Egyptian Journal of Aquatic Biology & Fisheries
Zoology Department, Faculty of Science,
Ain Shams University, Cairo, Egypt.
ISSN 1110 – 6131
Vol. 29(2): 531 – 548 (2025)
www.ejabf.journals.ekb.eg


# Evaluation of Function of Different Levels of Vitamin E on Reproductive Performance and Histological Study of Female Hybrid Red Tilapia (*Oreochromis* sp.)

**Alaa Sh. Griesh[1], Amal M. El-Nahla[2], Salah M. Aly[3], Mohamed F. Badran[1*]**

[1]Department of Aquatic Hatchery Production, Fish Farming and Technology Institute (FFTI), Suez Canal University, 41522 Ismailia, Egypt
[2]Department of Physiology, Faculty of Veterinary Medicine, Suez Canal University, Ismailia, Egypt
[3]Department of Pathology, Faculty of Veterinary Medicine, Suez Canal University, 41522 Ismailia, Egypt

***Corresponding author: mohamed.fekri.fish@suez.edu.eg**

**ARTICLE INFO**

**Article History:**
Received: Jan. 22, 2025
Accepted: Feb. 28, 2025
Online: March 11, 2025

**Keywords**:
Hybrid red tilapia,
Vitamin E,
Reproductive Performance,
Post spawning,
Ovaries histology

**ABSTRACT**

The hybrid red tilapia is highly sought after worldwide due to its delicious flavor, attractive appearance, ease of cultivation, adaptability to challenging environments, strong disease resistance, short generation time, and rapid growth. In the red tilapia hatchery, there is difficulty spawning of some red strains of tilapia and low viability red tilapia eggs and fry. Therefore, this study aimed to evaluate the function of different levels of vitamin E on reproductive performance and histological sections of hybrid female red tilapia *Oreochromis* spp. postspawning (at 60$^{th}$ days). Females were divided into four groups, including 4 vitamin E treatments (0, 50, 100, and 200mg kg$^{-1}$ diet). Each group had 3 replicates. The results showed that Vit. E had an effective impact on reproductive performance as oocyte diameter, egg diameter, fecundity, hatching rate, spawning percentage, and larval length at 60$^{th}$ day post-spawning and on histological sections of ovaries that showed high maturity with revealed yolk granules and yolk plates. In conclusion, adding vitamin E with different doses (50, 100, and 200mg/ kg of feed) had an effective influence on the reproductive performance and histological sections of the ovaries of females of hybrid red tilapia. Supplementing with a 50mg/ kg Vit. E dosage showed to be sufficient for a high reproductive performance of females of the hybrid red tilapia hatchery.

## INTRODUCTION

Aquaculture is considered one of the fastest-growing sectors in food industry globally, expanding daily **(Syed *et al.*, 2021)**. Among its various forms, tilapia farming holds a significant promise in addressing global challenges such as hunger, malnutrition, and poverty, especially in Africa **(Khalil *et al.*, 2024)**. Hybrid variant of *O. niloticus* and *O. mossambicus*, known as hybrid red tilapia, gained popularity widespread due to the appealing taste, attractive appearance, adaptability to harsh environments, omnivorous diet, disease resistance, rapid growth, and short reproductive cycle **(Haque *et al.*, 2016)**. Studies have shown that Mozambicans and red tilapia are particularly well-suited for cultivation in marine environments, demonstrating strong growth performance in such

conditions **(Tayamen *et al.*, 2002; Sallam *et al.*, 2017)**. However, production of Egyptian aquaculture seems to introduce low of red tilapia **(Zaki *et al.*, 2021)**.

Despite its advantages, red tilapia culture faces several challenges. Certain strains struggle with spawning, while others exhibit low survival rates among eggs and fry. These issues are often attributed to continuous inbreeding aimed at preserving the red coloration, which can lead to reduced genetic diversity and poor survival rates in early life stages **(Lovshin, 2000)**. Many regions of the world are unable to produce red tilapia commercially due to a lack of adequate quality egg supply. Furthermore, inappropriate broodstock management can lead to inbreeding, resulting in low-quality seed production **(Lingam *et al.*, 2021)**. Identifying suitable hybrid red tilapia candidates with sufficient genetic diversity for establishing founder populations remains a major hurdle, compounded by the limited availability of broodstock **(Mohamad *et al.*, 2021)**.

