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Effects of Some Feed Additives on Growth Performance and Physiological Parameters of *Oreochromis niloticus*

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ABSTRACT

A 100- day study was conducted to evaluate the influence of two natural additives in the tilapia diet on growth, food performance and physiological conditions. Hence, six treated diets (30 % protein, 3000 k cal) were used; the control (0% additives) and diets1, 2 and 3, containing 1%, 2% & 3% dried mulberry leaves (ML), respectively. Whereas, diets 4, 5 and 6 contained 1%, 2% & 3% dried guava leaves (GL). Fish were stocked in ponds with 30 fish/ part (2.5 x 2 x 1.5 metres), and an initial average weight of 24 ± 0.5 g. The best body weight gain, specific growth rate, feed conversion ratio and protein efficiency ratio were recorded for the fish fed on diets 4 & 5, which contained 1% & 2% of guava additive. Whereas, fish fed on diet 1 with 1% mulberry leaves ($p<0.05$) was rated the next, compared to fish fed on the control diet. The results revealed that the addition of guava leaves powder with levels 1% and 2 %, followed by 1% mulberry improved fish growth performance and feed utilization. In all fish treatments, the carcass composition data detected an increase in protein content accompanied with a decrease in fat percentage. Fish physiological measurements (plasma glucose, cholesterol and triglycerides) revealed that supplementing the Nile tilapia diets with the ML and GL showed a significant decrease ($p<0.05$) if compared with the control. An increase was recorded in the plasma total protein and globulin levels in all treated fish, compared to the control group, and accompanied with optimal liver enzymes activity. Hence, both ML and GL proved their beneficial impact on fish performance and nutrient utilization besides their role in enhancing the immune system of fish through vitamin C content in guava leaves that helps to maintain the health of blood vessels.

INTRODUCTION

Africa production of tilapia aquaculture accounted 1.2 MT **(FAO, 2017)**, last 10 years Tilapia species represented about 67% of total cultured species in 2014 **(Soliman & Yacout, 2016; Shaalan, *et al.*, 2018).** Nile tilapia (*Oreochromis niloticus*), blue tilapia (*Oreochromis aureus*) and the red tilapia are the common cultured tilapia species **(Sadek, 2011; Shaalan, *et al.*, 2018).**

The Nile tilapia is widely distributed in Africa and many of other parts of the world, Tilapia attributed to several factors, including high fecundity and rapid growth rates **(Trewaves, 1983; El-Shabrawy & Dumont, 2003)**

Use of natural unconventional feed sources, such as tree leaves, for animal production has attracted widespread attention.

Welker and Lim (2011) defined phytobiotics as substances that derived from plants to enhance fish health and growth. So, the use of compounds from plants is desirable to improve aquaculture, such as *Psidium guajava* **(Omitoyin *et al.*, 2019)**, *Echinacea purpurea* and *Allium sativum* **(Aly and Mamohamed, 2010),** mulberry and moringa **(Astuti *et al.*, 2012),** *Allium sativum* **(Shalaby *et al*., 2006)**, thyme and garlic **(Raky, 2009a)**, and ginger **(Raky, 2009b)**

Morus spp., such as Mulberry (*Morus alba*, Linn.) are widely distributed throughout Asia, Europe, Africa and the Americas, and have been used in animal production since the late 1980s. The mulberry leaves are highly palatable and easily digestible (70-90%) for herbivores and can also be fed to monogastrics. Mulberry leaves and young stems content on 15-28% crude protein that is similar to most legume forages (**Sanchez, 2000**). The nutritive value of mulberry leaves estimated as a protein source for beef cattle **(Huyen *et al*., 2012)**, sheep **(Kandylis *et al*., 2009)**, pigs **(Ly *et al*., 2001),** laying hens and broilers **(Al-Kirshi *et al*., 2009).**

