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Effect of Partially Replacing Corn Meal by Date Stone on Growth Performance in Nile Tilapia Fingerlings, Diets Supplemented with Marjoram: Effects of Date Stone on Growth Nile Tilapia

Gaber M.M.^{1*}, Labib E.H.², Omar E.A.³, Zaki M.A.⁴ and Nour A.M.⁵

^{*1}National Institute of Oceanography and Fisheries, Cairo Egypt.

²Research Center, Ministry of Agriculture and Land Reclamation, Dokki, Giza Egypt.

³Departments of Animal and Fish Production, Faculty of Agriculture, Saba Basha, Alexandria Univ., Egypt.

^{4,5}Departments of Animal and Fish Production, Faculty of Agriculture, El Shatby, Alexandria Univ., Egypt.

ARTICLE INFO	ABSTRACT
Article No.: 022013483/ DOI: 10.15580/GJAS.2014.1.022013483 Submitted: 20/02/2013 Accepted: 25/09/2014 Published: 15/01/2014 *Corresponding Author Gaber M.M. E-mail: gabermagdy@yahoo.com	A 3-month randomized factorial design 4x2 (four levels of Date stone (DS) (0, 15, 30, and 45%), two levels (0 and 0.03%) of Marjoram (M) and three replicates). The trial was conducted in twenty four glass aquaria. Ten fingerlings of Nile tilapia were placed in each aquarium with an average weight 15.40 ± 0.3 g fish. Fish were fed twice daily (six days a week) at a rate of 3% of body weight. The results revealed that, mean final weight (g/fish), SGR (% /day), feed conversion ratio, PPV and PER, were significantly ($p \leq 0.05$) affected by the levels of DS and level of M. And the best diet achieved which contain 15% DS and supplemented with 0.03 % M was not significantly different from control diet in fish performance. From the above results and the economic information of these study it can be concluded that, diet containing 15% of DS with 0.03% M exhibited the highest net profit and would seem to be the most desirable level of DS and M in the system studied.

Keywords:

Nile tilapia, date stone, Marjoram

INTRODUCTION

Annual production of date stone in Egypt is about 1.1 million tons representing 16% of total world production (FAO, 2004). Approximately 10-15% of this amount represents the total possible crop of date stone (110000-165000 tons annually). However, this amount was not all available, only those from manufactured date can be collected. Date stone are difficult to grind in an ordinary hammer mill (EL-Shazly et al., 1963). Therefore, they are first crushed and powdered with grain grinder stone mill. Yousif et al. (1996) reported that, the inclusion of the date stone and date pits did not influence fish growth. Increasing body protein and decreasing body fat was observed in fish fed on diets supplemented with date pits meal due to estrogenic action of date pits that was hypothesized as direct cause of this phenomenon. Azaza et al. (2008) noted that, waste date meal could be a substitute for soybean meal up to 300gkg⁻¹ without compromising growth of Nile tilapia. On the other hand El-Sayed et al. (2006) found that date pits based diets replaced with up to 75% wheat bran resulted in reduced growth rates and feed utilization efficiency of Nile tilapia. El-Sayed et al. further reported that, fungi degraded pits (DDP) could replace 300/kg dietary corn. While Belal (2008) found that, growth performance of Nile tilapia fed sprouted date pits diets are not different from control diet. Also, Gaber et al. (2012) found that DS could replace 150/kg dietary corn and which is not different from control diet. This study was carried out to determine the feasibility of using date stone as energy source for replacement corn meal in practical diets supplemented with 0.03% Digestarom on Nile tilapia fingerlings.

MATERIAL AND METHODS

Experimental diets

The date stone were collected from wet date. The date stone were dried in oven dried at 60-80°C for 72 hrs. Then crushed in disc crusher and then powdered with grain grinding stone mill. The other materials were finely ground in a house blender and used in the formulation of eight experimental diets isonitrogenous (303 g kg⁻¹) and isocaloric (18.4 kJ g⁻¹). The experiment was designed in a factorial design (4 x 2). Eight tested diets were formulated to contain 0, 15, 30, and 45% DS instead of YC, without or with 0.03 Marjoram as described in Table 1. Each ingredient was thoroughly mixed with the other ingredients, vitamins, minerals mix and 0.03 Marjoram was then added to the diet 5-8 with continuous mixing. (Marjoram, *Majorana hortensis* is a leaves extract (M) that stimulate digestibility). A few drops of soybean oil was added at the same time of mixing warm distilled water (45°C) which was slowly added until the diets began to clump. Diets were processed by a California pellet mill machine and dried for 48 hrs at 70°C in a drying oven. The experimental pellets were soft enough for the fish to take and retain. The processed diet particle size was 0.6 mm diameter

