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Nutritional Impact of Incorporation Thyme (*Thymus vulgaris*) Meal at Different Levels on Growth Performance, Biochemical Parameters and Economical Evaluation of the Nile Tilapia (*Oreochromis niloticus*)

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ABSTRACT

This study evaluated thyme meal (TM) as a plant protein source in fish feed, replacing 0, 10, 20, and 30% of soybean meal, which made up 40% of the control diet. The aim was to determine TM effect on the Nile tilapia growth performance. A total of 120 mon-sex fingerlings Nile tilapia, averaging 14.3± 0.64g, were acclimated and distributed into 12 aquariums (80×40×30cm, 60 liters), with 10 fish per aquarium, totaling 30 fish per each treatment group. TM contained 20% crude protein. Diets were similar in nitrogen content but varied in energy. The results showed significant improvements in total body weight gain (TBWG), average daily gain (ADG), and specific growth rate (SGR) with higher TM levels. Moreover, survival rates were 100% in all groups. The feed conversion ratio (FCR) improved significantly (P < 0.05) with higher TM inclusion. Fish body composition showed higher moisture, CP, and ash content, while dry matter (DM), organic matter (OM), EE, and gross energy decreased in TM groups. Furthermore, energy retention and protein production value (PPV) increased significantly (P < 0.05). Blood protein levels rose in all TM groups, with the highest values of protein and globulin in 10% group and non-significant effects on AST and uric acid compared to control. Feed costs decreased, enhancing cost-efficiency. In conclusion, thyme meal is an effective alternative plant protein source for tilapia diets, improving growth performance, feed efficiency, and biochemical parameters.

INTRODUCTION

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A shortage of animal proteins in numerous developing countries arises from the mismatch between the growth of the population and the increasing requirements of animal protein production (**Workagegn** *et al.*, **2014**). This imbalance between population growth and fish production, along with the rising demands for high-quality ingredients,

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has heightened the need for fish. However, as reported by the FAO (2006, 2009), wild fish catches have reached their limits, and the high cost of fish could lead to overfishing, jeopardizing food security for those reliant on fish as a primary food source. To address this, sustainable aquaculture production is essential to prevent fish protein from becoming scarce and expensive (FAO, 2012). Fish feeding constitutes the largest expense in semiintensive and intensive aquaculture systems, largely due to the cost of animal proteins such as fish meal (Hardy, 2010). Global production of fish meal and fish oil has fluctuated, averaging 6.5 million metric tons and 1.3 million metric tons, respectively, over the past two decades. Research has explored plant proteins as substitutes for animal proteins in fish feed to reduce costs. For example, in Egypt, a study successfully replaced fish meal with a blend of plant proteins in the Nile tilapia diets, showing no adverse effects on growth performance (El-Saidy & Gaber, 2003). Efforts to enhance aquaculture involve improving technologies, farming systems, and managing production costs (El-Sayed, 2021; Fayed et al., 2023). Herbal additives as thyme (Thymus vulgaris) are noted for their growth-promoting, antimicrobial, and antioxidant properties (Rota et al., 2008; Zak et al., 2012). Thyme has been shown to positively affect growth and immunity in various fish species (AlSafah & Al-Faragi, 2017; Zadmajid & **Mohammadi**, 2017). Given the increasing global population and food security concerns, thyme's potential in aquaculture is significant (Subasinghe et al., 2009; Silva et al., **2021**). This study aimed to evaluate the effects of replacing soybean meal with thyme meal at levels of 0, 10, 20, and 30% of soybean meal in a diet, where soybean meal constitutes 40% of the control diet, on the growth performance, feed utilization, body composition, blood parameters, and economic viability of the Nile tilapia.

MATERIALS AND METHODS

This study was conducted in the Fish Laboratory of the Animal Production Department, Biological Agriculture Research Institute, National Research Center, in collaboration with the Hydrobiology Department of the Veterinary Research Institute and Biotechnology Research Institute, National Research Center. The objective was to assess the effects of thyme meal inclusion at various levels (0, 10, 15, and 20%), replacing soybean meal on the Nile tilapia's growth performance, feed efficiency, body composition, blood parameters, and economic aspects.

Experimental setup

A total of one hundred twenty Nile tilapia, averaging 14.3 ± 0.64 g, were acclimated and randomly allocated to 12 aquariums ($80\times40\times30$ cm, 60 liters), with 10 fish per aquarium, totaling 30 fish per treatment group.

Dietary treatments

Thyme meal was used as a substitute for soybean meal, incorporated at 0, 10, 20, and 30% of the soybean meal content, which was 40% in the control diet. This resulted in thyme meal levels of 0g/ kg (D1), 40g/ kg (D2), 80g/ kg (D3), and 120g/ kg (D4) of the diet formulation (Table 1). The experimental diets were fed for 56 days, from mid-January to mid-March 2024.