Nutrition plays a crucial role in reproductive fish success, as the energy demands for gamete production and spawning behaviors are exceptionally high. Insufficient energy intake or poor nutrition can suppress reproductive functions **(Volkoff & London, 2018)**. Vitamins are one of the most significant nutrients for reproduction. They are not manufactured by animals; therefore, they must acquire them from their food to meet their needs **(Khalil *et al.*, 2024)**. **Fernández-Palacios *et al.* (2005)** revealed that vitamin E, for instance, has been shown to enhance spawning quality across various fish species. In the gilthead sea bream, diets deficient vitamin E decline fertilized eggs. A lack of vitamin E also decreased viable eggs with normal morphology in several species and larval survival was significantly improved with Vit. E inclusion in broodstock diets. Deficiency of Vit. E results in underdeveloped gonads, low fecundity and fertility and lower hatching rates and fry survival. Conversely, rich levels of dietary Vit. E in diets promote gonadal development, the gonadosomatic index, gonadal maturation, and improve egg quality, viability, hatching rates as well as the percentage of healthy larvae **(Volkoff & London, 2018)**.

Research has also established a direct relationship between dietary Vit. E levels and its concentration in ovarian tissues. Over the past two decades, the importance of Vit. E in improving development of gonadal, reproductive processes, as well as larval survival has been widely recognized. High levels of dietary Vit. E can mitigate egg deformities, enhance fecundity, and protect eggs from oxidative damage **(Izquierdo & Fernandez Palacios, 1997; Erdogan & Arslan, 2019)**. Therefore, the current study aimed to evaluate the use of Vit. E supplementation diets, and its impacts on reproductive performance, and histological sections of the female hybrid red tilapia.

## MATERIALS AND METHODS

The current study was carried out at hatchery unit and glass aquaria within the (AHPD) Aquatic Hatchery Production Department, FFTI at Suez Canal University (SCU), located in Ismailia, Egypt.

**1- Ethical approval**

The study adhered strictly to guidelines set by the Experimental Animal Care Committee. Additionally, it received approval from the Scientific Research Ethical Committee (SREC), Faculty of Veterinary Medicine, SCU **(Approval No. 2021016)**.

**2- Broodstock maintenance and tanks preparation**

A total of 72 female red tilapia hybrids were captured at FFTI, SCU, Ismailia, Egypt, and brought to the hatchery unit. Prior to handling, the fish were starved and sedated with Ms-222 (Tricaine methane sulfonate, dose: 100mg/ L, Argent Lab. Inc. Philippines) **(Popovic *et al.*, 2012)**, and then biological measurements of the fish were taken. Broodstocks were split into two sexes to differentiate between male and female by evaluating the genital papilla with gentian violent antiseptic on the urogenital orifice, which was conspicuous and pointed in males but rounder in females. Females' average body weight was 272.56 ± 34.84g, with a total length of 24.3 ± 2.5cm. Females were housed in 12 indoor fiberglass circular holding tanks, each with a maximum capacity of $3m^3$, filled with $1.5m^3$ saltwater under the water conditions indicated in Table (1), and around 50% of the water was replaced daily with aerated water. Females were separated into four equal groups and given four doses of Vit. E (0, 50, 100, and 200mg per kg diet). Each group comprised three replicates, with a stocking density of 6 fish per tank (4 fish per $m^3$) **(Nascimento *et al.*, 2014; Erdogan & Arslan, 2019)**.

The tanks were subjected to an artificial photoperiod of 12 hours of light/12 hours of darkness 2500 lux **(El-Sayed, 2020)**. Water was filtered using sandy filters and sterilized with ultraviolet units. Continuous aeration was continuously provided by air blower three diffuser stones per tank.

**3- Physico-chemical indicators of water of the experiment**

The water indicators were tested and controlled daily in the indoor holding tanks using a thermometer and dissolved oxygen (DO) meter (ExStik II D-0600, USA), refractometer (DRBS-300), and Milwaukee MW-100 pH meter.

**Table 1.** The experiment's water quality parameters

| | Parameters | Values |
|---|---|---|
| 1 | Temperatures | 28±1.5 °C. |
| 2 | DO | 7.2 ± 0.5 mg/L |
| 3 | pH | 7.2±0.7 |
| 4 | Salinity | 14±1 ppt |
| 5 | Ammonia & Nitrite | ≤0.01 mg/L |

**4- Feeding regimen**

Females were manually fed a commercial diet which contained: 30% crude protein, 6.2% crude fat, 4.5% fiber, 6.1% ash, and 4mm pellets twice daily for 60 days at a 3% feeding rate. Feed sources were obtained from Skretting Egypt Company (SEC). Vit. E was obtained from Karma care co. for veterinary product, Egypt. Vit. E was incorporated into the feed at four different levels: ($E_0$) 0mg, ($E_1$) 50mg, ($E_2$) 100mg, and ($E_3$) 200mg. Moreover, Vit. E levels were mixed following the process described by **Abdelhamid *et al.* (2023)** and **Griesh *et al.* (2024)**.