Mulberry plant is cultivated for silkworm feeding and their leaves contain a good quantity protein (21.1%) which also can be used as a total substitute (100%) for dietary fish meal for catfish. **(Bag *et al*., 2012)**

Guava is widely cultivated in Africa, and incorporated into agroforestry systems in India **(CABI, 2013)**. Guava leaves can be used as fodder **(Heuzé *et al*., 2015).**

Guava plant plays an important role as natural antioxidant due to its high content of polyphenols **(Jiménez-Escrig *et al*., 2001)**, employed for treating diarrhea and digestion **(Kumar *et al*., 2021)**, and used for hepatoprotection, cytotoxic, cardioactive, antiplasmodial, anti-inflamatory, due to its content of many metabolites like phenolic, flavonoid, carotenoid, terpenoid and triterpene, that found in leaves and fruits **(Gutiérrez *et al*., 2008).**

The guava (*Psidium guajava L*.) tree belonging to the Myrtaceae family, is a very unique and traditional plant which is grown due to its various medicinal and nutritive properties. **(Kumar *et al.*, 2021).**Also**, Jassal *et al*. (2019),** reported that guava leaves can be utilized as a novel and sustainable dietary source as they are a rich source of proteins, carbohydrates, and dietary fibers.

The present study was conducted to evaluate influence of two natural additives (mulberry and guava) dried leaves in tilapia diet to boost growth performance, feed utilization and physiological parameters.

MATERIALS AND METHODS

Experimental design:

Samples of the Nile tilapia, *Oreochromis niloticus* were collected from El Qanater El Khairya fish farm with initial body weight of 24 g. They were reared in two concrete ponds, each of which was divided into 8 equal parts by using nylon net with small mesh size (3.0 mm).For acclimatization, the stocked fish (with density 30 fish/part) were held under optimal conditions for2 weeks before starting the growth trial. The experiment lasted for 100 days. Fish were fed twice a day at 10:30 am and 14:30 pm and were biweekly weighed.

Water quality parameters:

The mean values of water quality parameter (±SE) were as follows: water temperature: 28.9 ± 0.3°C; dissolved oxygen: 5 - 6 ± 0.4 mg/L; pH: 7.5 ± 0.2; total ammonia: 0.023 ± 0.01 mg/L; nitrite: 0.025 ± 0.013 mg/ L and nitrate: 0.8 ± 0.4 mg/L. All water quality parameters were within the acceptable range for rearing the tilapia **(Makori *et al.*, 2017)**.

Preparing additives:

Leaves of *Psidium guaijava* L. (Guava), and *Morus alba L*. (Mulberry) were randomly harvested by hand from several trees. Leaves were washed by tap water, sun-dried and crushed into powder to be used in fish diets.

Experimental diets:

In this study, six experimental diets were used as follows: Diet 1 (1% ML), Diet 2 (2% ML), Diet 3 (3% ML), Diet 4 (1% GL), Diet 5 (2% GL), Diet 6 (3% GL) and the control (without additives). The diets formula and chemical composition were shown in Tables (1 & 2).

Table1. Experimental diets composition

Feed ingredient	Control	Mulberry leaves			Guava leaves		
		Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6
Fish meal 62 %	21	21	21	21	21	21	21
Soybean meal 44%	20	20	20	20	20	20	20
Yellow corn	33	33	32	32	33	32	32
Wheat bran	18	17	17	16	17	17	16
Sunflower oil	6	6	6	6	6	6	6
Premix*	2	2	2	2	2	2	2
Feed additive	0	1	2	3	1	2	3

* One kg premix contained:

Vitamins:- $48x10^5$ I.U (A), $6x10^2$ mg (B_6), 20 mg (Biotin), $8x10^5$ I.U. (D_3), 144 mg (E), 400 mg (B_1), 1600 mg (B_2), $4x10^3$ mg (Pantothenic acid), 4 mg (B_{12}), $4x10^2$ mg (Niacin), $2x10^5$ mg (Choline chloride) and 400 mg (folic acid).