| | Control | Replacing | Replacing | Replacing | Price |
|------------------------------|----------------|----------------|-----------------------|---------------|---------|
| Ingradiants | Zero | 10% of | 20% of | 30% of | of tone |
| lingieulents | TM | SBM | SBM | SBM | LE |
| | | by TM | by TM | by TM | |
| | \mathbf{D}_1 | \mathbf{D}_2 | D ₃ | D 4 | |
| Composition of tested diets | | | | | |
| Thyme meal(TM), (20% CP) | 0.00 | 4.00 | 8.00 | 12.00 | 15000 |
| Soybean meal (SBM), (44% CP) | 40.00 | 36.00 | 3200 | 28.00 | 33000 |
| Concentration (56% CP) | 17.00 | 19.00 | 22.00 | 24.00 | 25000 |
| Ground yellow corn (8% CP) | 28.00 | 28.00 | 28.00 | 28.00 | 12500 |
| Wheat bran (13% CP) | 10.00 | 8.00 | 5.00 | 3.00 | 14500 |
| Vegetable oil | 3.00 | 3.00 | 3.00 | 3.00 | 50000 |
| Salt (sodium chloride) | 1.00 | 1.00 | 1.00 | 1.00 | 5000 |
| Vitamins and minerals | 2.00 | 2.00 | 2.00 | 2.00 | 40000 |
| mixture** | | | | | |
| Price of ton fed (LE) | 24350 | 23840 | 23435 | 22925 | |
| Price of kg fed (LE) | 24.350 | 23.840 | 23.435 | 22.925 | |
| SBM: Soybean meal. | | | | | |

| Table 1. | Composition | of the different | experimental | diets |
|----------|-------------|------------------|--------------|-------|
| | | | | |

TS: Thyme meal.

** Vit. A (E672) (IU) 876.19, Vit. D3 (IU) 1141.39, Vit. E 114.30, Vit. K3 7.55, Vit. B1 13.71, Vit. B2 11.44, Vit. B6 15.33, Vit. B12 0.03, Niacin 60.96, Calpan 30.48, Folic Acid 3.04, Biotin 0.37, Vit. C 11.44, Selenium 0.27, Manganese 19.04, Iron 9.15, Iodine 0.77, Zinc 76.19, Copper 3.04, Cobalt 0.37, Choline Chloride 457.14, and Antioxidant 95.23 (Vit. vitamin; IU international unit).

Performance metrics

- Body Weight Gain (BWG): Final weight Initial weight
- Survival Rate (SR %): (Number of fish at end / Number of fish at start) × 100
- Specific Growth Rate (SGR): [ln(final weight) ln(initial weight)] / Experimental days × 100
- Feed Conversion Ratio (FCR): Total dry matter intake (g) / Total body weight gain (g)
- **Crude Protein Efficiency Ratio (CPER):** Total body weight gain (g) / Total crude protein intake (g)
- Feed Efficiency (FE %): Weight gain (g) / Feed intake (g)
- **Protein Productive Value (PPV %):** [(PR1 PR0) / PI] × 100, where PR1 is final body protein, PR0 is initial body protein, and PI is protein intake.

• Energy Retention (ER %): [(E - E0) / EF] × 100, where E is final carcass energy; E0 is initial carcass energy, and EF is the energy from feed intake.

Blood sampling

Blood was drawn from the caudal vein using a 3ml syringe after anesthetizing the fish with clove oil (0.5ml/ L). Samples were collected in centrifuge tubes, allowed to clot at room temperature, centrifuged at 3000rpm/ 15min, and the serum was stored at -20°C until analysis.

Body composition

Initially, 15 fish per group were used, and at the study's end, 6 fish per treatment were randomly selected for body composition analysis.

Analytical procedures

Dietary and body composition analyses followed AOAC (2016) standards. Biochemical assays included alanine aminotransferase (ALT) and aspartate aminotransferase (AST) (Reitman & Frankel, 1957), total proteins (Cannon *et al.*, 1974), globulin (total protein minus albumin), cholesterol (Ellefson & Caraway, 1976), glucose (Caraway & Watts, 1987), albumin, uric acid, and creatinine (Tietz, 1990). These assays were conducted using commercial kits (Spectrum-diagnostics, Egypt) and analyzed with an Agilent Cary UV-Vis spectrophotometer.

Energy calculations

Gross energy (kcal/kg DM) and metabolizable energy (ME) were calculated using established values (**Blaxter, 1968; MacRae & Lobley, 2003; NRC, 2011**). The protein energy ratio was also determined according to the method of **NRC (2011)**.

Statistical analysis

Data were analyzed using Duncan's multiple range test (**Duncan**, 1955) and oneway ANOVA with **SPSS** (2020) for mean separation.

RESULTS

Chemical analysis of experimental diets

The chemical composition of thyme meal (TM) is detailed in Table (2). TM contained 20% crude protein (CP), 7.42% ether extract (EE), 22.30% crude fiber (CF), 42.13% nitrogen-free extract (NFE), with a gross energy content of 4501kcal/ kg DM, and a metabolizable energy content of 297.51kcal/ kg DM. The protein energy ratio was 67.22mg CP/kcal ME.

For the experimental diets, CP levels ranged between 27.50 and 27.89%. Gross energy values varied from 4303 to 4400kcal/ kg, while metabolizable energy ranged from 337.13 to 346.15kcal/ kg. The protein energy ratio across the diets was between 79.44 and 82.73mg CP/kcal ME.

These values are generally adequate to meet the nutritional needs of the Nile tilapia. Although the diets were nearly iso-nitrogenous, there were differences in growth energy content, likely due to the higher EE and GE in thyme meal compared to soybean meal. The data in Table (3) reveal that the final weight (FW), total body weight gain (TBWG), average daily gain (ADG), and specific growth rate (SGR) of fish significantly (P<0.05) improved when their diets included 10, 20, and 30% of soybean meal (SBM) replaced with thyme meal. Additionally, all experimental groups achieved a 100% survival rate (SR) with no recorded mortality. The incorporation of thyme meal in the diet notably (P<0.05) influenced the aforementioned growth parameters.