**5- Mating process (Postspawning)**

On the 45$^{th}$ day, ripped females and males of hybrid red tilapia were placed in tanks with a water capacity of 1.5m$^3$ at sex rate of 4 females: 3 males and were fed for an additional 15 days until the 60$^{th}$ day. To minimize injuries to females caused by male aggression during spawning, males and females of similar size and weight were paired. A total of 36 male hybrid tilapia red, with weight of about 298.63±10.50g and length of 22.41± 1.86cm, were used. After 15 days (at 60$^{th}$ day), representative samples of different broodstock fish from each treatment were collected to determine reproductive parameters and histological examination of ovaries of females.

**6- Reproductive performance**

On the 60$^{th}$ day (post spawning), the females were fasted for 24 hours before sampling. They were captured and weighed, and from each tank two fish were dissected; their liver, and gonads were weighed. The reproduction performance was evaluated using the following parameters:

Viscerosomatic index % (**VSI**) = $\frac{Viscera\ weight\ (g)}{Body\ weight\ (g)} \times 100$

Hepatosomatic index % (**HSI**)= $\frac{Liver\ weight\ (g)}{Total\ Body\ weight\ (g)} \times 100$

Gonadosomatic index (**GSI**) = $\frac{Gonad\ weight}{Total\ Body\ weight} \times 100$

Fulton conditions factor (**K**) = $\frac{Body\ weight \times 100}{Total\ length^3}$

**(Hastings & Dickie, 1972; Badran *et al.*, 2019; Obirikorang *et al.*, 2019)**

**Oocyte diameter**

Fifty oocytes were extracted from the ovarian tissues and were put in a saline solution (0.9 % NaCl), and then transferred to a slide to measure their diameters. They were measured to the nearest 0.01µm by using an ocular micrometer on the binocular microscope at magnification of 40x and 10x, and the percentage occurrence was plotted

against their diameters class interval to confirm the ovulation pattern **(Badran *et al.*, 2019)**.

Egg diameters were measured using an ocular micrometer on a binocular microscope at magnification of 40x and 10x. For stage one eggs (n= 10), it was measured to the nearest 0.01mm under a calibrated binocular microscope. Since the eggs are ellipsoid-shaped, both axes (long and short) were measured and then calculated the mean egg diameter [Σ10 eggs (length + width)/ 20] **(Coward & Bromage, 1999)**.

Fecundity = (no. of eggs× gonad weight)/ (weight of gonad sample) **(Badran *et al.*, 2019)**.

Hatching rate = (Total no. of hatched eggs/Total no. of stocked eggs)*100 **(Erdogan & Arslan, 2019)**.

Spawning percentage = (no. of spawn females / no. of total females) *100 **(Gammanpila *et al.*, 2010)** and larval length.

### 7- Histological study of ovaries

Ovaries from a total of 24 fish were collected on the 45$^{th}$ day (pre-spawning) and another 24 fish on the 60$^{th}$ day (post spawning). Fish were transported alive to the laboratory and were examined physically to ensure they were free from any pathological abnormalities. Specimens of approximately one cubic cm were taken from different parts of ovary and fixed in neutral buffered formalin for about 24 hours. Clearing and paraffin embedding were performed using standard histological techniques, after one week they were dehydrated in ascending alcohol series (70, 80, 90, 95% then absolute alcohol), exposed to xylene and embedded in paraffin wax. Sections of 5-6µm thickness were cut, stained with Harris hematoxylin and Eosin, and mounted with DPX **(Ratcliffe, 1982; Thulasitha & Sivashanthini, 2013)**. Histological sections were captured using binocular light microscope (CX23LEDRFS2) equipped with Wi-Fi embedded camera (AC1200).

### 8- Statistical analysis of the experiment

Collected data were presented as means ± stander error (SE). Significant differences were calculated using one-way ANOVA at level of 0.05. Post hoc multiple tests were used for comparisons **(Duncan, 1955)**. Moreover, SPSS, Richmond, USA (version 22) was used in the statistical analysis, as described by **Dytham (2011)**.