Minerals premix:- $12x10^3$ mg Iron, $16x10^3$ mg Manganese, $12x10^2$ mg Copper, 120 mg Iodine, 80 mg Cobalt, 40 mg Selenium and $16x10^3$ mg Zinc.

Table 2. Chemical analysis of diet and additives

Chemical composition	Experimental diet	Guava leaves (GL)	Mulberry leaves (ML)
Crude protein %	29.18	7.44	8.76
Ether extract %	6.8	5.56	6.03
Crude fiber %	3.7	8.5	5
Nitrogen free extract %	47.7	68.26	72.72
Ash %	10	10.24	7.49
Metabolizable energy (kcal/kg)	2990.2	-	-
Protein/Energy ratio	98.71	-	-

Fish performance and feed utilization parameters

Fish growth performance and feed utilization parameters were calculated according to the method of **Cho and Kaushik (1985)** as the following:

Body weight gain (**BWG**, g /fish) = [final body weight (g) - initial body weight (g)];

Daily weigh gain, (**DWG**, g /fish /day) = [BWG (g) / Experimental period (days)];

Specific growth rate (**SGR**, %g/day) = 100 [Ln final weight - Ln initial weight] / Experimental period (day);

Feed conversion ratio (**FCR**) = feed intake (g) / body weight gain (g);

Protein efficiency ratio (**PER**) = gain in weight (g) / protein intake in feed (g);

Protein productive value (**PPV**, %) =100 [protein gain in fish (g) / protein intake in feed (g)]; and

Survival rate % = 100 [Initial number of fish stocked-Mortality] / Initial number of fish stocked

Physiological parameters

Blood parameters

At the end of experiment, blood samples were collected using heparinized syringes from the caudal veins of the fish. Blood was centrifuged at 3000rpm for 15 min. Samples were subjected to determination of plasma total protein (PTP) and plasma albumin (PA) according to the method of **Armstrong and Carr (1964)** and **Doumas *et al*., (1977)**, respectively. Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined according to **Rettman and Frankel (1975)**. Serum triglycerides (STG) and cholesterol (Chol.) were determined according to the method described by **Stein (1986)**. Alkaline phosphatase (ALP) activity was determined by using the method of **Williason (2003)**. While, glucose concentration was measured according to the method of **Trainder (1969)**.

Chemical analysis

Chemical analysis of the experimental diets, additives and fish body were conducted to determine the percentages of dry matter (DM %),crude protein (CP %), ether extract (EE %), crude fiber (CF %), and ash % according to the **AOAC method (2012)**. Nitrogen free extract (NFE %) was calculated by differences; deducting the sum of CP%, EE%, CF%, and ash% from 100.

Statistical analysis

The data were subjected to analysis of variance (ANOVA) using general linear models (GLM) procedure; the software used was SPSS (Version 16.0) **(SPSS, 1997)**. Duncan's multiple range tests **(Duncan, 1955)** was used to compare between means of the control and treated groups.

The model of analysis was as follows:

$$\mathbf{Y_{ij} = \mu + T_i + E_{ij}}$$

μ = the overall mean.

T_i = the effect of treatment and

E_{ij} = the random error.

RESULTS AND DISCUSSION

1. Mulberry (*Morus alba*) dried leaves additive

1.1. Growth performance and feed utilization of tilapia fed *Morus alba* leaves

The current findings presented in Table (3) reveal that, growth performance of fish fed on diet 1, which contain 1% ML, were improved with respect to total weight gain, specific growth and survival rates (110 g, 1.73 % and 92 %) respectively, compared to those fed the other diets.

Table (4) shows an improvement in feed the utilization values; FI (207.6 g), PPV (33.78 %), FCR (1.89) and PER (2.0), for the test of fish fed diet 1.