Regarding feed utilization, as shown in Table (4), the feed conversion ratio (FCR) improved significantly (P<0.05) with increasing the levels of thyme meal in the diets. The group fed diet D2 demonstrated the highest feed intake (526.25g), surpassing the control and other groups which recorded 496.40, 459.40, and 460.70g for D1, D3, and D4, respectively. Moreover, fish in the D2 group exhibited the highest crude protein intake (145.61g), compared to 136.51, 127.21, and 128.49g for D1, D3, and D4, respectively.

| | Experimental diets | | | | | | | | |
|-------------------------------|--------------------|----------------|----------------|-----------------|-----------------------|--|--|--|--|
| | | Control | Replacing | Replacing | Replacing | | | | |
| T 4 | Thymus | Zero | 10% of | 20% of | 30% of | | | | |
| Item | meal | TM | SBM | SBM | SBM | | | | |
| | (TM) | | by TM | by TM | by TM | | | | |
| | | \mathbf{D}_1 | \mathbf{D}_2 | \mathbf{D}_3 | D ₄ | | | | |
| Moisture | 7.82 | 7.90 | 7.17 | 6.45 | 7.18 | | | | |
| Dry matter (DM) | 92.18 | 92.10 | 92.83 | 93.55 | 92.82 | | | | |
| | | | Chemic | cal analysis of | n DM basis | | | | |
| Organic matter (OM) | 91.85 | 90.02 | 88.44 | 88.36 | 88.30 | | | | |
| Crude protein (CP) | 20.00 | 27.50 | 27.67 | 27.69 | 27.89 | | | | |
| Crude fiber (CF) | 22.30 | 5.20 | 5.46 | 5.70 | 5.82 | | | | |
| Ether extract (EE) | 7.42 | 4.80 | 4.14 | 4.49 | 4.53 | | | | |
| Nitrogen free extract (NFE) | 42.13 | 52.52 | 51.17 | 50.48 | 50.06 | | | | |
| Ash | 8.15 | 9.98 | 11.56 | 11.64 | 11.70 | | | | |
| Gross energy kcal/ kg DM | 4501 | 4400 | 4303 | 4318 | 4321 | | | | |
| Gross energy cal/ g DM | 4.501 | 4.400 | 4.303 | 4.318 | 4.321 | | | | |
| Metabolizable energy kcal/ kg | 297.51 | 346.15 | 336.84 | 337.37 | 337.13 | | | | |
| DM | | | | | | | | | |
| Protein energy ratio (mg CP/ | 67.22 | 79.44 | 82.15 | 82.08 | 82.73 | | | | |
| Kcal ME) | | | | | | | | | |

Table 2. Chemical analysis of thyme meal and the different experimental diets

SBM: Soybean meal.

TM: Thyme meal.

Gross energy (kcal/ kg DM) was calculated according to (Blaxter1968; MacRae and Lobley 2003). Where, each g CP = 5.65 Kcal, g EE = 9.40 kcal and g CF and NFE = 4.15 Kcal.

Metabolizable energy (ME): calculated using values of 4.50, 8.15 and 3.49 Kcal for protein, fat andcarbohydrate, respectively and calculated according to **NRC** (2011). Protein energy ratio (mg CP/ Kcal ME): Calculated according to **NRC** (2011).

| | Control | Replacing | Replacing | Replacing | _ | |
|----------------------------|-------------------|-------------------|-------------------|-------------------|-------|----------------|
| Item | zero | 5% of | 10% of | 15% of | | |
| Item | TM | SBM | SBM | SBM | SEM | Sign. |
| | | by TM | by TM | by TM | - | <i>P</i> <0.05 |
| | \mathbf{D}_1 | \mathbf{D}_2 | \mathbf{D}_3 | \mathbf{D}_4 | | |
| Number of fish | 30 | 30 | 30 | 30 | - | - |
| Initial weight, g (IW)/10 | 142 | 145 | 144 | 141 | 0.640 | NS |
| fish | | | | | | |
| Final weight, g (FW)/10 | 308 ^c | 338 ^b | 360 ^a | 368 ^a | 7.116 | * |
| fish | | | | | | |
| Total body weight gain, g | 166 ^d | 193° | 216 ^b | 227 ^a | 7.121 | * |
| (TBWG) | | | | | | |
| Duration experimental | | | 56 days | 6 | | |
| period | | | | | | |
| Average daily gain, g | 2.96 ^d | 3.45 ^c | 3.86 ^b | 4.05 ^a | 0.127 | * |
| (ADG) | | | | | | |
| Specific growth rate (SGR) | 1.18° | 1.34 ^b | 1.42^{a} | 1.45 ^a | 0.035 | * |
| Number of fish at the | 30 | 30 | 30 | 30 | - | - |
| starter | | | | | | |
| Number of fish at the end | 30 | 30 | 30 | 30 | - | - |
| Survival ratio (SR) | 100 | 100 | 100 | 100 | - | - |
| Number of dead fish | Zero | Zero | Zero | Zero | - | - |
| Mortality rate percentages | Zero | Zero | Zero | Zero | - | - |

Table 3. Growth performance, specific growth rate and survival ratio of different experimental groups

SBM: Soybean meal. TM: Thyme meal.

a, b, c and d:Means in the same row having different superscripts differ significantly (P<0.05).