## RESULTS

### 1- Reproductive parameters

$E_1$ showed a significant value in VSI, and GSI compared to control while no significant differences were shown between $E_2$ and $E_3$. HSI values showed no significant differences between control and treated groups (Fig. 1). Highest K factor was recorded

at $E_1$ (2.05) followed by $E_2$ (1.86), while the lowest value was recorded at control ($E_0$) (1.59) (Fig. 2).

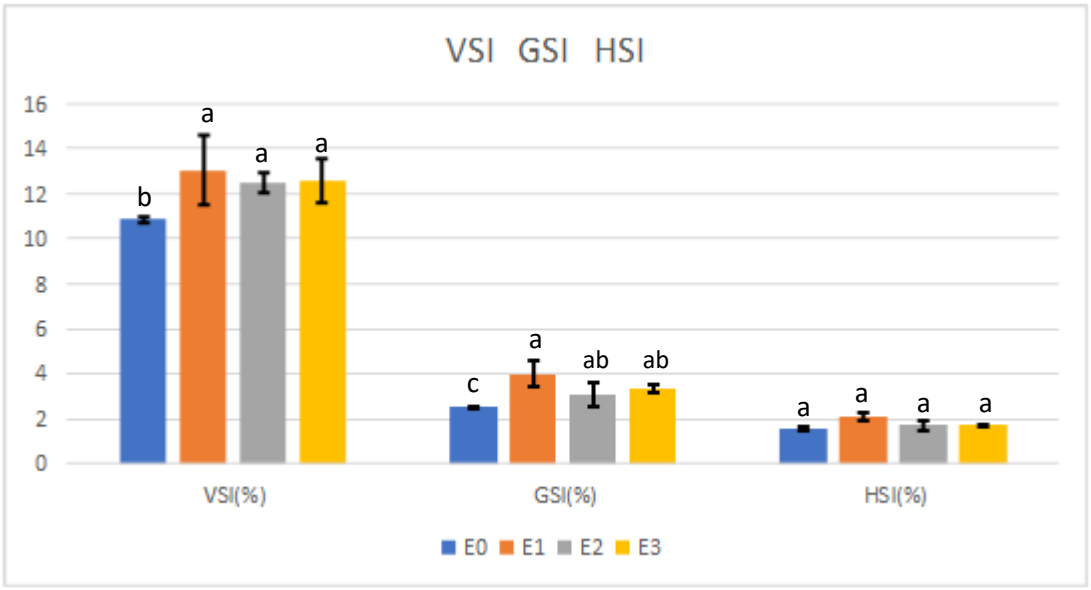


**Fig. 1.** Effects of different levels of VE on VSI, GSI, and HSI of female hybrid red tilapia at 60th day (post spawning). E0: (0 mg/kg of Vit. E), E1: (50 mg of Vit. E), E2 (100 mg of Vit. E), E3: (200 mg of Vit. E)

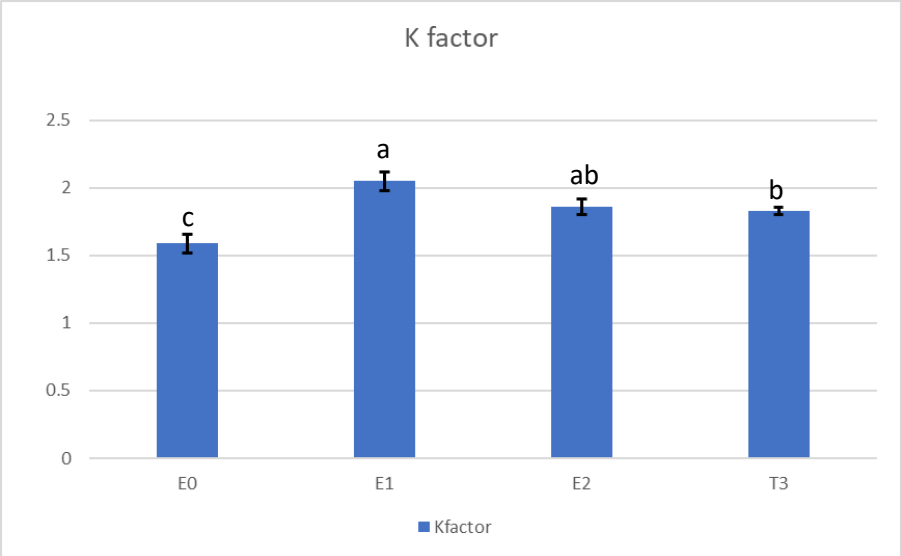


**Fig. 2.** The effect of different levels of VE on K factor of female hybrid red tilapia at 60th day (post spawning). E0: (0 mg of Vit. E), E1: (50 mg of Vit. E), E2 (100 mg of Vit. E), E3: (200 mg per kg of Vit. E).

