DOI: 10.4025/actascianimsci.v31i4.6929

Dietary α-tocopheryl acetate on fillet quality of tilapia

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ABSTRACT. This work compared the effects of dietary α-tocopheryl acetate on storage life of tilapia fillets. Three experimental diets containing increasing α-tocopherol levels (zero, 100, and 200 mg kg^{-1}) were used. The fish, with a mean initial weight of 184.23 ± 1.68 g, were fed diets for 63 days. After that period, they were slaughtered, filleted, and the fillets were grounded to accelerate lipid oxidation. Fish growth, survival, fillet yield, chemical composition and lipid oxidation of tilapia ground fillets were evaluated 0, 30 and 60 days after frozen storage. The results demonstrated that there was no significant difference between treatments for performance, and also, tocopherol did not influence the chemical composition values of fillets. Increased tocopherol levels in the feeds promoted a reduction in ground fillets lipid oxidation values.

Key words: lipid oxidation, *Oreochromis niloticus,* storage life, tocopherol supplementation.

RESUMO. Suplementação dietética de acetato de α-tocoferil na qualidade dos filés de tilápia. Este trabalho comparou os efeitos da suplementação dietética do acetato de α-tocoferil na vida de prateleira dos filés de tilápia. Três dietas experimentais contendo níveis crescentes de α-tocoferol (zero, 100 e 200 mg kg^{-1}) foram usadas. Os peixes, com peso médio inicial de 184,23 ± 1,68 g foram alimentados com as dietas durante 63 dias. Após esse período, os peixes foram abatidos, filetados, sendo os filés moídos para acelerar a oxidação lipídica. O crescimento, sobrevivência, rendimento de filé, composição química e oxidação lipídica dos filés de tilápia moídos foram avaliados 0, 30 e 60 dias após a estocagem congelada. Os resultados demonstraram que não houve diferença significativa entre os tratamentos para desempenho, e também, que o tocoferol não influenciou na composição química dos filés. O aumento nos níveis de tocoferol nas dietas promoveu a redução dos valores de oxidação nos filés moídos.

Palavras-chave: oxidação lipídica, *Oreochromis niloticus*, vida de prateleira, suplementação de tocoferol.

Introduction

Vitamin E is a generic descriptor attributed to a group of lipid-soluble, structure-related compounds, occurring naturally in α-, β-, γ-, or δ-tocopherols, and the four corresponding tocotrienols. Among them, α-tocopherol has the highest vitamin E activity (NRC, 1993). Furthermore, α-tocopherol is a potent biological antioxidant that protects biomembranes (WANG et al., 2006) and lipid components containing unsaturated fatty acids against attack from oxygen free radicals (YAMAMOTO et al., 2001).

Lipid oxidation is a serious problem for biological materials containing unsaturated fatty acids, a fact that is particularly important for fishes since they normally contain a greater amount of highly unsaturated fatty acids (HUFA) than other species of animals (HUANG et al., 2004). Lipid oxidation in fish products can contribute to the development of a rancid flavor and changes in color, texture, and nutritional value (JENSEN et al., 1998), mainly affecting their acceptability for consumption (NOGALA-KALUCKA et al., 2005).

Dietary supplementation with α-tocopherol has been the most commonly used form of vitamin E incorporation (BOTSOGLOU et al., 2003). Studies have shown that the fish requirement for tocopherol may vary due to many factors, including the interrelations of this vitamin with other factors such as the amount of polyunsaturated fatty acids (SHIAU; SHIAU, 2001; LÓPEZ-BOTE et al., 2003), the oxidative stability of the diets (HUANG; HUANG, 2004) and the dietary presence of other antioxidant substances including, for example, vitamin C (GUO et al., 2001; SHIAU; HZU, 2002).

Recently, authors have tested the effects of supplementation with different levels of vitamin E in diets on growth, lipid stability (FOGAÇA;

SANT'ANA, 2007), inflammatory response (MARTINS et al., 2008), as an immunostimulant (CHEN et al., 2004) in several fish species. The essentiality of dietary α-tocopherol for normal tilapia growth was clearly demonstrated in a study by Shiau and Shiau (2001). Those authors observed higher weight gain, with reduced values for fish given the basal diet (without tocopherol supplementation) with incremental increases in dietary vitamin E level up to the requirement.