SEM: Standard error of mean

NS: Not significant

*: Significant at *P*<0.05.

| Item | Control zero TM | Replacing 10% of SBM by TM | Replacing 20% of SBM by TM | Replacing 30% of SBM by TM | SEM | Sign. <i>P</i> <0.05 |
|---------------------------|-----------------------|-------------------------------------|-------------------------------------|-------------------------------------|-------|-------------------------|
| | \mathbf{D}_1 | \mathbf{D}_2 | D_3 | \mathbf{D}_4 | - | |
| Total body weight gain, g | 166 ^d | 193 ^c | 216 ^b | 227 ^a | 7.121 | * |
| (TBWG) | | | | | | |
| Feed intake (FI), g | 496.40^{a} | 526.25 ^a | 459.40 ^c | 460.70° | 8.500 | * |
| Feed conversion ratio | 2.99 ^d | 2.73 ^c | 2.13 ^b | 2.03 ^a | 0.122 | * |
| (FCR) | | | | | | |
| Feed crude protein % | 27.50 | 27.67 | 27.69 | 27.89 | - | - |
| Crude protein intake | 136.51 ^b | 145.61 ^a | 127.21 ^c | 128.49 ^c | 2.260 | * |
| (CPI), g | | | | | | |
| Protein efficiency ratio | 1.216 ^d | 1.325 ^c | 1.698 ^b | 1.767 ^a | 0.071 | * |
| (PER) | | | | | | |

Table 4. Feed utilization of the different experimental groups

SBM: Soybean meal.TM: Thyme meal.,a, b, c and d: Means in the same row having different superscripts differ significantly (P<0.05),SEM: Standard error of mean, *: Significant at (P<0.05), FCR: Expressed as g of DM intake / g gain, PER: Expressed as g of g gain / g CP intake.

Blood parameters of the different experimental groups

The blood parameter results shown in Table (5) indicate that incorporating thyme meal (TM) into fish diets significantly (P<0.05) influenced several biochemical markers. There was a notable (P<0.05) increase in total protein levels across all groups treated with thyme-substituted meal (TSM) compared to the control, with the highest protein concentration observed in the group receiving 10% TSM. Albumin levels significantly (P<0.05) rose in the 20 and 30% TSM groups, with the highest albumin concentration found in the 30% TSM group. Globulin levels showed the highest significant increase (P<0.05) in the 10% TSM group, while a significant decrease (P<0.05) was observed in the 30% TSM group.

Regarding liver and kidney functions, the concentrations of aspartate aminotransferase (AST) and uric acid did not show significant changes (P>0.05) in the TSM-treated groups compared to the control. However, alanine aminotransferase (ALT) reached its highest significant value (P<0.05) in the 30% TSM group, and the creatinine level was significantly (P<0.05) elevated in the 20% TSM group. In terms of glucose and cholesterol concentrations, glucose levels significantly increased in the 10% and 30% TSM groups, while cholesterol levels significantly decreased in the 20% and 30% TSM groups compared to the control.

| | | Experime | ental diets | | | | | |
|-----------------------|-------------------------------|-------------------------------------|-------------------------------------|-------------------------------------|-------|-------------------------|--|--|
| Item | Control zero TM | Replacing 10% of SBM by TM | Replacing 20% of SBM by TM | Replacing 30% of SBM by TM | SEM | Sign. <i>P</i> <0.05 | | |
| T-4-1 (-/ | $\frac{D_1}{2.05^{\text{b}}}$ | D_2 | $\frac{D_3}{2 co^3}$ | D_4 | 0.10 | * | | |
| l otal protein (g/dl) | 2.95° | 3.74" | 3.69 | 3.70° | 0.10 | * | | |
| Albumin (g/dl) | 1.30° | 1.19 ^c | 1.75° | 3.13 ^a | 0.07 | * | | |
| Globulin (g/dl) | 1.65 ^b | 2.55 ^a | 1.94^{ab} | 0.57 ^c | 0.14 | * | | |
| Glucose (mg/dl) | 25.01 ^b | 52.89 ^a | 21.11 ^b | 40.80^{a} | 4.86 | * | | |
| Cholesterol (mg/dl) | 135.60 ^a | 122.19 ^a | 99.97 ^b | 75.09 ^c | 5.26 | * | | |
| | | Liver funct | ion | | | | | |
| AST (Unit/l) | 177.75 ^a | 177.59 ^a | 194.66 ^a | 160.45 ^a | 15.43 | NS | | |
| ALT (Unit/l) | 69.39 ^b | 59.20 ^c | 60.60^{bc} | 85.09 ^a | 2.38 | * | | |
| Kidney function | | | | | | | | |
| Creatinine (mg/dl) | 7.73 ^b | 9.07^{ab} | 9.88 ^a | 8.80^{ab} | 0.56 | * | | |
| Uric acid (mg/dl) | 3.81 ^a | 3.18 ^a | 3.55 ^a | 3.39 ^a | 0.39 | NS | | |
| ab) (a () | | | | | | | | |

| Table 5. Blood parameters of the | different | experimental | groups |
|----------------------------------|-----------|--------------|--------|
|----------------------------------|-----------|--------------|--------|

SBM: Soybean meal.

TM: Thyme meal,

a, b, c and d: Means in the same row having different superscripts differ significantly (P<0.05).

SEM: Standard error of mean,

*: Significant at P<0.05.

NS: non-significant.

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase.

Fish body composition of different experimental groups

The data presented in Table (6) reveal that the Nile tilapia fed diets supplemented with thyme meal exhibited a significant (P<0.05) increase in body composition parameters, including moisture, crude protein (CP), and ash content. Conversely, there was a significant (P<0.05) reduction in the levels of dry matter (DM), organic matter (OM), ether extract (EE), and gross energy compared to the control group.