These results of growth performance and feed utilization are semi close to those of **Bag *et al.* (2012),** who fed sting catfish on the mulberry leaves**.**

In literature, no enough information is found about adding mulberry leaves in fish diet. The present results showed that growth parameters of the fish fed on (levels 2 % and 3 %) mulberry leaves additive recorded a negative slightly affect, when compared to the control. **Astuti *et al.*(2012)** recorded that mulberry leaves which contained secondary compounds (saponin, phytic and tannin) may affect fish performance. This result may be due to the presence of anti-nutritional factors and complex carbohydrates in mulberry leaves **(NRC, 2011)**.

1.2. Body chemical composition of fish fed *Morus alba*

The chemical composition of fish body presented in Table (5)shows that the highest protein and ash percentage (56.49 & 21.83) were recorded for fish fed on diet 1,and at the same time, the lowest lipid content was detected.

Results of **Miao *et al*. (2020**) agree with those of the present study noting that the low level of dietary mulberry leaf meal increased protein content and ash content of the whole body compared to higher levels of the additive. This finding coincides with that of **Khalil *et al*. (2015**) who fed the Nile tilapia on *Eruca sativa* L. additive and noticed that the highest protein level was accompanied with the lowest lipid, which also agrees with the findings of **El-Ebiary and Zaki (2003)** and **Abdelhamid *et al*.(2007)**.

Table 3. Growth performance of the Nile tilapia fed mulberry (ML) and guava leaves (GL) (Mean ± SE)

Parameter	Experimental diet						
	Control	Mulberry leaves			Guava leaves		
		Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6
Initial weight	24±0.12	24± 0.11	24±0.10	25±0.09	27±0.15	28±0.13	27±0.91
Final weight	127^{d} ±0.63	134^{c}±1.01	114^{e}±0.82	100^{g}±0.95	174^{b}±0.65	178^{a}±0.55	110^{f}±0.59
Body weight gain	104^{c} ± 0.95	110^{b} ± 1.01	92^{d} ± 0.81	75^{f} ±0.65	148^{a} ±0.59	150^{a} ±0.71	84^{e} ±0.69
Daily weight gain	1.04^{c} ±0.01	1.1^{b} ±0.02	0.92^{d} ±0.01	0.75^{f} ±0.01	1.48^{a} ±0.02	1.50^{a} ±0.03	0.84^{e} ±0.01
Specific growth rate	1.67^{c} ±0.03	1.73^{b} ±0.01	1.56^{d}±0.04	1.39^{f} ±0.03	1.86^{a} ±0.05	1.85^{a} ±0.02	1.41^{e} ±0.06
Survival %	90 ±0.91	92 ±1.10	91 ±1.02	91 ±0.97	94 ±1.03	95 ±0.85	91 ±0.99

Table 4. Feed utilization parameters of fish fed ML and GL (Mean ± SE)

Parameters	Experimental diets						
	Control	Mulberry leaves			Guava leaves		
		Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6
Feed intake	197.7^{d}±1.03	207.6^{c}±1.32	182.9^{e}±1.40	166.2^{g}±1.05	262.1^{a}±1.51	247.8^{b}±1.26	181.9^{f}±1.42
Feed (FI) conversion ratio (FCR)	1.91^{b} ±0.01	1.89^{c} ±0.2	1.99^{b} ±0.01	2.23^{a} ±0.02	1.78^{d} ±0.01	1.78^{d} ±0.002	2.18^{a} ±0.01
Protein efficiency ratio (PER)	1.97^{c} ±0.01	2.0^{b} ±0.02	1.88^{d} ±0.01	1.69^{f} ±0.01	2.12^{a} ±0.01	2.11^{a} ±0.01	1.73^{e} ±0.01
Protein productive value (PPV)	29.35^{c} ±0.2	33.78^{a} ±0.3	29.56^{c} ±0.1	27.20^{d}±0.2	30.92^{b} ±0.2	30.67^{b} ±0.1	26.16^{e} ±0.1

1.3. Physiological parameters of fish fed *Morus alba*

Results revealed that, supplementing the Nile tilapia diets with the ML significantly ($p<0.05$) decreased plasma glucose, serum triglycerides and total cholesterol compared to those in the control group (Table 6). These results are similar to data of **Raky (2009b)** who used ginger additive and those of **Khalil *et al.* (2015) using** rocket leaves additive in the Nile tilapia diets.