Improved product quality has been achieved by feeding on supra-nutritional levels of α-tocopheryl acetate before slaughter (RUFF et al., 2002b). This has been demonstrated by Ruff et al. (2003), who suggested supplementation of feeds with at least 550 mg α-tocopheryl acetate kg^{-1} diet for market size turbot two months prior to slaughter improved fillet quality. Others studies on the Atlantic salmon *Salmo salar* (ONIBI et al., 1996; HAMRE et al., 2004), sea bass *Dicentrarchus labrax* (GATTA et al., 2000; PIRINI et al., 2000) and hybrid tilapia *Oreochromis niloticus x O. aureus* (SHIAU; HZU, 2002) also demonstrated that when dietary vitamin E level increased, lipid peroxidation in *post-mortem* muscle was inhibited.

Thus the objective of this work was to evaluate the effects of increased α-tocopherol supplementation in diets (zero, 100, and 200 mg α-tocopheryl acetate kg^{-1} feed) on the lipid stability of Nile tilapia.

Material and methods

Feeding trial and diet

The feeding trial was developed at the Aquaculture Center (Universidade Estadual Paulista, Unesp, Caunesp, Jaboticabal, São Paulo State, Brazil). Juveniles of tilapia were obtained from the Aquatic Organisms Nutrition Laboratory (at Caunesp) and acclimated to pond conditions for two weeks.

Four-hundred and twenty sex-reversed Nile tilapia juveniles, with a mean initial weight of 184.23 ± 1.68 g, were randomly distributed among twelve earthen fishponds of 50 m^2 area (5 m of width for 10 m of length) and 1 m depth, to each of which water was supplied at a rate of approximately 11 L $min.^{-1}$ in a flow-through system at a stocking density of 0.70 fish m^{-2}. The water-quality physicochemical parameter means during the experimental period were: dissolved oxygen 6.13 ± 0.95 mg L^{-1}, temperature of 28.05 ± 1.55°C in the morning and 29.06 ± 1.67°C in the afternoon, and pH 7.20 ± 0.28.

The diet (Table 1) was prepared as 4 mm pellets by cooking-extrusion using a semi-industrial extruder (Caunesp, Jaboticabal, São Paulo State, Brazil). The α-tocopheryl acetate (Rovimix E-50 adsorbate, Roche commercial product) was diluted in soybean oil and added after extrusion. Three experimental diets containing increasing α-tocopheryl acetate levels (zero, 100, and 200 mg kg^{-1}) were used. The quantities of α-tocopherol determined in the diets were 24.45, 135.98, and 230.74 mg kg^{-1} feed. The feed were frozen storage at -18°C and samples for analysis of α-tocopherol were collected three times during the experimental period (with zero, 30 and 60 days of frozen storage).

Table 1. Tocopherol concentrations and proximate compositions of the experimental diets.
Tabela 1. *Concentração de tocoferol e composição proximal das dietas experimentais.*

Ingredients (%) *Ingredientes (%)*	Diet 1 *Dieta 1*	Diet 2 *Dieta 2*	Diet 3 *Dieta 3*
Corn *Milho*	34.24	34.24	34.24
Wheat bran *Farelo de trigo*	14.58	14.58	14.58
Rice bran *Farelo de arroz*	10.00	10.00	10.00
Soybean meal *Farinha de soja*	28.41	28.41	28.41
Fish meal *Farinha de peixe*	11.70	11.70	11.70
Soybean oil *Óleo de soja*	1.50	1.50	1.50
DL-Methionine *DL-metionina*	0.38	0.38	0.38
L-lysine *L-lisina*	0.19	0.19	0.19
Vitamin supplement (without vitamin E) *Suplemento vitamínico (sem vitamina E)*	0.25	0.25	0.25
Mineral supplement *Suplemento mineral*	0.25	0.25	0.25
Proximate composition			
D.M. (%)[1] *M.S.*	90.24	89.87	89.85
DE (kcal kg^{-1})[2] *ED*	3,214	3,214	3,214
CP (%) *PB*	26.38	25.90	25.55
Fat (%) *Lipídios*	3.27	3.18	3.41
Ash (%) *Cinzas*	7.77	7.71	7.72
TNC (%)[b] *CNE*	36.49	38.16	38.79
α-tocopherol (mg kg^{-1}) *α-tocoferol*	24.45	135.98	230.74

[1]Dry matter. [2]Digestible energy. DE calculated based on DE values for each ingredient. [3]Rude protein. [4]Total non-structural carbohydrate; TNC% = DM% - (CP% + Fat% + Crude fiber% + Ash%).
[1]Matéria seca. [2]Energia digestível. ED calculada com base nos valores de ED de cada ingrediente. [3]Proteína bruta. [4]Carboidratos não-estruturais; CNE% = MS% - (PB% + lipídios% + fibra bruta% + cinzas%).