1

| | | Experimental diets | | | | | | | |
|-------------------------------|--|---|---|---|---|-----------|--------------------------------|--|--|
| Item | Body compositio n of initial fish | Control zero TM D ₁ | Replacing 10% of SBM by TM D ₂ | Replacing 20% of SBM by TM D ₃ | Replacing 30% of SBM by TM D ₄ | SE M | Sign. <i>P<</i> 0. 05 | | |
| Moisture | 76.52 | 72.75 ^c | 73.62 ^b | 76.93 ^a | 73.59 ^b | 0.48 3 | * | | |
| Dry matter (DM) | 23.48 | 27.25 ^a | 26.38 ^b | 23.07 ^c | 26.41 ^b | 0.48 3 | * | | |
| Chemical analysis on DM basis | | | | | | | | | |
| Organic matter (OM) | 85.33 | 88.79 ^a | 87.80 ^b | 85.43° | 81.75 ^d | 0.81 9 | * | | |
| Crude protein (CP) | 60.94 | 58.03 ^d | 62.67 ^b | 60.97 ^c | 64.16 ^a | 0.69 5 | * | | |
| Ether extract (EE) | 24.39 | 30.76 ^a | 25.13 ^b | 24.46 ^c | 17.59 ^d | 1.41 1 | * | | |
| Ash | 14.67 | 11.21 ^d | 12.20 ^c | 14.57 ^b | 18.25 ^a | 0.81 9 | * | | |
| Gross energy kcal/ 100g | 573.58 | 617.01 ^a | 590.31 ^b | 574.40 ^c | 527.85 ^d | 9.77 6 | * | | |
| Gross energy cal/ g DM | 5.7358 | 6.1701 ^a | 5.9031 ^b | 5.7440 ^c | 5.2785 ^d | 0.09 8 | * | | |

Table 6. Fish body composition of initial and different experimental groups fed tested diets

SBM: Soybean meal.

TM: Thyme meal.

a, b, c and d: Means in the same row having different superscripts differ significantly (P<0.05).

SEM: Standard error of mean, *: Significant at *P*<0.05.

Gross energy (kcal/ kg DM) was calculated according to **Blaxter (1968)** and **MacRae and Lobley (2003)**. Where, each g CP = 5.65 Kcal, g EE = 9.40 kcal and g CF and NFE = 4.15 Kcal.

Energy retention (ER)% and protein productive value (PPV)%

The results presented in Table (7) demonstrate that incorporating thyme meal (TM) into the diets of the Nile tilapia at various levels led to a significant (P<0.05) increase in energy retention percentage (ER%) and protein productive value percentage (PPV%) compared to the control group (D1). Specifically, the ER% values were enhanced by 3.11, 25.65, and 14.23% over the control. Additionally, the PPV% values showed a significant (P<0.05) increase of 25.54, 53.33, and 73.05% in the D2, D3, and D4 groups, respectively, compared to the control (D1).

| Experimental diets | | | | | _ | | | | |
|---|----------------------------------|-------------------------------------|--|--|--------|-------------------------|--|--|--|
| Item | Control zero TM | Replacing 10% of SBM by TM | Replacing 20% of SBM <u>by TM</u> | Replacing 30% of SBM <u>by TM</u> | SEM | Sign. <i>P</i> <0.05 | | | |
| | \mathbf{D}_1 | \mathbf{D}_2 | D_3 | D_4 | | | | | |
| Initial weight (IW), g | 142 | 145 | 144 | 141 | 0.640 | NS | | | |
| Final weight (FW), g | 308 ^c | 338 ^b | 360 ^a | 368 ^a | 7.116 | * | | | |
| | Calculation the energy retention | | | | | | | | |
| Energy content in final body fish 10 fish | 6.1701 ^a | 5.9031 ^b | 5.7440 ^c | 5.2785 ^d | 0.098 | * | | | |
| Total energy at the end in body fish (10 fish) | 1900 ^c | 1995 ^b | 2068 ^a | 1942 ^{bc} | 20.594 | * | | | |
| Energy content in initial body fish (cal /g) | | | 5.7358 | | | | | | |
| Total energy at the start in body fish (E_0) | 814 ^{ab} | 832 ^a | 826 ^{ab} | 809 ^b | 3.674 | * | | | |
| Energy retained in body fish $(E-E_0)$ | 1086 ^c | 1163 ^b | 1242 ^a | 1133 ^{bc} | 18.400 | * | | | |
| Energy of the feed intake (Cal / g feed) | 4.400 | 4.303 | 4.318 | 4.321 | - | - | | | |
| Quantity of feed intake | 496.40^{b} | 526.25 ^a | 459.40 ^c | 460.70 ^c | 8.500 | * | | | |
| Total energy of feed intake (EF) | 2184 ^b | 2264 ^a | 1984 ^c | 1991 ^c | 37.262 | * | | | |
| Energy retention (ER) % | 49.82 ^d | 51.37 ^c | 62.60 ^a | 56.91 ^b | 1.537 | * | | | |
| Calcul | lation the p | protein produ | ctive value (I | PPV) % | | | | | |
| Crude protein % in final body fish | 58.03 ^d | 62.67 ^b | 60.97 ^c | 64.16 ^a | 0.695 | * | | | |
| Total protein at the end in body fish (PR ₁) | 178.73 ^d | 211.82 ^c | 219.49 ^b | 236.11 ^a | 6.351 | * | | | |
| Crude protein % in initial body fish | | | 60.94 | | | | | | |
| Total protein at the start in body fish (PR ₂) | 86.53 ^{ab} | 88.36 ^a | 87.75 ^{ab} | 85.93 ^b | 0.389 | * | | | |
| Protein Energy retained in body fish | 92.20 ^d | 123.46 ^c | 131.74 ^b | 150.18 ^a | 6.350 | * | | | |
| $(\mathbf{PR}_3) = (\mathbf{PR}_1 - \mathbf{PR}_2)$ | | | | | | | | | |
| Crude protein in feed intake (CP %) | 27.50 | 27.67 | 27.69 | 27.89 | - | - | | | |
| Total Protein intake (PI), g | 136.51 ^b | 145.61 ^a | 127.21 ^c | 128.49 ^c | 2.260 | * | | | |
| Protein productive value (PPV) % | 67.54 ^d | 84.79 ^c | 103.56 ^b | 116.88 ^a | 5.638 | * | | | |