The plasma total protein (PTP),for fish fed on the three ML levels, recorded optimum values higher than those of the control. While, plasma albumin values showed an increase through all tested fish groups ($p>0.05$). The present results are similar to those of **Metwally (2009) who used** garlic as an additive**,** and those of **Raky (2009a, 2009b)**who added garlic, thyme and ginger to diets of the tilapia; in addition, they concur with those of **Khalil *et al*. (2015)**who added rocket leaves.

Table (6) shows that mulberry leaves reduced the activities of AST and ALT, compared to those in the control diet. This finding is in agreement with those of **Metwally (2009)** and **Zaki *et al*. (2012)** who fed *O. niloticus* on garlic and different medicinal plants..

Table 5. Chemical analysis of whole fish body of the Nile tilapia (Mean ± SE)

Parameter	Experimental diet						
	Control	Mulberry leaves			Guava leaves		
		Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6
Moisture %	70.15^{f}±0.05	73.65^{b}±0.05	71.25^{e}±0.07	69.35^{g}±0.04	75.15^{a}±0.5	72.2^{c}±0.06	71.45^{d}±0.05
Crude protein %	56.7^{a}±0.53	56.49^{a}±0.42	54.58^{b}±0.63	52.55^{c}±0.19	54.71^{b}±0.75	54.53^{b}±0.51	52.99^{c}±0.92
Ether extract %	17.1^{e}±0.01	19.91^{d}±0.02	22.39^{c}±0.04	24.41^{a}±0.04	17.28^{e}±0.2	18.81^{d}±0.1	23.38^{b}±0.1
Ash %	25.17^{a}±0.04	21.83^{c}±0.2	20.9^{d}±0.1	17.89^{e}±0.09	24.41^{b}±0.06	24.54^{b}±0.06	21.61^{c}±0.04
Nitrogen free extract %	1.03^{e}±0.02	1.77^{d}±0.2	2.13^{c}±0.04	4.15^{a}±0.06	3.62^{b}±0.3	2.12^{c}±0.03	2.02^{c}±0.09
Dry matter %	29.85^{b}±0.05	26.35^{f}±0.05	28.75^{c}±0.05	30.65^{a}±0.05	24.85^{g}±0.05	27.65^{e}±0.05	28.55^{d}±0.05

Table 6. Physiological parameters of tilapia fed on guava and mulberry leaves supplemented diets (Mean ± SE)

Parameter	Experimental diet						
	Control	Mulberry leaves			Guava leaves		
		Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6
Plasma Glucose. (mg/ dl)	104.3^{a} ±1.04	85^{d} ±0.91	84^{d} ±0.90	82.9^{e} ±0.87	92^{c} ±0.75	89^{c} ±0.81	96^{b} ±0.59
Plasma total protein (g/dl)	2.82^{c} ±0.11	3.12^{b} ±0.09	3.10^{b} ±0.12	3.11^{b} ±0.07	3.25^{a} ±0.21	3.31^{a} ±0.06	3.10^{b} ±0.10
Plasma albumin (g/dl)	1.6^{b} ±0.02	1.79^{a} ±0.11	1.65^{b} ±0.03	1.72^{ab} ±0.01	1.38^{c} ±0.04	1.28^{d} ±0.02	1.41^{c} ±0.01
Plasma globulin (g/dl)	1.22^{e} ±0.01	1.33^{d} ±0.02	1.45^{c} ±0.04	1.39^{cd} ±0.03	1.87^{b} ±0.01	2.03^{a} ±0.10	1.69^{b} ±0.05
S. triglycerides (mg/dl)	62.06^{a} ±0.58	57.4^{d} ±0.75	58.9^{c} ±0.63	57.0^{d} ±0.54	60.10^{bc} ±0.47	59^{c} ±0.68	60.8^{b} ±0.81
Total Cholesterol (mg/ dl)	127.9^{a} ±1.09	119.0^{b} ±1.21	118.5^{b} ±1.32	113.4^{d} ±1.19	117.6^{c} ±1.35	118.8^{b} ±1.45	119.2^{b} ±1.07
Alkaline phosphatase (U/L)	49^{a} ±0.42	42^{d} ±0.36	42^{d} ±0.47	39^{e} ±0.53	44.1^{c} ±0.61	45.5^{b} ±0.50	44^{c} ±0.41
Alanine aminotransferase(U/L)	46.1^{a} ±0.32	39.2^{b} ±0.29	36.3^{c} ±0.48	34.9^{d} ±0.42	36.0^{c} ±0.27	36.0^{c} ±0.52	39^{b} ±0.39
Aspartate aminotransferase(U/L)	118.5^{a} ±1.07	87.8^{f} ±0.98	91^{e} ±1.02	91.5^{e} ±0.90	107.8^{c} ±1.11	112^{b} ±1.04	101^{d} ±0.99