and 2 mm length. The experimental fish were fed the test diets for one week as adaptation period to adapt them to these test diets. After the adaptation period was completed, fish in each aquarium were reweighed, and their initial weights were recorded. Fish in each aquarium were fed twice daily (six days a week) at a rate of 3% of body weight for 90 days.

The experimental work of the present study was carried out at the Department of Animal and Fish Production, Faculty of Agriculture (El-Shatby); Alexandria University, Egypt.

Culture condition

Nile tilapia (*Oreochromis niloticus*) fingerlings obtained from Berseek Fish Hatchery, El-Behera Governorate were used in the present study. Fish were placed randomly in twenty four glass aquaria with dimensions of 100×40×30 cm and 100 l capacity of water per aquarium, three replicates per treatment were used in this study. Each aquarium stocked with ten fingerlings of Nile tilapia with an average initial body weight of 15.40 ± 0.3 g fish. Each aquarium was cleaned daily in order to prevent accumulation of fecal materials and reduce the growth of algae, and the same amount of fresh water was used to refill the aquaria. Water was partially changed once every three days using fresh water. Aeration was continuously provided using an air blower. The test diets were fed to triplicate aquaria two times daily at the rate of 3% of body weight for 90 days.

Procedure of Marjoram leaf extraction

Procedure for extraction carried out according to Thakare (2004) as follows:

1. 100 g of plant samples was put in 1000 ml ethanol solvent (95/100 ml).
2. The flasks of all the solvents were stoppard and agitated with a magnetic stirrer for 24 hours at room temperature.
3. Then the solution was centrifuged at 3000 rpm for 10 min and the residue then extracted with 500 ml of methanol for 24 hours with stirring at room temperature.
4. Evaporate Methanol from the solution at approximately 42°C by using a rotary evaporator.
5. The extracts were collected and diluted with ethanol solvent to about 150 ml and stored at 4 °C for further use.

Preparation of samples

At the end of twelve weeks of growth study, fish per tank were collected for analysis. Fish were homogenized for whole body analysis and frozen at -18 °C. The homogenized samples were oven dried at 60 - 80°C for 48 hrs. Proximate analyses of whole body, protein, lipid, and ash were performed according to standard AOAC (2000) methods and gross energy (Ballistic bomb calorimeter, Gallenkamp, England).

Water quality

Water quality parameters (temperature, dissolved oxygen, pH, ammonia, nitrate and nitrite) were monitored to ensure water quality remained well within limits recommended for Nile tilapia. Water temperature and dissolved oxygen were measured every other day using an YSI Model 58 oxygen meter (Yellow Springs Instruments, Yellow Springs, OH). Ammonia and nitrite were measured at wkly intervals. Alkalinity was monitored twice weekly using the titration methods of Golterman *et al.* (1978). pH was monitored twice weekly using an electronic pH meter (pH pen Fisher Scientific, Cincinnati, OH). During the feeding trial, the water quality parameter averaged ($\pm$ SD): water temperature $27.8 \pm 0.8^\circ\text{C}$ dissolved oxygen $4.8 \pm 0.4 \text{ mg l}^{-1}$; pH 7.4 ± 0.6 ; ammonia $0.01 \pm 0.04 \text{ mg l}^{-1}$; nitrite $0.1 \pm 0.05 \text{ mg l}^{-1}$; nitrate $1.5 \pm 2 \text{ mg l}^{-1}$; alkalinity $181 \pm 46 \text{ mg l}^{-1}$.

Calculations and statistical analysis

Calculations of growth parameters were conducted according to Cho and Kaushik (1985). Data were analyzed by analysis of variance (ANOVA) using the SAS ANOVA procedure (Statistical analysis system, 1988). Duncan's multiple range tests was used to compare differences among individual means. Treatment effects were considered significant at $P < 0.05$. All percentages and ratio were transformed to arcsine values prior to analysis (Zar, 1984).

RESULTS

There were no significant differences in water quality parameter among the treatments during experimental period. Water quality parameters were within the acceptable range for Nile tilapia growth (Stickney, 1979).

