Arch. Pol. Fish. (2016) 23: 47-52
DOI 10.1515/aopf-2016-0005

DE GRUYTER OPEN

RESEARCH ARTICLE

Growth performance and feed utilization of keureling (*Tor tambra*) fingerlings fed a formulated diet with different doses of vitamin E (alpha-tocopherol)

Zainal A. Muchlisin, Ayu A. Arisa, Abdullah A. Muhammadar, Nur Fadli, Iko I. Arisa, Mohd N. Siti-Azizah

Received – 22 June 2015/Accepted – 20 January 2016. Published online: 31 March 2016; ©Inland Fisheries Institute in Olsztyn, Poland
Citation: Muchlisin Z.A., Arisa A.A., Muhammadar A.A., Fadli N., Arisa I.I., Siti-Azizah M.N. 2016 – Growth performance and feed utilization of keureling (*Tor tambra*) fingerlings fed a formulated diet with different doses of vitamin E (alpha-tocopherol) – Arch. Pol. Fish. 24: 47-52

Abstract. The objective of the present study was to determine the optimum dosage of vitamin E (alpha-tocopherol) in the diet of keureling, *Tor tambra* (Val.) fingerlings for optimal growth performance and feed utilization. Five doses of vitamin E were tested: 0 mg kg^{-1} feed (control); 150 mg kg^{-1} feed; 300 mg kg^{-1} feed; 450 mg kg^{-1} feed; 600 mg kg^{-1}. The feed ratio was 5% body weight, which was delivered twice daily at 08:00 and 17:00 for 60 days. The results showed that higher growth performance, feeding conversion ratios, feed efficiency, protein retention, and protein digestibility were obtained at 600 mg kg^{-1} feed, but the value was not significantly different from the other doses. The optimal dose in terms of the hepatosomatic index and survival rate was 300 mg kg^{-1}. Hence, it was concluded that the optimum, most economical dose of vitamin E supplement for keureling (*T. tambra*) was 150 mg kg^{-1} feed, because this value was not significantly different from the doses of 300 and 600 mg kg^{-1} feed.

Keywords: alpha-tocopherol, feed efficiency, feed conversion, semah, tambra

Z.A. Muchlisin [✉], A.A. Arisa, A.A. Muhammadar, I.I. Arisa
Department of Aquaculture, Faculty of Marine and Fisheries, Syiah Kuala University, Banda Aceh 23111, Indonesia
e-mail: muchlisinza@unsyiah.ac.id

N. Fadli
Department of Aquaculture, Faculty of Marine and Fisheries
Syiah Kuala University, Banda Aceh 23111, Indonesia

M.N. Siti-Azizah
School of Biological Sciences, Universiti Sains Malaysia
Penang, Malaysia

M.N. Siti-Azizah
Centre for Marine and Coastal Studies, Universiti Sains Malaysia
Penang, Malaysia

Introduction

Aceh Province, Indonesia has an abundance of fishery resources with at least 114 species of freshwater and brackish-water fishes recorded in this region (Muchlisin and Siti-Azizah 2009). In addition, Muchlisin et al. (2015a) report that 73 species freshwater and brackish-water fishes inhabit the Tripa peat swamp forest, while Rudi et al. (2009) and Rudi and Muchsin (2011) report that at least 97 species of coral reef fishes occur in Aceh waters. Furthermore, Muchlisin (2013) reports that 14 freshwater species of high economic fish value are found in Aceh waters, including *Tor tambra* (Val.), which is known locally as keureling.

Presently, keureling is the main fish targeted by most inland fishermen in Aceh Province and other parts of Indonesia because it can obtain prices of up to 35 USD kg^{-1} at local markets (Muchlisin et al.

Table 1
Ingredient formulations (g kg^{-1}) of the experimental diets containing 30% crude protein used in the study

	Dietary vitamin E level				
Raw materials (g kg^{-1})	A (0 mg kg^{-1})	B (150 mg kg^{-1})	C (300 mg kg^{-1})	D (450 mg kg^{-1})	E (600 mg kg^{-1})
Soybean meal	250	250	250	250	250
Corn flour	54	54	54	54	54
Fine rice bran	270	270	270	270	270
Fish meal	190	190	190	190	190
Cassava meal	20	20	19.85	19.70	19.40
Ebi shrimp meal	190	190	190	190	190
Fish oil	20	20	20	20	20
Vitamin E	0	0.15	0.30	0.45	0.60
Mixed vitamins*	3	3	3	3	3
Mixed minerals**	3	3	3	3	3
Total (g)	1000	1000	1000	1000	1000
Proximate composition of dry weight of diet					
Crude protein (%)	30.1	30.1	30.2	30.2	30.4
Crude lipid (%)	8.8	8.6	8.6	8.7	8.6
Crude fiber (%)	2.8	2.9	2.6	2.7	2.7
Ash (%)	5.1	5.1	4.8	4.8	4.6

