 1 % *thyme powder*	t3 1.5 % *thyme powder*	t4 2 % *thyme powder*
Rbcs (10^{12} cells/l)	1.330±0.13 A	3.935±1.109 A	2.410±0.560 A	1.380±0.106 A
Hb (g/dl)	10.900±2.01 A	10.350±0.886 A	8.650±0.460 A	9.250±0.106 A
Mch (pg)	320.000±4.64 A	33.000±1.418 B	32.000±1.418 B	30.000±1.418 B

Mchc (g/dl)	24.000±19.39 A	26.250±0.177 A	26.500±0.496 A	26.800±1.205 A
Mcv (fl)	100.000±8.15 A	95.500±7.78 A	92.000±2.87 A	92.000±6.28 A
Plt (10^9cells/l)	8.000±7.12 A	13.500±3.191 A	22.050±1.382 A	15.000±2.127 A
Wbc (10^9cells/ l)	9.000±0.000 A	9.000±0.000 A	8.500±0.212 A	9.000±0.000 A
Granulocytes (%)	1.100±2.13 A	0.850±0.460 A	0.850±0.248 A	1.600±0.141 A
Lymphocytes (%)	8.500±0.83 B	85.600±2.831 A	85.350±1.170 A	82.200±4.397 A
Monocytes (%)	0.000±0 A	0.200±0.070 A	0.450±0.177 A	0.250±0.035 A

Different letter in same rows mean significant differences ($p<0.05$).
Values of alt, ast, total protein, globulin and albumin in fish plasma were no significant($p <0.05$) amon treatments.

Table 4: Effect of thyme powder (thymus vulgaris) on some blood biochemical parameters of young common carp (c. Carpio)

Parameters	t1 control	t2 1 % *thyme powder*	t3 1.5 % *thyme powder*	t4 2 % *thyme powder*
Alanine aminotransferase activity (alt)	130.450±3.63 A	71.750±44.539 A	128.165±25.613 A	64.100±39.482 A
Aspartate aminotransferase activity (ast)	586.99± 2.26 A	419.135±241.414 A	490.755±28.223 A	387.465±213.776 A
Total proteins	45.560±2.51 A	29.225±1.904 A	30.945±1.131 A	39.870±8.815 A
Globulin (g/dl)	44.500±0.24 A	29.000±2.056 A	30.500±1.276 A	38.450±8.049 A
Albumin (g/dl)	0.920±0.14 A	0.225±0.159 A	0.365±0.124 A	1.395±0.782 A

Different letter in same rows mean significant differences ($p<0.05$).
Significant differences ($p<0.05$) were noted in among treatments in all parameters that listed in table 6 except hepatosomatic index, fish weight index and meat weight index. Highets condition factor was in treatment 3. Also, highest values of meat weight index were observed in treatment

Table 5: Effect of thyme powder (thymus vulgaris) on some physio-biological parameters of young common carp (c. Carpio).

Parameters	t1 control	t2 1 % *thyme powder*	t3 1.5 % *thyme powder*	t4 2 % *thyme powder*
Hepatosomatic index	0.142±0.08 A	0.524±0.0201 A	2.116± 0.0201 A	0.566 ±0.56 A
Spleenosomatic index	1.420±0.08 Ab	0.924±0.011 C	1.562± 0.039 A	1.231 ±0.066 B
Gillsomatic index	6.534±0.04 BC	6.934±0.210 B	5.344±0.190 C	8.316±0.315 A
ntestine weight index	5.398±0.21 A	1.429±0.256 B	5.082±0.342 A	3.864±0.205 A
Fish weight index	83.475±0.7003 A	86.190±0.229 A	85.675±0.501 A	82.938±0.083 A
Meat weight index	61.080±1.07 A	60.414±0.424 A	62.003± 1.291 A	57.548±1.980 A
Condition factor	1.760± 0.02 AB	2.013±0.002 AB	2.154±0.056 A	1.709±0.128 B

Different letter in same rows mean significant differences ($p<0.05$).

Dissussion

Thyme and its derivatives have been used as supplements in fish diets primarily to enhance growth and immunity [19, 20, 21].

The current study showed significant increase specific growth rate (SGR) and feed convertion ratio along the whole periods in *C. carpio* fed on thyme where 8.551±0.147% and 0.685±0.084 respectively, at the level 2% of adding thyme powder . Also, [10] documented that Oreochromis niloticus fed diets supplemented with thyme (2%) levels positively affected growth performance and found that specific growth rate and feed convertion ratio were 3.65% and 2.59 respectivley. On the other hand, [22] showed no enhancement in stellatus juvenile growth performance after feeding 1% of dietary thyme. Still, thyme supplementation has improved of meat biochemical quality composition, due to a significant decrease in the percentage of water, respectively increasing the percentage of protein. Our results agree with [23], who noticed that in Nile tilapia fed diet supplemented with thyme and rosemary an increase in growth characteristics were noted. Blood is a patho-physiological indicator of the whole body and the counts of haematological indices in blood offer a reflection to the health status of fish by detecting any disruption occurring due to the use of immunostimulants [24].