Table 1: composition of experimental diets fed to Nile tilapia fingerlings (g.100 g⁻¹diet)

Ingredients	Diets							
	Without Supplement M				With D 0.03% M			
	1 (0%)	2 (15%)	3 (30%)	4 (45%)	5 (0%)	6 (15%)	7 (30%)	8 (45%)
Fish meal	23.0	23.0	23.0	23.0	23.0	23.0	23.0	23.0
Soybean meal	29.0	29.0	29.0	29.0	29.0	29.0	29.0	29.0
Yellow corn meal	45.0	38.25	38.25	24.75	45.0	38.25	38.25	24.75
Date stone (DS) ¹	-	6.75	6.75	20.25	-	6.75	6.75	20.25
Soybean oil	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Vita. & min ²	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Marjoram	--	--	--	--	0.03	0.03	0.03	0.03
Proximate analyses (%)								
Moisture	9.09	9.08	9.36	9.52	9.11	9.05	9.33	9.52
Crude protein	30.45	30.49	30.32	30.30	30.45	30.49	30.32	30.30
Crude fat	6.27	6.30	6.16	6.20	6.27	6.30	6.16	6.10
Ash	7.71	7.72	8.6	9.56	7.73	7.75	8.9	9.56
Crude fiber	4.89	4.9	4.58	4.11	4.89	4.9	4.58	4.11
NFE ⁴	50.64	50.55	49.99	49.93	50.64	50.55	49.99	49.93
GE (kjg ⁻¹) ⁵	18.4	18.4	18.4	18.4	18.4	18.4	18.4	18.4
P/E ⁶	69.33	69.34	69.75	69.70	69.33	69.34	69.75	69.70

¹. Date stone meal, decorticated waste seed and grinded to fine particles.

². Premix supplied according to Xie et al. (1997)

³. Marjoram (M) is a dried combination of natural attracting spices that stimulate digestibility

⁴. NFE (Nitrogen free extract) = 100 - (%moisture + %C. protein + %C. fat + %C. fiber + ash)

⁵. Gross energy (GE) = calculated as 5.621.64, 39.5 and 17.24 kjg⁻¹ of protein, fat and carbohydrate respectively according to NRC, 1993).

⁶. P/E ratio (protein to energy ratio) = mg protein /kj GE.

Table 2: amino acid composition content of basal diet and amino acid requirements of Nile tilapia (g.100 g⁻¹diet)

Indispensable amino acid	Required ¹	Diet 1 (control)
Arginine	1.6	1.52
Histidine	0.65	0.97
Isoleucine	1.18	1.32
Leucine	1.29	1.81
Lysine	1.95	1.69
Methionine	1.02	0.88
Phenylalanine	1.43	1.22
Threonine	1.43	1.37
Valine	1.06	1.39

¹ From Santiago & Lovell (1988).

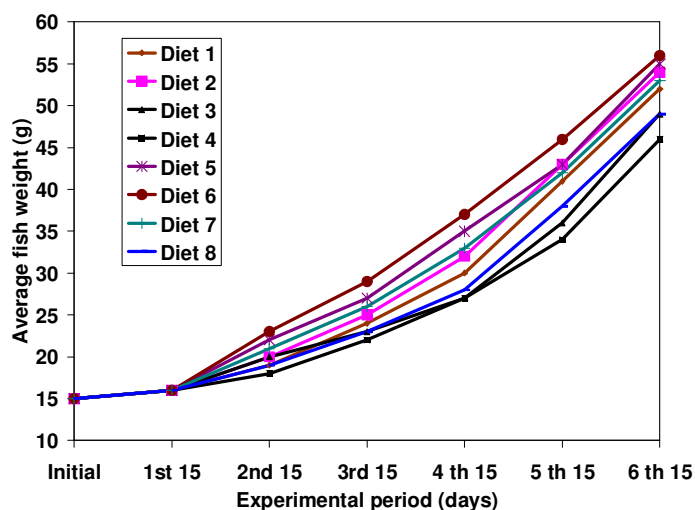


Figure 1 Effect of different levels of date stone diets supplemented with 0% and 0.03% Digestarom[®] on weight gains of Nile tilapia fingerlings.