*kg^{-1} of vitamin mix (kg^{-1}) : Ascorbic acid (45.0 g), Inositol (5.0 g), Choline Chloride (75.0 g), Niacin (4.5 g), Riboflavin(1.0 g), Pyridoxine HCL (1.0 g), Thiamine mono (0.92 g), D-calcium pantothenate (3.0 g), Vitamin A (1.8 m.I.u), Vitamin D3 (3.320 m.I.u), Menadione (1.670g), D-Biotin (20.0 mg), Folic acid (90.0 mg), Vitamin B12 (1.350 mg), Cellulose (Q.S to 1000 mg)
**kg^{-1} of mineral mix: Calcium phosphate, 379.65 g; Calcium lactate, 327 g; Ferrous sulphate, 25 g; Magnesium sulphate, 137 g; Potassium chloride, 50 g; Sodium chloride, 60 g; Potassium iodide, 0.15 g; Copper sulphate, 0.785; Manganese oxide, 0.8 g; Cobalt carbonate, 0.1 g; Zinc oxide, 1.5 g; Sodium selenite, 0.02 g

2015b). Since demand for this species is met by wild catches, resources are rapidly decreasing. According to Kottelat et al. (1993), the genus *Tor* as a whole is threatened by ecological perturbation and overfishing; therefore, the development of keureling cultures to meet market demand for this species should be prioritized to conserve wild populations. Cultures of keureling have already been initiated in Aceh Province. However, their growth rates and feed protein digestibility were found to be unsatisfactory in captivity (Muchlisin, unpublished data).

In this study we evaluated the role of vitamin E (alpha-tocopherol) on the growth performance, feed conversion ratio, and protein digestibility of keureling (*T. tambra*) fingerlings. In general, vitamin E plays an important role as an intracellular antioxidant, in the peroxidation reaction of the unsaturated fatty acids of biomembranes, and in the respiratory function of muscle cells (Linder 1992). Vitamin E contains alpha-tocoferol, which is important in fish gonad development (Halver 1989), it acts as an anti stressor (Li et al. 2008), and it is also involved in the immune system (Watanabe et al. 1991, Clerton et al. 2001). Nekoubin et al. (2012) and Li et al. (2014) reported that dietary vitamin E showed significant effects on the growth performance and antioxidant status of angelfish, *Pterophyllum scalare* (Schultze), and grass carp, *Ctenopharyngodon idella* (Val.), respectively. To date, no information is available on the vitamin E requirements for keureling (*T. tambra*) fingerlings; hence, the present study is important to address this problem.

Table 2
Growth performance, survival rate, hepatosomatic index, feed efficiency, feed conversion ratio, protein retention on the carcass, and protein digestibility of keureling (*T. tambra*) fingerlings according to dose levels of vitamin E in the diet

	Doses of Vitamin E (mg kg^{-1})				
Parameters	0	150	300	450	600
Total body weight gain (g)	0.35 ± 0.12^{a}	0.49 ± 0.20^{a}	0.33 ± 0.32^{a}	0.54 ± 0.17^{a}	0.54 ± 0.12^{a}
Specific growth rate (% day^{-1})	0.30 ± 0.20^{a}	$0.33 \pm 0.0.75^{a}$	0.32 ± 0.64^{a}	0.35 ± 0.11^{a}	0.36 ± 0.13^{a}
Daily growth rate (g day^{-1})	0.007 ± 0.002^{a}	0.010 ± 0.002^{a}	0.007 ± 0.005^{a}	0.011 ± 0.003^{a}	0.011 ± 0.002^{a}
Hepatosomatic index (%)	31.23 ± 2.24^{a}	32.53 ± 3.50^{a}	41.52 ± 8.72^{a}	36.68 ± 4.67^{a}	39.56 ± 5.12^{a}
Survival rate (%)	54.76 ± 4.12^{a}	54.17 ± 7.21^{a}	64.29 ± 12.37^{a}	$56.55 \pm 6,27^{a}$	55.56 ± 9.62^{a}
Feed efficiency (%)	53.36 ± 37.10^{a}	53.79 ± 9.49^{a}	54.44 ± 9.49^{a}	59.41 ± 19.99^{a}	65.29 ± 32.31^{a}
Feed conversion ratio	3.93 ± 4.46^{a}	1.89 ± 0.30^{a}	1.88 ± 0.36^{a}	1.80 ± 0.53^{a}	1.77 ± 0.71^{a}
Protein retention on carcass (%)	6.41 ± 2.08^{a}	8.11 ± 3.81^{a}	10.06 ± 6.31^{a}	18.88 ± 1.65^{b}	23.88 ± 1.17^{b}
Protein digestibility (%)	88.13 ± 3.12^{a}	90.53 ± 7.12^{a}	90.43 ± 6.23^{a}	89.97 ± 6.02^{a}	91.23 ± 6.72^{ab}