Table 7. Energy retention (ER) and protein productive value (PPV) % of different experimental groups

SBM: Soybean meal, TM: Thyme meal.

a, b, c and d: Means in the same row having different superscripts differ significantly

(P<0.05).SEM: Standard error of mean.

NS: Not significant.

*: Significant at *P*<0.05.

Economical evaluation of different experimental groups

The economic evaluation results shown in Table (8) indicate that incorporating thyme meal (TM) into the feed formulation led to a reduction in feed costs, decreasing from 24.350 LE per kg in the control diet (D1) to 23.80, 23.435, and 22.925 LE per kg for diets D2, D3, and D4, respectively. Additionally, the net improvement in economic efficiency was recorded at 8.53, 27.68, and 30.61% for D2, D3, and D4, respectively, compared to the control diet, which did not include TM.

| | Tested diets | | | | | |
|--------------------------------------|-----------------------|-------------------------------------|-------------------------------------|-------------------------------------|--|--|
| Item | Control zero TM | Replacing 10% of SBM by TM | Replacing 20% of SBM by TM | Replacing 30% of SBM by TM | | |
| | \mathbf{D}_1 | \mathbf{D}_2 | \mathbf{D}_3 | \mathbf{D}_4 | | |
| Costing of kg feed (LE) | 24.350 | 23.840 | 23.435 | 22.925 | | |
| Relative to control (%) | 100 | 97.91 | 96.24 | 94.15 | | |
| Feed conversion ratio (FCR) | 2.99 | 2.73 | 2.13 | 2.03 | | |
| Feeding cost (LE) per Kg weight gain | 72.81 | 65.08 | 49.92 | 46.54 | | |
| Relative to control (%) | 100 | 89.38 | 68.56 | 63.92 | | |
| Net improving in feeding cost (%) | Zero | 8.53 | 27.68 | 30.61 | | |

Table 8. Economical evaluation of different experimental groups

SBM: Soybean meal, TM: Thyme meal, LE. : Egyptian pound, Feed cost (L.E) FCR×FI. Cost per Kg diet: Diet formulation calculated according to the local prices at year 2024, as presented in Table (1).

DISCUSSION

The findings of this study demonstrate that growth performance and survival rates were significantly enhanced (P<0.05) in the groups of fish fed diets where 10%, 20%, and 30% of soybean meal (SBM) was replaced with thyme meal (TM). Specifically, parameters such as final weight (FW), total body weight gain (TBWG), average daily gain (ADG), and specific growth rate (SGR) all showed marked improvement. Additionally, survival rates (SR) were maintained at 100%, with no mortality observed in any of the experimental groups. The feed utilization data further revealed a significant (P<0.05) improvement in the feed conversion ratio (FCR) as the inclusion levels of TM in the diets increased. The D2 group exhibited the highest feed intake and crude protein intake compared to the control and other experimental groups.

These results align with those reported by **Ghafarifarsani** *et al.* (2022b), who found that the dietary inclusion of thyme essential oil (TVO) in common carp (*Cyprinuscarpio*) significantly improved final body weights and reduced FCR. Their study also reported no mortality in the groups supplemented with TVO. Other studies, such as those by **Dorojan** *et al.* (2014), **Emeish and El-Deen** (2016) and **Yilmaz** *et al.* (2016), have similarly documented the beneficial effects of thyme and its derivatives on fish growth and immunity across various species, including Nile tilapia (*Oreochromis niloticus*) and rainbow trout (*Oncorhynchusmykiss*).

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Regarding biochemical parameters, the current study supports the safety of using 10% TSM, which led to the highest serum protein and globulin levels without affecting liver or kidney function, apart from a rise in glucose levels, possibly indicating stress. These findings are consistent with **Ghafarifarsani** *et al.* (2021b), who observed enhanced serum protein levels in rainbow trout fed a diet supplemented with thyme extract oil. However, higher concentrations of TSM were associated with increased albumin, ALT, and creatinine levels and a reduction in globulin, suggesting potential liver stress or immune suppression, as noted by **Yousefi** *et al.* (2018) and **Hajirezaee** *et al.* (2020).

Fish body composition analysis revealed that diets containing TM significantly (P<0.05) increased moisture, crude protein (CP), and ash contents while decreasing dry matter (DM), organic matter (OM), ether extract (EE), and gross energy content compared to the control group. These outcomes are consistent with studies such as Srivastava *et al.* (2020), who found that dietary supplementation with ashwagandha root powder improved flesh quality in *L. rohita* fingerlings. Similar effects were noted in other research focusing on the replacement of traditional protein sources with alternatives like black soldier fly larvae meal (BSFM), which impacted lipid and protein contents in various fish species.