The high level of liver enzymes (ALT and AST) in blood is usually an indication of cellular damage in stressed fish **(Chatterjee *et al.*, 2006; Soosean *et al.*, 2010)**

The current results are promising and assume that the mulberry leaves additive protected the membrane integrity of the liver cells against stressors.

The optimal values of alkaline phosphatase (ALP) were recorded for those fed ML compared to those of the control as shown in Table (6).

The present results of physiological parameters are in agreement with those of **Bag *et al.* (2012)** who mentioned that, adding mulberry leaf meal in sting catfish diet was accepted with respect to the reaction of fish and improved their immunity against common diseases.

Georgieva *et al*. (2020) recorded that diets supplemented with the phytoextracts of curcumin, paprika, thyme, oregano and garlic enhanced ALT and AST activities and increased plasma proteins in common carp.

2. Guava (*Psidium guaijava*) dried leaves additive

2.1. Growth performance and feed utilization of fish fed *Psidium guaijava* leaves

Present results showed that, best values of body weight gain (148 g & 150 g) and specific growth rate (1.86 % &1.85 %) were recorded for fish fed on diets 4 & 5, which contained 1% & 2% guava leaves (Table 3). The present result revealed that using 1% or 2% guava leaf additive can enhance tilapia growth. This result coincides with that of **Omitoyin *et al*. (2019)** who found that the Nile tilapia fed on diet containing guava additive recorded the best SGR (1.47 & 1.56 %) at levels 0.25 % & 0.50 %. Moreover, **Setufe *et al*. (2018)** noticed that *Clarias gariepinus* fed on diets containing guava leaves additives enhanced growth performance of fish. Table (4) shows the best values of PER (2.12, & 2.11), FCR (1.78 & 1.78), respectively, and the best results of feed intake (262.1 & 247.8g) and PPV (30.92 & 30.67) attributed to the tilapia fed on diets 4 and 5 and performed better than other groups ($p<0.5$). These results agree with those of **Omitoyin *et al*. (2019)** who reported that the best FCR, PER and FI were recorded for the Nile tilapia fed on guava additive.

These results are somewhat similar with those obtained by **Raky (2009a)**, who using thyme and garlic (powder and oil) additives observed that, the results of FCR, PER and PPV for the tilapia fingerlings were improved.