Mean values (±SD) in the same row with the same superscripts did not differ significantly ($P > 0.05$)

Materials and Methods

Experimental design and diets

Five doses of vitamin E (D alpha-tocopherol, Roche International) were tested in this study: 0 mg kg^{-1} or no vitamin E as the control (A); 150 mg kg^{-1} (B); 300 mg kg^{-1} (C); 450 mg kg^{-1} (D); E 600 mg kg^{-1} (E). Each treatment was in three replicates. An iso-protein formulated diet (30% protein) was utilized in this study. The diet was formulated using the following raw materials: fishmeal (45% crude protein), soybean meal (42% crude protein), corn flour (10% crude protein), fine rice bran (9% crude protein), cassava flour (1.5% crude protein), and ebi shrimp flour (45% crude protein) (Table 1). The materials were purchased at a local market in Banda Aceh. The materials were mixed and pelleted using an extruder and then sun dried for 24 hours prior to use with the experimental fish. The pellet was crushed into 0.2-0.3 mm using a mortar before being fed to the experimental fish.

Experimental fish

A total of 150 *T. tambra* fingerlings with an average body weight of 2.69 g and an average total body length of 6.5 cm were used in this study. The experimental fish were weaned for experimental diets for one week prior to use in this experiment. The fish were stocked in plastic aquariums (volume 25 L) at a stocking density of 10 fish per $aquarium^{-1}$ with a water volume of 15 L. The fish were fed two times daily (08:00 and 17:00) with the respective experimental diet at a ratio of 5% body weight day^{-1} for 60 days. The water quality during the experiment was as follows: dissolved oxygen – 10.5-12.0 mg L^{-1}; pH – 7.5-7.9; water temperature – 29-30°C.

Proximate analysis of carcass and feces

Three individuals were collected randomly from each experimental aquarium at the end of the experiment. The carcasses were used for protein content analysis, while the feces were collected three hours after feeding, dried at room temperature, and then processed for protein content. The proximate compositions of the raw materials, the experimental diets, the carcass, and feces were analyzed according to AOAC procedures (1990). Dry matter was calculated by weight loss after 72 h at 70°C. Crude protein was measured using the Kjeldahl technique. About one gram samples were weighed and placed into

a Kjeldahl beaker, then 10 g catalyst and 25 ml sulfuric acid were added to the beaker. The samples were heated at 250°C for 20 minutes and slowly shaken. The temperature was then increased to 350°C for 2 hours. After the samples had been cooled for 10 minutes, 300 ml distilled water was added to each beaker. The diluted samples were distilled, followed by titration using 0.1 N HCl (Muchlisin et al. 2006).

Parameters and data analysis

Total weight gain, daily growth rate, specific growth rate, survival rate, and hepatosomatic index were analyzed. Measurements were taken at ten-day intervals from all fish in every tank for total body length (± mm) and body weight (± g). The survival rate was calculated as follows: SR = $[(N_o-N_t)/N_o] \times 100$, where SR= survival rate (%), N_t = total fish died during of the experiment, N_o = total fish at the start of the experiment. Weight gain was calculated as follows: $W_g = W_t - W_o$, where W_g = weight gain (g), W_t = average body weight of fish at the end of the experiment (g), W_0 = average body weight of fish at the start of the experiment (g). Specific growth rate was determined using the following formula: SGR = $[(\text{Ln } W_t - \text{Ln } W_o) / t] \times 100$. Daily growth rate was calculated using the following formula: DGR= $(W_t - W_o)/ t$, where SGR = specific growth rate (% day^{-1}), DGR= daily growth rate (g day^{-1}), t = duration of the feeding experiment (day). Feed efficiency was calculated using the formula as follows: FE = 1/ FCR × 100%, where FE= feed efficiency (%), FCR = feed conversion ratio calculated as follows: FCR = F / $(W_t - W_o)$, where F= total feed intake (g). Protein retention on the carcass was examined using the formula proposed by Takeuchi et al. (1981) as follows: PR = [(CP- IP) / FP] × 100, where PR = protein retention on the carcass (%), CP = protein content on the carcass at the end of the experiment (%), IP = protein content on the carcass at the beginning of the experiment (%), FP = protein content of the formulated diet (%). Protein digestibility (PG) was calculated as follows: PG (%) = [(protein in diet – protein in the feces)/ protein in the diet] × 100. All data were subjected to analysis of variance (ANOVA), followed by comparison of means using Duncan's multiple range test at a 95% confidence level (P = 0.05).