Fish were hand fed twice a day (9:00 and 17:00) to apparent satiation. Pellets were distributed slowly, permitting all fish to eat. The fishes received food for 63 days. All fish were individually weighed once every two weeks. Mortality and feed intake were recorded daily.

At the end of the trial, the fish were slaughtered by thermal shock in boxes containing ice and water (1:1) and then eviscerated and manually filleted. The fillets were ground in order to accelerate lipid

oxidation due to the incorporation of oxygen into the muscle tissue. The samples were frozen at -18°C for 60 days, and the analyses realized with zero, 30 and 60 days of frozen storage.

The following performance indices were determined for each treatment (n = 140):

Feed conversion ratio (FCR) = feed intake (g)/ weight gains (g);

Mean weight gain (WG) = (Final weight – Initial weight) x 100/Initial weight;

Apparent feed intake (AFI) = (Mean feed intake per treatment)/(Mean number of fish);

Specific growth rate (SGR) = (ln final weight – ln initial weight) × 100/Time;

Survival rate (S) = (initial number of fish/final number of fish)*100;

Fillet yield (FY) = (Fillet weight × 100)/Total Weight.

A fillet yield evaluation was made for samples of ten fishes from each plot.

Analysis

Proximate analysis was performed on feed and fillets according to standard analytical methods (AOAC, 2000). All samples were stored at -18°C and thawed at 5°C for 24h prior to analysis. Twenty grams of feed from each treatment was ground prior to analysis and four ground fillets samples were taken from each treatment, per time interval, and analysis were performed with three replicates. Moisture was determined on approximately 5 g of ground fillet, by oven drying at 105°C for 24h, following technique 950.46 (AOAC, 2000). Results were means of three determinations and were expressed as grams of water 100 g^{-1} of fillet. Crude lipid concentration was determined by petroleum ether extraction in a Soxhlet device (AOAC method 960.39; AOAC, 2000). Crude protein was determined according to the Kjeldahl method, following AOAC 928.08 (AOAC, 2000). The nitrogen-to-protein conversion factor considered was 6.25. Ash was isolated by incineration at 550°C for 6h in a muffle furnace (Quimis Inc., São Paulo State, Brazil) (AOAC method 900.02; AOAC 2000).

High Performance Liquid Chromatography (HPLC) determined α-tocopherol in the feed, according to the methodology described by Mestre Prates et al. (2006). Lipid oxidation was evaluated by the formation of Thiobarbituric Acid Reactive Substances (TBARS), according to Vyncke (1970). TBARS was performed on the ground fillets on day 0 (5 days post-harvest in -18°C storage), day 30 and at week 8 (in -18°C storage). Ten grams of the fillet was used for the TBARS test. Frozen fillets were thawed at 5°C in a refrigerator overnight. Five mL of the distillate was used for color development, and the color was measured at 532 nm with a spectrophotometer. Results were expressed as mg of malonaldehyde per kg of fillets. The oxidation quantification was calculated by the standard curve: $y = 0.1152x$ ($r^2 = 0.996$).

On each sampling occasion, four samples were selected from each treatment batch to be subjected to the different analyses (proximate composition and TBARS). All measurements were carried out in triplicate. The data were analyzed by Statistical Analyses System – SAS, version 6.12 software package (SAS Institute Inc., Cary, NC, USA). The main effects of dietary treatment and storage time on lipid oxidation, as well as interactions between dietary treatment and storage time on lipid oxidation and proximate composition were determined by Tukey-Kramer test, and the significance was defined either at $p < 0.05$.

Results and discussion

Results

No difference in growth was observed between groups (Table 2). After nine weeks of feeding, the fish had reached a mean weight of 460.1 g on average. Weight gain (WG) of the fish fed diets containing zero mg α-tocopheryl acetate kg^{-1} was not significantly different ($p < 0.05$) from those fed 200 mg kg^{-1} of the supplement. Feed conversion ratio (FCR) and specific growth rate (SGR) followed similar trends as growth performance. Survival (S) of the fish fed diets containing zero, 100 and 200 mg of α-tocopheryl acetate kg^{-1} showed no significant difference ($p < 0.05$).