The study also highlighted the positive impact of TM on energy retention (ER%) and protein productive value (PPV%). Significant increases (P<0.05) in these parameters were observed in all experimental groups compared to the control, with ER% improving by 3.11%, 25.65%, and 14.23% and PPV% by 25.54%, 53.33%, and 73.05% for groups D2, D3, and D4, respectively. These findings are in line with those of **Abo-State** *et al.* (2021) and **Fayed** *et al.* (2023), who reported similar improvements in ER% and PPV% when fish diets were supplemented with alternative protein sources like β -glucan, and BSFM.

CONCLUSION

Based on the results obtained in this study, it can be concluded that incorporating thyme meal as a non-conventional protein source to replace 10%, 20%, and 30% of soybean meal in fish diets effectively improves growth performance, feed utilization, and feed conversion ratios. It also enhances total protein levels, increases DM, CP, EE, and gross energy contents in fish body composition, and improves energy retention and protein productive values. Additionally, the inclusion of thyme meal in the diets resulted in a reduction in feeding costs, making it an economically viable option for fish farming.

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REFERENCES

- Abo-State Hanan, A.; El-Monairy, M.M.; Hammouda Y.A. and Hassan H.M.A. (2021). Effect of dietary supplementation of manna oligosaccharide and β-glucan on the performance and feed utilization of Nile tilapia fingerlings. Curr. Sci. Intl, 10 (1) ISSN: 2077- 4435 DOI: 10.36632/csi/2021.10.1.21www.curresweb.com : 226-233.
- Ali, B.A. and El-Feky A. (2019). Enhancing growth performance and feed utilization using prebiotics in commercial diets of Nile Tilapia (*Oreochromis niloticus*) fingerlings. EJNF, 22 (1): 219-225.
- AlSafah, A.H. and Al-Faragi J.K. (2017). Influence of thyme (*Thymus vulgaris*) as feed additives on growth performance and antifungal activity on *Saprolegnia sp.* in Cyprinuscarpio L. J Entomol Zool Stud, 5(6): 1598-1602.
- **AOAC.** (2016). Official Methods of Analysis, 18th ed. Association of Official Analytical Chemists, Washington, DC, USA (2005). All content following this page was uploaded by Hayder N. Al-Mentafji on 02 February 2016.
- Blaxter, K.L. (1968). The energy metabolism of ruminants. 2nded. Charles Thomas Publisher. Spring field. Illinois, USA.
- **Cannon, D.C.; Olitzky, I. and Inkpen, J.A. (1974)**. Proteins. In: Clinical Chemistry, principles and techniques, 2nd ed., R.J. Henery, D.C. Cannon, J.W. Wink Elman (eds.), Harper and Row New York, pp: 407-421.
- **Caraway, W.T. and Watts, N.B. (1987).** Carbohydrates In: Fundamentals of clinical chemistry. 3^{ry} ed. Edited by Tietz, N.W. Philadelphia, WB Saunders. P: 422-47.
- **Dorojan, O.G.; Placinta, S. and Petrea, S. (2014)**. The influence of some phytobiotics (thyme, seabuckthorn) on the growth performance of stellate sturgeon (A. stellatus, Pallas, 1771) in an industrial recirculating aquaculture system. J Anim Sci Biotechnol, 47(1): 205-210.
- Duncan, D.B. (1955). Multiple Rang and Multiple F-Test Biometrics, 11: 1-42.<u>https://doi.org/10.2307/3001478</u>https://www.jstor.org/stable/3001478doi:10.2307/3 001478<u>https://scihub.yncjkj.com/10.2307/3001478https://en.wikipedia.org/wiki/Dunca</u> <u>n%27s_new_multiple_range_test</u>
- Ellefson, R.D. and Caraway, W.T. (1976). Fundamentals of clinical chemistry. Ed Tietz, N.W. Philadelphia, WB Saunders. p: 506.
- El-Saidy, D.M.S.D. and Gaber, M.M.A. (2003). Replacement of fish meal with a mixture of different plant protein sources in juvenile Nile tilapia, *Oreochromis niloticus* (L.) diets. Aquac. Res., 34: 1119-1127.
- **El-Sayed, A.F.M. (2021)**. Use of biofloc technology in shrimp aquaculture: a comprehensive review, with emphasis on the last decade. Rev. Aquacult., 13: 676–705.