Results

The results showed that the total body weight gain ranged from 0.33 g to 0.54 g, the daily growth rates were 0.007 to 0.011 g day^{-1}, and the specific growth rate was 0.30 – 0.36% day^{-1}, while the survival rate was 54.76-64.29%, and hepatosomatic index was 31.23-41.52% (Table 2). The study revealed food efficiency and food conversion ratio values that ranged from 53.36% to 65.29% and 1.89 to 3.93, respectively. The protein retention on the carcass was 6.41-23.88%, while the protein digestibility was 88.13.44-91.23% (Table 2). In general, higher growth performance, feed conversion ratio, feed efficiency, protein retention, and protein digestibility were found at 600 mg kg^{-1} feed, but these values did not differ significantly from those of other doses except with regard to protein retention and protein digestibility. In addition, the hepatosomatic index and survival rate were higher at the dosage of 300 mg kg^{-1}, but they were not significantly different at the other dosages. The ANOVA test revealed that the dosages of vitamin E in the diets did not have significant effects on the growth performance, survival, hepatosomatic index, feed efficiency, or feed conversion ratio of *T. tambra* fingerlings (P > 0.05), but that they did have a significant effect on protein retention on the carcass and protein digestibility (P < 0.05).

Discussion

The study revealed that the addition of vitamin E to the diet effected better performance trends in comparison to the control group (without vitamin E), where 600 mg kg^{-1} diet resulted in a higher growth rate, hepatosomatic index, feed efficiency, feed conversion ratio, protein retention on the carcass, and

protein digestibility. However, these values were not significantly different from other doses except with regard to protein retention and protein digestibility. Similar studies were reported by Amlashi et al. (2012) for beluga, *Huso huso* (L.), tilapia, *Oreochromis niloticus* (L.) (Lim et al. 2009), and Cowey et al. (1983) for rainbow trout, *Oncorhynchus mykiss* (Walbaum), where no differences in growth performance among tested doses of vitamin E were observed. However, a contrasting result was reported for *P. scalare* in which dietary vitamin E had a significant impact on body weight increase, specific growth rate, and food conversation ratio with increasing levels of vitamin E, and the best result in growth performance was obtained at 600 mg kg^{1} (Nekoubin et al. 2012).

Overall, dietary vitamin E (alpha-tocopherol) did not have a significant effect on growth performance, but it had a positive effect on carcass quality (protein content on the carcass) and the protein digestibility rate as recorded in this study. Tocopherol deficiency leads to reduced fat and protein content in the ovaries and carcass (Steffens 1989). According to Tokuda et al. (2000), tocopherol plays an important role in fish reproduction. For example, the addition of tocopherol to a sea bream diet resulted in significantly improved fecundity (Izquierdo et al. 2001). Furthermore, vitamin E also has a significant effect on fish health as reported by Falahatkar et al. (2012) who observed higher resistance among beluga juveniles fed dietary vitamin E. A similar result was also reported by Montero et al. (2001); dietary vitamin E had a positive effect on reducing stress in sea bream, *Sparus aurata* L.

In general, optimal requirements for dietary vitamin E is species dependent: for example, 131.9 mg kg^{-1} of feed for rohu, *Labeo rohita* (Hamilton) (Sahoo and Mukherjee 2002); 100-200 mg kg^{-1} for *Clarias batrachus* (L.) (Roy and Molllah 2009); 60-240 mg kg^{-1} of diet for channel catfish, *Ictalurus punctatus* (Raf.) (Gatlin et al. 1992) 35-300 mg kg^{-1} of diet for chinook salmon, *Oncorhynchus tshawytscha* (Walbaum) (Thorarinsson et al. 1994); 100 mg kg^{-1} grass carp, *C. idella* (Li et al. 2014); 99 mg kg^{-1} for *Cirrhinus mrigala* (Hamilton) (Paul et al. 2004). However, alfa-tocopherol hypervitaminosis resulted in poor growth, toxic liver reaction, and death (Udo and Afia 2013). Hence, based on this study, it is suggested that the optimum level of vitamin E for *T. tambra* is 150 mg kg^{-1} feed, as it is not significantly different from 300 mg kg^{-1} and 600 mg kg^{-1} feed.