Table (3): Growth performance and nutrient utilization of Nile tilapia fed the experimental diets
Values are mean± standard deviation. Values in the same row with same superscripts are not significantly different (P≥0.05).

Classification	Average body weight		SGR	Feed intake	FCR	PPV	PER
	(g/fish)						
	Initial	Final		(g/fish)			
Date stone levels							
0%	15.42±0.06	53.5±2.0 ^b	1.48±0.04 ^a	63.1±0.1 ^b	1.51±0.2 ^a	33.84±3.5 ^b	2.06±0.2 ^a
15%	15.42±0.05	54.7±1.1 ^a	1.50±0.03 ^a	63.2±1.5 ^a	1.51±0.2 ^a	34.59±4.5 ^a	2.05±0.2 ^a
30%	15.40±0.08	50.2±1.7 ^b	1.41±0.04 ^b	61.4±1.6 ^c	1.70±0.2 ^b	32.84±4.4 ^c	1.97±0.3 ^b
45%	15.39±0.05	47.4±1.6 ^c	1.33±0.04 ^c	62.4±1.1 ^b	1.85±0.2 ^c	29.45±3.1 ^d	1.79±0.2 ^c
Marjoram							
0%	15.4±0.6	49.5±3.0 ^b	1.40±0.1 ^b	63.0±1.2	1.84±0.13 ^b	29.27±1.8 ^b	1.78±0.1 ^b
0.03%	15.4±0.05	52.±2.9 ^a	1.47±0.1 ^a	62.1±1.4	1.53±0.11 ^a	36.09±3.4 ^a	2.17±0.1 ^a
DSxD	N.S	**	**	**	*	**	NS

Values are mean± standard deviation. Values in the same row with same superscripts are not significantly different (P≥0.05).

¹IBW=initial body weight. ²FBW=final body weight. ³. SGR, specific growth rate= (Ln FBW-Ln IBW)/90x100.

⁴. FI= feed intake ⁵ FCR, feed conversion ratio=dry feed fed/ body weight gain.

⁶ PER, protein efficiency ratio= final body weight gain/protein intake X100.

⁷.PPV%, protein productive value = {(final body

Table (4): whole body composition (% wet weight basis) of fish at the end of the experiment

Classification	Moisture	Crude protein	Ether extract	Ash	Energy (Kj/100g)
Date Stone levels (DS)					
0%	74.26±0.86 ^a	15.60±0.3 ^b	6.03±0.4 ^a	4.69±0.13 ^a	592.01±26.9
15%	74.18±0.5a ^b	15.48±0.2 ^{bc}	5.78±0.1 ^c	4.59±0.18 ^b	587.9±17.6
30%	73.7±0.2b ^c	15.70±0.2 ^a	5.91±0.03 ^{ab}	4.67±0.06 ^b	599.2±11.1
45%	73.9±0.3 ^b	15.59±0.3 ^b	5.97±0.1 ^a	4.6±0.4 ^b	601.8±3.7
Digestarom ® (D)					
0%	74.11±0.5	15.56±0.2	5.92±0.1	4.65±0.1a	596.8±11.2
0.03%	73.94±0.5	15.63±0.3	5.93±0.31	4.62±0.13b	593.7±21.6
DSXD	**	*	*	*	NS

Values are mean± standard deviation. Values in the same row with same superscripts are not significantly different (P ≥ 0.05).

Table (5): Economic information for Nile tilapia

Items	Diets							
	Without supplement D				With supplement D			
	1 (0%)	2(15%)	3(30%)	4(45%)	5(0%)	6(15%)	7(30%)	8(45%)
Food cost kg diet (LE)	2.9	2.7	2.8	2.7	2.94	2.8	2.8	2.77
No. fish stocked/ m ³	100	100	100	100	100	100	100	100
No fish harvested m ³	100	100	100	100	100	100	100	100
Harvested (kgm ⁻¹)	5.17	5.5	5.4	5.3	5.8	5.8	5.6	5.3
Food used(kg/m ⁻¹)	6.3	6.3	6.3	6.3	6.3	6.2	6.0	6.3
Fingerling cost (LE) ¹	18.27	18.2	18.1	18.0	18.5	17.4	17.0	17.5
Food cost ²	15.0	15.0	15.0	15.0	15.0	15.0	15.0	15.0
Total cost (LE)	33.27	33.3	33.2	33.0	33.5	32.4	32.0	32.5
Value of harvest (8.6 LE. kg ⁻¹)	41.4	42.2	41.2	41.0	46.4	46.4	44.8	42.4
Net profit (LE)	8.13	8.9	8.0	8.0	12.9	14.0	12.8	9.9

¹LE= Lever Egyptian, one Dollar equal 6.12 LE.