Table 2. Performance parameters of Nile tilapia fed diets containing increased tocopherol levels.
Tabela 2. *Parâmetros de desempenho de tilápias do Nilo alimentadas com dietas contendo níveis crescentes de tocoferol.*

Dietary tocopherol[1] *Tocoferol dietético*	FW (g) *PF*	WG (%)[2] *GP*	AFI (g)[3] *CRA*	FCR[4] *CAA*	SGR[5] *TCE*	FY (%)[6] *RF*	S (%)[7] *S*
F Value *F valor*	0.88	0.88	0.17	0.79	0.86	8.15*	0.24
0 mg	484.70 (5.41)	162.76 (3.35)	480.18 (0.92)	1.60 (0.09)	1.54 (0.63)	32.74 (1.29)[a]	96.43 (3.59)
100 mg	505.72 (15.61)	176.63 (10.25)	486.34 (7.08)	1.48 (0.06)	1.60 (0.56)	31.70 (0.26)[ab]	96.43 (3.59)
200 mg	479.90 (10.21)	174.78 (6.50)	471.25 (8.00)	1.50 (0.09)	1.49 (0.49)	29.88 (1.56)[b]	87.86 (22.42)

Values are mean (SD) of quadruplicate groups of 35 fishes (n = 4). Means in the same column with different superscripts are significantly different at *$p < 0.05$. Final weight (FW), weight gain (WG), apparent feed intake (AFI), Feed conversion ratio (FCE), specific growth rate (SGR), fillet yield (FY), and survival (S). [1]mg kg^{-1} feed. [2]WG = (Final weight - Initial weight) × 100/Initial weight. [3]AFI = (Mean feed intake per treatment)/Mean number of fish. [4]FCR = Mean intake/mean weight gain. [5]SGR = (ln final weight – ln initial weight) × 100/Time. [6]FY = (Fillet weight × 100)/Total Weight. [7]S = (initial no. of fish/final no. of fish)*100.

*Valores médios (desvio padrão) de quatro parcelas de 35 peixes (n = 4). Médias seguidas de letras diferentes nas colunas diferem entre si pelo teste de Tukey ($p < 0,05$). Peso final (PF), ganho de peso (GP), consumo de ração aparente (CRA), conversão alimentar aparente (CAA), taxa de crescimento específico (TCE), rendimento de filé (RF) e sobrevivência (S). [1]Níveis expressos em mg kg^{-1} de ração. [2]GP = (Peso final - Peso inicial) x 100/Peso inicial. [3]CRA = (Consumo médio de ração por tratamento)/Número médio de peixes. [4]CAA = Consumo médio/ganho médio de peso. [5]TCE = (ln peso final – ln peso inicial) x 100/Tempo. [6]RF = (Peso do filé x 100)/Peso Total. [7]S = (nº inicial de peixes/nº final de peixes)*100.*

Processing yield (FY) was significantly lower for fish fed diets containing 200 mg α-tocopheryl acetate kg^{-1} when compared with the control group. Fishes fed diets containing 100 mg α-tocopheryl acetate kg^{-1} showed intermediate values of fillet yield.

Dietary α-tocopheryl acetate supplementation affected fillets composition of tilapia (Table 3). Moisture and ash were significantly different ($p < 0.05$) among the groups. No statistical difference ($p > 0.05$) was observed for crude protein or crude lipid (Table 3). Crude protein ranged between 19.97 and 20.67%, and crude lipid averaged 2.3%. Fillet composition of the group fed diets containing 100 mg α-tocopheryl acetate kg^{-1} showed lower ash (2.21%) and higher moisture contents (77.76%).

Table 3. Tilapia fillet proximate composition after 9 weeks of feeding the experimental diets.
Tabela 3. *Composição proximal dos files de tilápia após 9 semanas de alimentação com as dietas experimentais.*

Dietary tocopherol (mg kg^{-1}) *Tocoferol dietético*	Moisture (%) *Umidade*	Crude Fat (%) *Lipídios*	Crude Protein (%) *Proteína Bruta*	Ash (%) *Cinzas*
0	76.12(1.75) [b]	0.63(0.01) [a]	20.67(1.27) [a]	2.33(0.07) [ab]
100	77.96(0.60) [a]	0.56(0.03) [a]	19.97(0.69) [a]	2.21(0.08) [b]
200	75.04(1.09) [b]	0.64(0.02) [a]	20.13(0.74) [a]	2.46(0.15) [a]

Values are mean (SD) of triplicate groups of four samples (n = 4). Means in the same column with different superscripts are significantly different at *p < 0.05.
Médias seguidas de mesmas letras não diferem pelo teste de Tukey a 5% de probabilidade. Análises de quatro amostras realizadas em triplicata (n = 4).