In conclusion, dietary vitamin E did not have a significant effect on the growth performance or survival rate, but it significantly affected protein retention on the carcass and protein digestibility. However, the best results were noted in the fish fed dietary vitamin E as compared to those that did not receive vitamin E; a dose of 600 mg kg^{-1} resulted in higher growth performance, feed efficiency, feed conversion, protein retention, and digestibility, but these values did not differ significantly from those of other doses. Therefore, it was concluded that the dietary vitamin E dose of 150 mg kg^{-1} feed was the optimum one for *T. tambra*.

Acknowledgements. The study was financed by a Research Grant from the Directorate General of Higher Education (DGHE), Republic of Indonesia under the MP3EI scheme. We express appreciation to anonymous reviewers for their critical comments and suggestions. The technical assistance by all members of the Aquaculture Research Group of Syiah Kuala University, Banda Aceh, Indonesia is also acknowledged.

Author contributions. Z.A.M. conceived and designed the experiments, interpreted the data and approved the final manuscript, A.A.A., I.I.A. contributed materials and performed the research, A.A.M, N.F. interpreted the data, statistically analyzed the data and wrote the draft manuscript, M.N.S. statistically analyzed the data, reviewed the manuscript and substantially improved the language quality of the paper.

References

Amlashi A.S., Falahatkar B., Sharifi S.D. 2012 – Dietary vitamin E requirements and growth performance of young-of-the-year beluga, *Huso huso* (L.) (Chondrostei: Acipenseridae) – Arch. Pol. Fish. 20: 299-306.

AOAC 1999 – AOAC official methods 199.10 – Association of Official Analytical Chemists, Maryland, USA, 40-50 p.

Clerton P., Traoutaud D., Verlhac V., Gabaudan J., Deschaux J. 2001 – Dietary vitamin E and rainbow trout (*Oncorhynchus mykiss*) phagocyte functions: effect on gut and on head kidney leucocytes – Fish Shellfish Immunol. 11: 1-13.

Cowey C.B., Cho C.Y., Watanabe T. 1983 – Finfish nutrition in Asia methodelogical apphroaches to research and development – Tokyo University of Fisheries, Tokyo.

Falahatkar B., Amlashi S.A., Conte F. 2012 – Effect of dietary vitamin E on cortisol and glucose response to handling stress in juvenile Beluga *huso huso* – J. Aquat. Anim. Health 24: 11-16.

Gatlin D.M., Bai S.C., Erickson M.C. 1992 – Effects of dietary vitamin E and synthetic antioxidants on composition and storage quality of channel catfish, *Ictalurus punctatus* – Aquaculture, 106: 323-332.

Halver J.E. 1989 – Fish nutrition, Second Edition – Academic Press Inc. San Diego, California, 789 p.

Izquierdo M.S., Fernández-Palacios H., Tacon A.G.J. 2001 – Effect of broodstock nutrition on reproductive performance of fish – Aquaculture, 197: 25-42.

Kottelat M., Whitten A.J., Kartikasari S.N., Wirjoatmodjo S. 1993 – Freshwater fishes of Western Indonesia and Sulawesi – Periplus Editions Limited, Singapore, 293 p.

Li. P., Wang X., Gatlin III D.M. 2008 – RRR-α-Tocopheryl succinate is a less bioavailable source of vitamin E than all-rac-α-tocopheryl acetate for red drum, *Sciaenops ocellatus* – Aquaculture, 280: 165-169.

Li J., Liang X.F., Tan Q., Yuan X., Liu L., Zhou Y., Li B. 2014 – Effects of vitamin E on growth performance and antioxidant status in juvenile grass carp *Ctenopharyngodon idellus* – Aquaculture, 430: 21-27.

Lim C., Yildirim-Askoy M., Li M.H., Welker T.L., Klesius P.H. 2009 – Influence of dietary levels of lipid and vitamin E growth and resistance of Nile tilapia to *Streptococcus iniae* challenge – Aquaculture, 298: 76-82.

Linder M.C. 1992 – Biochemistry nutrion and metabolism – University of Indonesia Press, Jakarta, 781 p.