² Feed cost of 1 kg ingredients used were 6 LE for fish meal, 23.9 LE for soybean meal, 1.75 LE yellow corn meal, and 0.5 LE date stone, 12 LE for Digestarom ®, 6.5 LE for soybean oil, 5.0 LE vitamin and minerals, ingredient price at start of 2012.

Feed quality

The amino acids as well as the nutrient contents of the diets are presented in tables (1 & 2). The essential amino acid of the basal diets meets the requirements of Nile tilapia (Santiago and Lovell, 1988).

Growth performance

The changes in mean body weight (g/fish) of Nile tilapia fed on eight tested diets containing 0, 15, 30, and 45% DS instead of YC, without or with 0.03 Marjoram during the period of the experiment (3 months) are shown in Fig. 1. In the beginning of the experiment, mean weight was not significantly different among the treatments (P>0.05). The overall averages ± SD of Nile tilapia 15.4±0.5 g/fish for weight. At the end of the experiment, the mean weight ranged between 49.6±3.5 g at the highest level date stone and 55.9±2.9 g at lowest level date stone. The mean weight was significantly affected by date stone, and M supplementation rate. The overall data for final individual weight, and specific growth rate (SGR) for Nile tilapia reared at four levels of date stone and M supplementation for a period of 3 months in aquarium are presented in Table 3. It can be concluded from this table that, mean final weight (g/ fish) and SGR (%)

/day) were significantly (P < 0.05) affected and the best fish weight was obtained with the lowest level date stone and supplemented with 0.03% D.

The results of feed conversion ratio (FCR), protein productive value (PPV %) and protein efficiency ratio (PER) are presented in Table 3. Feed conversion ratio and protein productive value, protein efficiency ratio were significantly (P ≤ 0.05) affected by level date stone and exhibited the best results at the lowest level date stone and supplemented with Marjoram (M).

Feed intake was significantly higher in Nile tilapia fed on 15 % DS at 0.03% M than other DS levels. The feed conversion ratio (FCR) of Nile tilapia fed on 15 % DS at 0.03% M was not significantly different (P>0.05) when compared with control diet (Diets with 0% DS). Increasing dietary levels of DS from 15 to 45% caused decreasing feed intake and deterioration (FCR). Protein efficiency ratio (PER) , protein productive value (PPV) were significantly higher at 15% DS and supplemented with 0.03% M. The lowest PER, PPV were observed at 45 % DS without and with M.

Whole body composition

The chemical compositions of whole body parameters of Nile tilapia fingerlings fed diets containing different levels of DS supplemented without and with 0.03% M are summarized in table 4. Value of moisture and crude protein (CP%) content were highly significantly ($P<0.05$) different between all fish groups. However, ether extract (EE %) ash and energy contents were significantly not ($P>0.05$) different among all fish groups. Incorporation DS at 30 % replacement level only did significantly increase protein content.

DISCUSSION

Approximately 15% of corn meal (CM) energy could be replaced by DS, and result in growth rates in Nile tilapia higher than a CM- based diet. This is the first time to our knowledge that date stone meal has been demonstrated to be effective in replacing CM in fish diets. Based on feed intake the palatability of control diet and diets 2, 3, 5, 6 and 7 appeared to be better than the two diets 4 and 8 (Table 3). Feed intake was significantly higher ($P<0.05$) in Nile Tilapia fed diets containing 15 % DS without and with 0.03% M. The feed conversion ratio (FCR) of Nile tilapia fed DS at 15 % inclusion levels was significantly different ($P>0.05$), compared with those fed other levels of DS. Increasing dietary DS levels from 15 to 45 % caused significant deterioration in FCR ($P<0.05$). On the other hand, El-Sayed *et al.* (2006) noted that increasing dietary DP levels from 25 to 100% did not cause any further deterioration in FCR ($P>0.05$). There were significant differences in weight gain and feed conversion (FCR) was significantly greater with diets 15 %DS at 0.03% M than with the other diets. Diet 2 contained 15% CM and represented the highest level of substitution, which was significantly different from the other diets (Table 3). These results are in agreement with the results of El-Sayed *et al.* (2006) and Yousif *et al.* (1996). While Osman *et al.* (2001) found that growth performance values were significantly ($P<0.05$) higher in Nile tilapia fed diet which contain 50 % date pits treated with H_2SO_4 and NaOH, whereas fish fed diet containing 50 % untreated date pits gave the lowest values ($P>0.05$) in our experiment. Also, Belal (2008) found that growth performance of Nile tilapia fed diet containing fungi *Trichoderma reesei*-degraded date pits DDP could replace 300 g kg^{-1} of dietary corn with better growth results, when compared with those fish fed the other diets.