- Emeish, W.F. and El-Deen, A.G.S. (2016). Immunomodulatory effects of thyme and fenugreek in sharp tooth catfish, *Clariasgariepinus*. AVMJ, 62(150): 45-51. doi: 10.21608/avmj.2016.169988.
- FAO. (2006). Fisheries Topics: Fisheries and Aquaculture Resources: FAO, Rome, Italy.
- FAO. (2009). Fisheries Topics: Fisheries Statistics and Information, FAO, Rome, Italy.
- **FAO.** (2012). The State of the World Fisheries and Aquaculture Part I: World Review of Fisheries and Aquaculture, FAO, Rome, Italy.
- Fayed, W.M.; Mansour, A.T.; Zaki, M.A.; Omar, E.A.; Moussa, N.A.A.; Taha, E.M. and Sallam, G.R. (2023). Water quality change, growth performance, health status in response to dietary inclusion of black soldier fly larvae meal in the diet of Nile tilapia, *Oreochromis niloticus*, Ann. Anim. Sci., DOI: 10.2478/aoas-2023-0088
- Ghafarifarsani, H.; Hoseinifar, S. H.; Sheikhlar, A.; Raissy, M.; Chaharmahali, F. H.; Maneepitaksanti, W .; Mehwish, F. and Van Doan, H. (2022). The effects of dietary thyme oil (*Thymus vulgaris*) essential oils for common carp (*Cyprinuscarpio*): growth performance, digestive enzyme activity, antioxidant defense, tissue and mucus immune parameters, and resistance against Aeromonas hydrophila. Aquac. Nutr., (1), 7942506. Sep.14:2022:7942506.doi: 10.1155/2022/7942506. Collection 2022.
- **Ghafarifarsani, H.; Kachuei, R. and Imani, A. (2021b)**. Dietary supplementation of garden thyme essential oil ameliorated the deteriorative effects of aflatoxin B1 on growth performance and intestinal inflammatory status of rainbow trout (*Oncorhynchusmykiss*). Aquac, 531, 735928.
- Hajirezaee, S.; Mohammadi, G. and Naserabad, S.S. (2020). The protective effects of vitamin C on common carp (*Cyprinuscarpio*) exposed to titanium oxide nano-particles (TiO₂-NPs). Aquac, 518, 734734.
- Hardy, R.W. (2010). Utilization of plant proteins in fish diets: effects of global demand and supplies of fishmeal. Aquac. Res., 41: 770-776.
- Ji, S.C.; Takaoka, O.; Jeong, G.S., *et al.* (2007). Dietary medicinal herbs improve growth and some non-specific immunity of red sea bream Pagrus major. Fish. Sci., 73(1):63-69. doi: 10.1111/j.1444-2906.2007.01302.x.
- MacRae, J. and Lobley, G.E. (2003). Some factors which influence thermal energy losses during the metabolism of ruminants Quelques facteurs des pertes d'energie thermique liées au métabolisme des ruminants Einige Faktoren mit Einfluss auf die Wärmeenergieverluste während des Stoffwechsels bei Wiederkäuern. <u>Livestock Production ScienceVolume 9, Issue 4</u>, July 1982, Pages 447-45 Accepted 16 December 1981, Available online 1 October 2003. <u>https://doi.org/10.1016/0301-6226(82)90050-1</u>
- NRC. (2011). National Research Council. Nutrient Requirement of Fish. National Academy Press, Washington, DC, USA.
- **Reitman, S.** and **Frankel, S.** (1957). Colorimetric determination of glutamic oxaloacetic and glutamic pyruvic transaminases, Am. J. Clin. Pathol., 28: 53-56.

- 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. doi: 10.1016/j.foodcont.2007.07.007.
- Silva, A. S.; Tewari, D.; Sureda, A.; Suntar, I.; Belwal, T.; Battino M., et al. (2021). The evidence of health benefits and food applications of *Thymus vulgaris L*. Trends Food Sci. Technol., 117, 218-227. doi: 10.1016/j.tifs.2021.11.010.
- **SPSS (2020)**. Statistical Package for Social Science (**Software version**: 22.0).
- Srivastava, A.; Ansal, M.D. and Khairnar, S.O. (2020). Effect of Ashwagandha (Withania somnifera) root powder supplementation on survival, growth and flesh quality of an Indian major carp, Labeorohita (Ham.) fingerlings. Amim Nutr Feed Techn, 20: 515-524 DOI: 10.5958/0974-181X.2020.00045.1
- Subasinghe, R.; Soto, D. and Jia, J. (2009). Global aquaculture and its role in sustainable development. Rev. Aquacult. 1, 2–9. doi: 10.1111/j.1753-5131.2008.01002.x
- **Tietz, N.W. (1990)**. Clinical guide to laboratory tests. 2nd ed. Philadelphia, WB Saunders. P: 566.
- Workagegn, K.B.; Ababbo, E.D.; Yimer, G.T. and Amare, T.A. (2014). Growth performance of the Nile tilapia (*Oreochromis niloticus*) fed different types of diets formulated from varieties of feed ingredients. J Aquac Res Development 5: 235 doi:10.4172/2155-9546.1000235
- Yilmaz, E.; Sebahattin, E. and Yilmaz, S. (2015). Influence of carvacrol on the growth performance, hematological, non-specific immune and serum biochemistry parameters in rainbow trout (*Oncorhynchusmykiss*). J. Nutr. Sci., 6(5): 523-531. doi: 10.4236/fns.2015.65054
- Yilmaz, S.; Ergün, S. and Çelik, E.Ş. (2016). Effect of dietary spice supplementations on welfare status of sea bass, *Dicentrarchuslabrax L.* Proceedings of the National Academy of Sciences, India Section B: Biol. Sci., 86(1): 229-237. doi: 10.1007/s40011-014-0444-2.
- Yousefi, M.; Hoseini, S.M.; Vatnikov, Y.A.; Nikishov, A.A. and Kulikov, E.V. (2018). Thymol as a new anesthetic in common carp (*Cyprinuscarpio*): Efficacy and physiological effects in comparison with eugenol. Aquac., 495, 376-383.
- Zadmajid, V. and Mohammadi, C. (2017). Dietary thyme essential oil (*Thymus vulgaris*) changes serum stress markers, enzyme activity, and hematological parameters in gibel carp (*Carassiusauratusgibelio*) exposed to silver nano-particles. Iran. J. Fish. Sci., 16(3): 1063-1084.
- 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, CBEES, 4: 220-227.

Zargar, A.; Rahimi-Afzal, Z.; Soltani, E.; Mirghaed, T.A.; Ebrahimzadeh, M.H.A., Soltani M., et al. (2019). Growth performance, immune responseand disease resistance of rainbow trout (*Oncorhynchusmykiss*) fed *Thymus vulgaris* essential oils. Aquac. Res. 50, 3097-3106. doi: 10.1111/are.14243.