Montero D., Tort L., Robaina L., Vergara L., Izquierdo M.S. 2001 – Low vitamin E in diet reduces stress resistance of gilthead seabream (*Sparus aurata*) juveniles – Fish Shellfish Immun. 11: 473-490.

Muchlisin Z.A. 2013 – Study on potency of freshwater fishes in Aceh waters as a basis for aquaculture development programs – J. Iktiol. Indon. 13: 91-96.

Muchlisin Z.A., Siti-Azizah M.N. 2009 – Diversity and distribution of freshwaters fish in Aceh waters Northern – Sumatera, Indonesia – Int. J. Zool. Res. 5: 62-79.

Muchlisin Z.A., Hashin R., Chien A.C.S. 2006 – Influence of dietary protein levels on growth and egg quality in brodstock female bagrid catfish (*Mystus nemurus* Cuv. & Val.) – Aquacult. Res. 37: 416-418.

Muchlisin Z.A., Akyun Q., Rizka S., Fadli N., Sugianto S., Halim A., Siti-Azizah M.N. 2015a – Ichthyofauna of Tripa Peat Swamp Forest, Aceh province, Indonesia – CheckList 11: 1560.

Muchlisin Z.A., Batubara A.S., Siti-Azizah M.N., Adlim M., Hendri A., Fadli N., Muhammadar A.A., Sugianto S. 2015b – Feeding habit and length weight relationship of keureling fish, *Tor tambra* Valenciennes, 1842 (Cyprinidae) from the western region of Aceh Province, Indonesia – Biodiversitas, 16: 89-94.

Nekoubin H., Hosseynzadeh M., Imanpour M.R., Asgharimoghadam A., Raki M., Montajami S. 2012 – Effect of vitamin E (A-tocopheryl) on growth reproductive performance and survival rate of angel fish *(Pterophyllum scalare)* – World J. Zool. 7: 285-288.

Paul B.N., Sarkar S., Mohanty S.N. 2004 – Dietary vitamin E requirement of mrigal, *Cirrhinus mrigala* fry – Aquaculture, 242: 529-536.

Roy A., Mollah M.F.A. 2009 – Effects of different dietary levels of vitamin E on the ovarian development and breeding performances of *Clarias batrachus* (Linnaeus) – J. Bangladesh Agril. Univ. 7: 183-191.

Rudi E., Muchsin I. 2011 – The coral reef fish in Aceh water and its sorrounding. Lubuk Agung, Indonesia, 215 p.

Rudi E., Elrahimi S.A., Kartawijaya T., Herdiana Y., Setiawan F., Shinta P., Campbell S.J., Tamelander J. 2009 – Reef fish status in northern Acehnese reef based on management type – Biodiversitas, 10: 87-92.

Sahoo P.K., Mukherjee S.C. 2002 – Influence of high dietary α-tocopherol intakes on specific immune response, non-specific resistance factors and disease resistance of healthy and aflatoxin B1- induced immunocompromised Indian major carp, *Labeo rohita* (Hamilton) – Aquacul Nut. 8: 159-167.

Steffens W. 1989 – Principles of fish nutrition – Ellis Horwood, New York, 384 p.

Takeuchi M., Ishii S., Ogiso T. 1981 – Effect of dietary vitamin E on growth, vitamin E distribution and mortalities of the fertilized eggs and fry in ayu *(Plecoglossus altivelis)* – Bull. Tokai Reg. Fish. Res. Lab. 104: 111-122.

Thorarinsson R., Landolt M.L., Elliott D.G., Pascho R.J., Hardy R.W. 1994 – Effect of dietary vitamin E and selenium on growth, survival and the prevalence of *Renibacterium salmoninarium* infection in chinook salmon (*Oreochynchus tshawytscha*) – Aquaculture, 121: 343-358.

Tokuda M., Yamaguchi T., Wakui K., Sato T., Ito M., Takeuchi M. 2000 – Tocopherol affinity for serum lipoprotein of Japanese flounder *Paralichthys olivaceus* during the reproduction period – Fish. Sci. 66: 619-624.

Udo I.U., Afia O.E. 2013 – Optimization of dietary vitamin E (tocopherols) in fish: a review – Niger. J. Agricult. Food Environ. 9: 99-107.

Watanabe T., Fujimura T., Lee M.J., Fukusho K., Satoh S., Takeuchi T. 1991 – Effect of polar and nonpolar lipids from krill on quality of eggs of red seabream *Pagrus major* – Nippon Suisan Gakk. 57: 695-698.