The oocyte diameter (OD) in all treated groups showed higher significant value when compared to $E_0$. The same trend was revealed in larval length (LL). Egg diameter (ED), fecundity and hatching rate (HR) were significantly increased in $E_1$ as compared to

control or each treated groups $E_2$ and $E_3$. The peak of spawning percentage (SP) was recorded in $E_1$ and $E_3$ (Table 2).

**Table 2.** Impact of different Vit. E levels on oocyte diameter (OD), (ED) egg diameter, fecundity, hatching rate (HR), spawning percentage (SP) and larval length (LL) of female hybrid red tilapia at 60th day (post spawning)

| Treatment | OD (mm) | ED (mm) | Fecundity | HR (%) | SP (%) | LL (mm) |
|---|---|---|---|---|---|---|
| **$E_0$** | $0.60 \pm 0.04^{b}$ | $0.92 \pm 0.01^{c}$ | $1073.33 \pm 117.95^{c}$ | $63.67 \pm 3.22^{c}$ | $50.77 \pm 8.75^{b}$ | $2.67 \pm 0.13^{b}$ |
| **$E_1$** | $0.86 \pm 0.04^{a}$ | $1.16 \pm 0.02^{a}$ | $2523.57 \pm 320.28^{a}$ | $92.50 \pm 1.61^{a}$ | $73.22 \pm 5.13^{a}$ | $3.07 \pm 0.03^{a}$ |
| **$E_2$** | $0.80 \pm 0.06^{a}$ | $1.04 \pm 0.02^{b}$ | $1760.83 \pm 111.56^{b}$ | $77.06 \pm 2.23^{b}$ | $61.81 \pm 2.45^{ab}$ | $3.03 \pm 0.09^{a}$ |
| **$E_3$** | $0.79 \pm 0.06^{a}$ | $1.09 \pm 0.01^{b}$ | $1834.17 \pm 133.15^{b}$ | $78.67 \pm 2.17^{b}$ | $69.41 \pm 1.51^{a}$ | $3.00 \pm 0.10^{a}$ |

Data were presented as Mean ± SE within columns. Means were significant at $P< 0.05$. E0: (0mg of Vit. E), E1: (50mg of Vit. E), E2 (100mg of Vit. E), E3: (200mg of Vit. E)

## 2- Histological examination

### *2.1. Effect of different Vit. E levels on histological study of females' gonads at 45th day (pre-spawning)*

Fig. (3-I) exhibits: a- A microphotograph of a histological section of $E_0$ ovaries in hybrid red tilapia display a normal histological structure, featuring different development stages of oocytes, primarily cortical alveolar oocyte and mature, ripped oocyte. b- A microphotograph of a histological section of 50mg\kg diet group of ovaries of hybrid red tilapia showing typical structure of mature ripped oocyte, chromatin nuclear oocyte and perinuclear oocyte. c- A microphotograph of a histological section of 100 mg\kg diet group of ovaries of hybrid red tilapia showing atresia, mild degeneration and deformities of ovarian follicles and presence of chromatin and perinuclear oocyte. d- A microphotograph of a histological section of 200mg\kg diet group of ovaries of hybrid red tilapia showing enhancement of ovarian follicle in comparison with other groups. This section shows all developmental stages in the ovarian tissue, including nucleolar chromatin, perinucleolar, cortical alveolar, and mature ripe oocytes.