Dietary α-tocopherol level influenced lipid oxidation, as demonstrated by TBARS results (Table 4). TBARS produced for fish fed 0 mg α-tocopheryl acetate kg^{-1} diet were significantly higher than other fish ($p < 0.05$).

Table 4. Mean TBARS values (mg kg^{-1} fillet) for increase tocopherol supplementation levels.
Tabela 4. *Médias de valores de SRATB (mg kg^{-1} filé) segundo os níveis de suplementação de tocoferol.*

	TBARS (mg MDA kg^{-1}) *SRATB (mg MDA kg^{-1})*	
Storage time *Tempo de estocagem*	0 day *0 dias*	30 days *30 dias*
Level of tocopherol kg^{-1} diet *Nível de tocoferol kg^{-1} dieta*		
0 mg	2.78(0.50) [b]	5.16(0.57) [c]
100 mg	1.26(0.10) [a]	3.20(0.30) [b]
200 mg	0.94(0.24) [a]	1.63(0.17) [a]

Values are mean ± SD of triplicate groups of four samples (n = 4). Means in the same column with different superscripts are significantly different at *p < 0.05.
Médias seguidas de mesmas letras não diferem pelo teste de Tukey a 5% de probabilidade. Análises de quatro amostras realizadas em triplicata (n = 4).

The linear relationship between dietary α-tocopherol level and TBARS values was significant ($p < 0.05$) in fillets stored frozen for 30 and 60 days. Fillets TBARS values increased from time zero to time 30 (days) and decreased from time 30 to time 60 (days) in -18°C storage. In samples taken during the period between 30 and 60 days of frozen storage, fillet TBARS values for treatment with 200 mg α-tocopheryl acetate kg^{-1} diet was significantly lower than for other groups ($p < 0.05$).

Discussion

The same weight gain was observed between control and the group receiving 100 mg α-tocopheryl acetate kg^{-1}, which is the amount representing the tilapia requirements, according to the National Research Council (NRC, 1993) (50-100 mg of vitamin E kg^{-1} diet). These results are in agreement with Huang et al. (2004), who evaluated an *O. niloticus* × *O. aureus* tilapia hybrid that received supplementation feeds containing zero, 50 and 100 mg vitamin E kg^{-1} and did not observe growth differences among treatments. Huang and Huang (2004) compared the supplementation of two levels of tocopherol in diets (11 and 105 mg kg^{-1}) for transgenic-GH Coho salmon (*Oncorhynchus kisutch*) and did not observe statistical differences between these treatments for a 70-day experimental period.

Comparisons of the effects of dietary treatments among growth studies are often difficult due to many variables that affect growth (CHAIYAPECHARA et al., 2003). In this work, the fishes were held in earthen ponds, where additional vitamin could be consumed, and no effect is expected of vitamin E on growth under these conditions. Similar results were obtained in Atlantic halibut *Hippoglossus hippoglossus* (RUFF et al., 2002a), Atlantic salmon (LYGREN et al., 2000; SCAIFE et al., 2000), and rainbow trout *Oncorhynchus mykiss* (KIRON et al., 2004). Tocher et al. (2002) also observed no differences of performance with α-tocopheryl acetate supranutritional supplementation for turbot (*Scophthalmus maximus* L.) and sea bream (*Sparus aurata* L.)

Fillet yield (FY) was significantly higher ($p < 0.05$) for fish fed diets without supplementation than for fish fed diets containing 200 mg of α-tocopheryl acetate kg^{-1} (Table 2), but it is also in agreement with values found in the literature because fillet yield depends not only on the filleter's skill, but also especially on the anatomical shape of the fish (CONTRERAS-GUZMÁN, 1994). Nile tilapia fillet yield data (skinless) can be found in the literature ranging from 25.4% to values near 42%, owing to body weight and filleting methods, with comparisons for forms of decapitation, and skin and fin removal (MACEDO-VIEGAS; SOUZA, 2004).

More recent studies have determined yields ranging from 32.0 to 33.4% (GARDUNO-LUGO et al., 2003) and from 34.4 to 38.0% (RUTTEN et al., 2004; 2005) in tilapias with mean weights of

450 and 700 g, respectively. Then, more works must be conduced to defined standard fillet yield for Nile tilapia.