It is worth mentioning that, the guava leaves enhanced growth performance (gain in weight, SGR, FCR PER, FI and PPV).This may be due to the presence of minerals in guava leaves such as calcium, potassium, magnesium and phosphorus. Additionally, guava leaf diets contained phytochemicals such as terpenoids, steroids, flavonoids and phenols**. (NRC, 2011; Kumar *et al.*, 2021).**

Dietary inclusion of medicinal plants such as garlic **(El-Saidy & Gaber, 1997)**; onion and garlic (**Zaki & El-Ebiary, 2003)**; *Allium sativum* and *Thymus vulgari*s **(Raky, 2009a**); honey bee pollen (**EL-Asely *et al.*, 2014**); barely and onion **(Marzouk *et al.*, 2017)** increase fish growth performance, nutrients utilization, chemical composition and survival rate.

The present data showed that, the survival rate increased in all fish groups fed on diets containing additives compared to the control. This finding is in agreement with those of **Pachanawan *et al*. (2008)** and **Omitoyin *et al*. (2019)** who recorded that, fish diets supplemented with guava dried leaves and its extraction reduced mortality of *A. hydrophilla* infected tilapia without side effects on tested fish. Furthermore, **Arima and Danno (2002)** recorded that, four antibacterial compounds isolated from guava leaves may improve fish growth and survival.

2.2. Chemical composition of fish fed *Psidium guaijava*:

The results of carcass composition cleared that the protein content of fish fed diet 4 & 5 was higher than those fed diet 6, and at the same time diet 4 & 5 showed the lowest lipid content. These results agree with those of **Khalil *et al*. (2015**) who fed the Nile tilapia fingerlings on rocket leaves *Eruca sativa* L. additive and noticed that highest protein values were accompanied with the lowest lipid.

On contrary to the present findings, **Abo-State *et al.*(2017)** found that there are no significant differences in carcass composition of the Nile tilapia when fed different levels of phytogenic feed additives (Silaacid encapsulated).

2.3. Physiological parameters of fish fed *Psidium guaijava*:

Blood results presented in Table (6) reveal that plasma glucose (Gluc.), serum triglycerides (STG) and cholesterol (Chol.) of fish fed diet supplemented by three levels of guava leaves decreased significantly ($p<0.05$), compared to those of the control.

These results are similar to the data of **Raky (2009b)** who verified that plasma glucose, serum triglycerides and total cholesterol decreased with the addition of ginger additive to the Nile tilapia diets. On the other hand, the present results of plasma total protein (PTP) for those fed guava leaves recorded highest values (3.25, 3.31, & 3.10 g/dl) compared to the control (2.82 g/dl). Additionally, the higher plasma globulin (PG) 1.87, 2.03, 1.69 values were recorded for the fish fed the three levels of the same additive when compared to the control group.

Metwally (2009) mentioned that plasma total protein of the Nile tilapia fed on *Allium sativum* were higher than that in the control.

Regarding liver enzymes of ALT (36.0, 36.0, and 39.0 U/L) and AST (107.8, 112.0, and 101.0 U/L) of the fish fed diet treated with guava revealed the optimum levels compared to fish fed the control diet (46.1, 118.5 U/L).The same trend was recorded with respect to the optimum alkaline phosphatase (ALP) values (45.5, 44.1, and 44 U/L) having the lowest values compared to fish fed on the control diet.

The current best results of ALT & AST are in agreement with those of **Omitoyin *et al*. (2019)** who recorded that, the *P. guajava* extract enhanced the activities of liver function when added to the Nile tilapia diet. This finding suggests that the guava leaves protect the liver health against stressors.

The current study detected an improvement in the blood parameters accompanied with growth parameters enhancement. This observation may be due to the stimulation of pancreatic enzymes secretion that are necessary in nutrient digestion and adsorption (**Frankic *et al.*, 2009**). In this respect, **Marzouk *et al.* (2017)** recorded an enhancement in the growth performance and the hematological parameters of the Nile tilapia when fed on diets supplemented with barely and onion.

CONCLUSION

The present findings presented promising results by using the two natural additives (1 % Mulberry and 2 % Guava) of dried leaves for the diets of the Nile tilapia to enhance growth performance, feed utilization, physiological condition, and protect factor to maintain membrane integrity of the liver cells against stressors and immune response system of fish.

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