In the present experiment, the lowest final fish weight was obtained by fish fed on diets containing 45% DS replacing CM, without and with 0.03% M. similar results have been reported by Yousif *et al.* (1996), who found that blue tilapia (*Oreochromis aureus*) fed date pits as carbohydrate source without growth enhancing supplement had significantly poor performance. The poor performance of tilapia fed DS may have been due to their high contents of simple sugars (Yousif *et al.*, 1996), whereas tilapia is known to utilize complex sugars more efficiently than simple sugars. In support, Shiau and Lin (1993) studied the effects of starch or fructose on the growth and feed

efficiency of (*Oreochromis niloticus* x *Oreochromis aureus*) hybrids. They found that starch was better utilized than glucose. This result is also applied to specific growth rate (SGR). The results showed that diets containing 15 % DS without or with 0.03% M as feed additives was significantly different from control diets (1) in its effect on fish performances. The present results are in agreement with the finding of Belal and Al-Owafeir (2009) who found that growth performance, including SGR of Nile tilapia fed the control diet are similar to fish fed date pits fed at 15 and 30 levels. Similar results have been reported for probiotics use in diets for tilapia fingerling by Khattab *et al.* (2004); Salem (2008); Eid and Mohamed (2007); Mohamed *et al.* (2007), El-Dakar *et al.* (2007), Carnevali *et al.* (2006) and Gaber *et al.* (2012).

Increasing dietary DS levels from 15 to 45 % in the experimental diets with other additives increased significantly ($P<0.05$) protein efficiency ratio (PER) and protein productive value (PPV%) about control diet (2, 3,4 and 5 vs diet 1). Similar results have been reported by El-Sayed *et al.* (2006) who noted that PER ,PPV% and EU% were significantly affected ($P<0.05$) by increasing date pits levels in Nile tilapia diets. Also significant ($P<0.05$) improvement was observed with M addition in FCR, PER and PPV within each replacement category. Protein efficiency ratio (PER) and protein productive value (PPV %) were significantly higher on the replacement rate of 15 % DS and 0.03 % M. The lowest protein productive value (PPV %) (30.7 %) was observed with diet (1) as opposed by Osman *et al.* (2001). These results are in agreement with the results of Mehrim (2001); Diab *et al.* (2002); Khattab *et al.* (2004); Mohamed *et al.* (2007), Eid and Mohamed (2008) and Gaber *et al.* (2012) for tilapia. The present results confirm those obtained by Azaza *et al.* (2008) in their work on waste date meal fed to Nile tilapia and Abd El-Maksoud *et al.* (1999) on tilapia fed diets supplemented with Marjoram leaves.

Calculations of economical efficiency of the tested diets based on cost one kg gain in weight of Nile tilapia in comparison with the control group are shown in Table 5. Feed costs and fingerlings cost m^{-1} (L.E) were the highest for the control diet (33.5 L.E) and gradually decreased with the increasing levels of DS and with addition of the M. At the level of DS replacement (15%) with 0.03% M , it was found that Nile tilapia could be produced cheaper than fish fed on the control diet. These results may indicate that the costs of one kg of diet declined by the incorporation of non conventional energy sources supplemented with 0.03% M due to its low price and this is in agreement with El-Sayed *et al.* (2006), Belal and AL-Owafeir (2009) and Gaber *et al.* (2012). From the previous results, it could be concluded that the diet containing 15% DS with 0.03 % Marjoram as dietary energy sources in formulation of Nile tilapia diets instead of 15 % yellow corn have positive improvement effects on Nile tilapia performance. From feed utilization data and from the economical point of view the diet containing 15% DS and supplemented with 0.03% M could be recommended as feed for Nile tilapia fingerlings.

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