The results of the present study indicated that supplemented thyme in fish diet increased considerably the RBCs count (3.935 ± 1.10910^{12} cells/l) and lymphocyte% (85.600±2.831). These results agree with [25] who recorded that the RBCs count, HB and PCV% in Nile tilapia fed diet supplemented with 1% of thyme were significantly increase compared to control. Similarly, [26] reported that the RBC in rainbow trout, Oncorhynchus mykiss fed thymol- carvacrol supplemented diet were slightly higher than the control. Similar results were reported by [27] who found that the RBC count, Hb concentration and PCV in rainbow trout fed dietary carvacrol were significantly higher in comparison to control group. Also, our results were in line with the results of [28] who found that the RBC value in fish fed 1% of thyme supplementation for 45 days significantly increased compared with control. Also, lymphocyte percentage significantly differed significantly in groups fed thyme supplemented diets compared to control.

The bio-somatic indices are considered as environmental stress indicators of fish [29]. The results in the current study revealed significant increase of (Spleen-somatic index, intestine- somatic index, gill-somatic index and condition factor) (1.231 ±0.066, 3.864±0.205, 8.316±0.315 and1.709±0.128) in fish received thyme-supplemented diet for 60 days at the level 2%. [23] Stated that significant increase of intestine- somatic index in fish received thyme-supplemented diet for 6 weeks and significant increase of HSI in group fed rosemary treated diet for 2 weeks.In the same respect, significant increase of ISI in O. niloticus fed on 2% ginger [30] and HBP [31] were recorded. Moreover, a little positive effect on bio-somatic indices of African catfish fed with rosemary extract was observed by [32]. The increase of intestine- somatic index may be due to an increase in thickness of intestinal tract villi [33]. In contrast, the increase of HSI could be attributed to increase in size and number of liver cell in fish that exposed to stress [34].

Conclusion

According to our results it was shown that dietary supplementation of *T. vulgaris* improved the growth rate of carp fingerlings. Also, *T. vulgaris* had a significant effect on physiological parameters such as RBC and lymphocyte. Also, it affected biological and nutritional parameters such as weight gain, relative growth rate, specific growth rate, feed conversion ratio and feed effieciency of common carp. While this study suggests future studies on thyme powder as feed to common carp in other aspects such as meat quality atttibuteies.

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تأثير مسحوق الزعتر (*Thymus vulgaris*) على النمو واستهلاك الأعلاف والصفات الدموية للكارب الاعتيادي (*C.carpio*).

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قسم علوم الحيوان، كلية هندسة العلوم الزراعية، جامعة السليمانية، السليمانية ، العراق.

الخلاصة
هدفت هذه الدراسة إلى تقييم تأثير كفاءة الزعتر (***Thymus vulgaris***) على صفات النمو ، والمعايير الكيموحيوية والصفات البيولوجية وصفات الدم للكارب الاعتيادي. لهذا الغرض تم في هذه الدراسة تربية 108 إصبعيات من اسماك الكارب تراوحت اوزانها بين 75.18 -75.36 غم تم توزيعها عشوائيا الى أربع مجموعات. تم تغذية الأسماك بتراكيز مختلفة من الزعتر: 1 ٪ (المعاملة 2) ، 1.5 ٪ (المعاملة 3) و 2 ٪ (المعاملة 4).المجموعة الأولى كانت بمثابة مجموعة سيطرة دون أي إضافة للزعتر. أجريت التجربة في قسم علوم الحيوان / كلية الهندسة الزراعية / جامعة السليمانية. استمرت التجربة لمدة 8 أسابيع ، بعد 60 يوما من التغذية تم جمع عينات الدم لدراسة صفات الدم الخاصة بالدراسة. أظهرت نتائج مسار التغذية في المجموعات المعاملة بمكملات الزعتر زيادة كبيرة وعلى مستوى المعنوية (***P<0.05***) في صفة زيادة الوزن وذلك في المعاملة الثالثة (3) ، أيضا تحسنت نسبة كفاءة التحويل الغذائية وكان هناك فرق كبير بين (المعاملة 3 و 4) مقارنة بـ (المعاملة 1 و 2).بالنسبة لمؤشر صحة الأسماك حيث لم يكن هناك اختلاف كبير في إنزيمات الكبد بين المعاملات وعلى مستوى معنوية (***P<0.05***) بالنسبة لصفات الدم حيث تم ملاحظة اختلاف كبير في قيمة ال (***Mch***) بين معاملة السيطرة مقارنة بالمعاملات الأخرى ، وبالنسبة لبقية صفات الدم الأخرى ، لم يلاحظ أي فروق معنوية .لم تكن هناك فروق معنوية لمؤشر الكبد، مؤشراللحوم الأسماك سمات البيولوجية الأخرى شوهدت اختلافات كبيرة بين المعاملات. ولوحظت أعلى قيمة لعامل الحالة في المعاملة (2) ولوحظ اختلاف كبير بين المعاملة 3 و 4.على أساس بياناتنا ، اضافة مسحوق الزعتر يؤدى الى تحسين معدل النمو و صفات الدم الكيموحيوية لأسماك الكارب الاعتيادي.

الكلمات المفتاحية: الكارب الاعتيادي ، النمو ، صفات الدم ، الزعتر ، البيوكيميائية.