The results on the composition of tilapia fillets were as expected (Table 3) and very close to those reported by Huang et al. (2004) for juvenile hybrid tilapia *O. niloticus* × *O. aureus* (78.40, 79.10, and 79.00% moisture; 1.32, 1.31, and 1.25% ash; and 16.40, 18.30, and 18.90% crude protein, for diets containing 20, 108, and 206 mg α-tocopheryl acetate kg^{-1}, respectively). Huang and Huang (2004) observed lower values of moisture (74.60, 74.90, and 74.60%), ash (1.56, 1.53, and 1.52%); and crude protein (16.10, 16.20, and 16.40%) for diets containing zero, 120, and 200 mg α-tocopheryl acetate kg^{-1}, respectively; when compared with the results shown in Table 3.

It is known that variations in the chemical composition of fishes are closely related to nutrition, live area, fish size, catching season, and seasonal and sexual variations as well as other environmental conditions (ERKAN; ÔZDEN, 2006). It is unclear how the dietary α-tocopheryl acetate supplementation affected fillet composition of tilapia in our experiment because no differences were observed in similar papers that evaluated vitamin E supplementation in feeds with regard to the chemical composition of tilapia (HUANG; HUANG, 2004), sea bream (PIRINI et al., 2000), and trout *Oncorhynchus mykiss* (CHAIYAPECHARA et al., 2003). Therefore, the variations in composition found in the present work between treatments were minimal and did not affect the nutritional value of tilapia fillet.

Vitamin E is a potent and widely studied antioxidant in biological systems (YAMAMOTO et al., 2001). The principal antioxidant role of vitamin E is to neutralize free radicals that could initiate a chain reaction, particularly among unsaturated fatty acids in membranes (BURTON; TRABER, 1990). When the animal is slaughtered, the rate and extent of oxidation are influenced by pre-slaughter events (stress), and also by post-slaughter factors, such as techniques used during slaughter, *post mortem* decline in pH, and carcass temperature (SANT'ANA; MANCINI-FILHO, 1995).

In the present work, the group that received 200 mg of α-tocopheryl acetate kg^{-1}, double the tilapia requirement, showed lower lipid oxidation ($p < 0.05$) compared to other treatments with 30 days of frozen storage (Table 5), since lipid oxidation decreases in muscle tissues when dietary α-tocopheryl acetate levels are increased. Research has shown consistently that vitamin E supplementation causes accumulation of α-tocopherol in muscle tissue, resulting in improved lipid stability (MITSUMOTO et al., 1993). Huang et al. (2004) found TBARS values of 36.3 and 23.5 nmol MDA kg^{-1} (which corresponds to 2.62 and 1.69 mg MDA kg^{-1}) in Coho salmon fillets for diets containing zero and 100 mg vitamin E kg^{-1}. Huang and Huang (2004), working with *Oreochromis niloticus* × *O. aureus* tilapia hybrids, observed values of 2.16, 1.87, and 1.77 nmol MDA kg^{-1} fillet (which corresponds to 0.16, 0.13, and 0.12 mg MDA kg^{-1} fillet), for diets containing zero, 120, and 200 mg vitamin E kg^{-1}, respectively. Gatta et al. (2000) investigated the quality of fresh sea bass fillets when the fish were fed three diets containing 139, 254, and 493 mg vitamin E kg^{-1}, and observed that the lipid oxidation values analyzed by the TBARS method were statistically different only for the group with lower supplementation than the others.

The addition of tocopherol to the diet may influence the oxidation rates of the processed products, since the increase in the level of α-tocopheryl acetate in the diets provided lower lipid oxidation in the ground fillets. Levels above 200 mg vitamin E kg^{-1} feed resulted in better lipid stability, and may extend the quality of *post mortem* fillets during frozen storage.

Further studies on supplementing the diet with high concentrations of antioxidants such as α-tocopherol are warranted. Furthermore, tocopherol content determination must be performed on fillets, to give the exact concentrations of tocopherol present in fillets regardless of the loss of tocopherol *in vivo* during the feeding trial.

Also, determining TBA on the diets, hematocrit, erythrocyte and leukocyte number as well as liver histology of fish would have been helpful in evaluating the vitamin E status of the diets and fish. By doing so, the efficacy of endogenous antioxidant could be better valuated.

Conclusion

The supplementation was effective in preventing lipid oxidation, thus maintaining the stability of Nile tilapia ground fillets.

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Received on April 24, 2009.
Accepted on September 11, 2009.