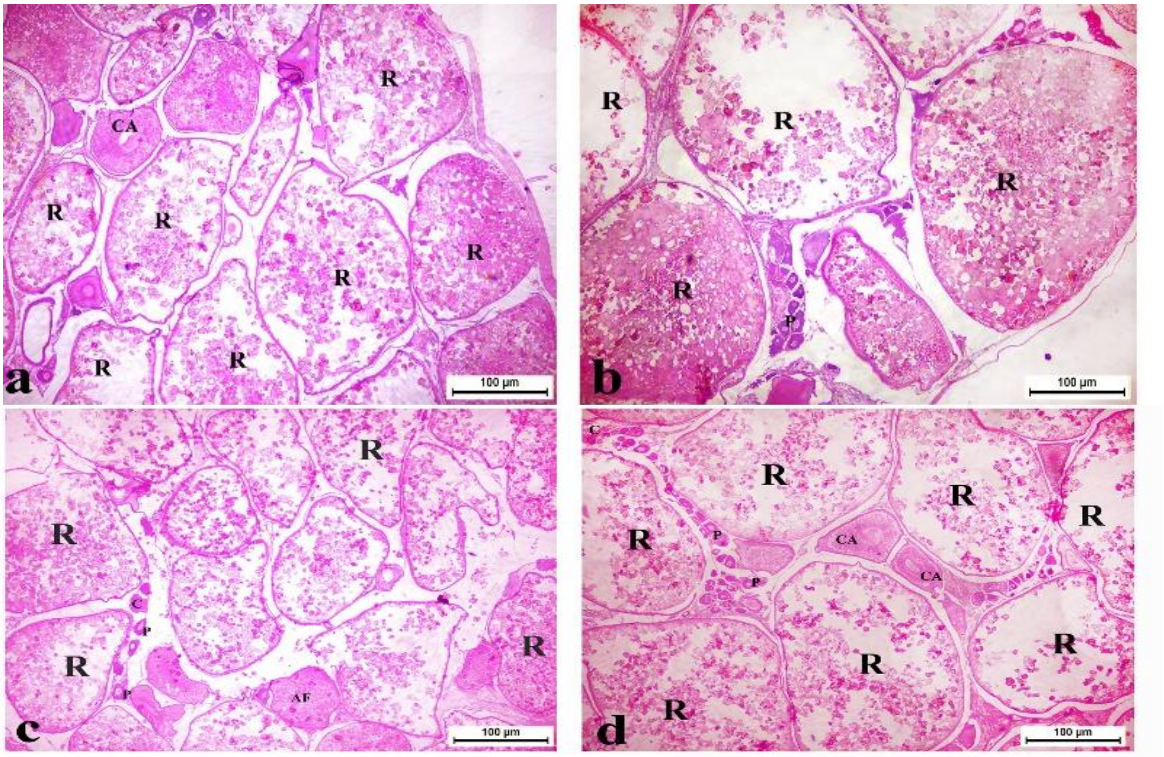


**Fig. (3-I).** Female ovary of hybrid red tilapia fed with dietaries of different Vit. E a: oocyte statue of control ($E_0$), b: 50mg Vit. E\kg diet, c: 100 mg. Vit. E\kg. diet and d: 200mg Vit. E\kg. diet groups at 45$^{th}$ day (pre-spawning). R: ripped oocyte, CA: cortical alveoli, C: chromatin nuclear oocyte, P: perinuclear oocyte, AF: atresia follicle

### *2.2. Effect of different Vit. E levels of on histological study of females' gonads at 60$^{th}$ day (post spawning)*

Fig. (3-II) exhibits: e- A microphotograph of a histological section of control group of ovaries of hybrid red tilapia showing moderate degeneration and necrosis of ovarian follicles with inter-follicular edema and atretic oocyte. f- A microphotograph of a histological section of (50mg\kg diet) group of ovaries of hybrid red tilapia showing atresia and mild edema between follicles. g- A microphotograph of a histological section of (100mg\kg diet) group of ovaries of hybrid red tilapia showing mild improvement of developmental stages with formation of ripping stages and some follicles showed atresia and empty oocyte. h- A microphotograph of a histological section of 200mg\kg diet group of ovaries of hybrid red tilapia showing normal structure and improvement with Fig. (3-II) exhibits: e- A microphotograph of a histological section of control group of

ovaries of hybrid red tilapia showing moderate degeneration and necrosis of ovarian follicles with inter-follicular edema and atretic oocyte. f- A microphotograph of a histological section of (50mg\kg diet) group of ovaries of hybrid red tilapia showing atresia and mild edema between follicles. g- A microphotograph of a histological section of (100mg\kg diet) group of ovaries of hybrid red tilapia showing mild improvement of developmental stages with formation of ripping stages and some follicles showed atresia and empty oocyte. h- A microphotograph of a histological section of 200mg\kg diet group of ovaries of hybrid red tilapia showing normal structure and improvement with presence of all development stages mainly chromatin nucleolar oocyte, perinucleolar oocytes and mature ripped oocytes.

.

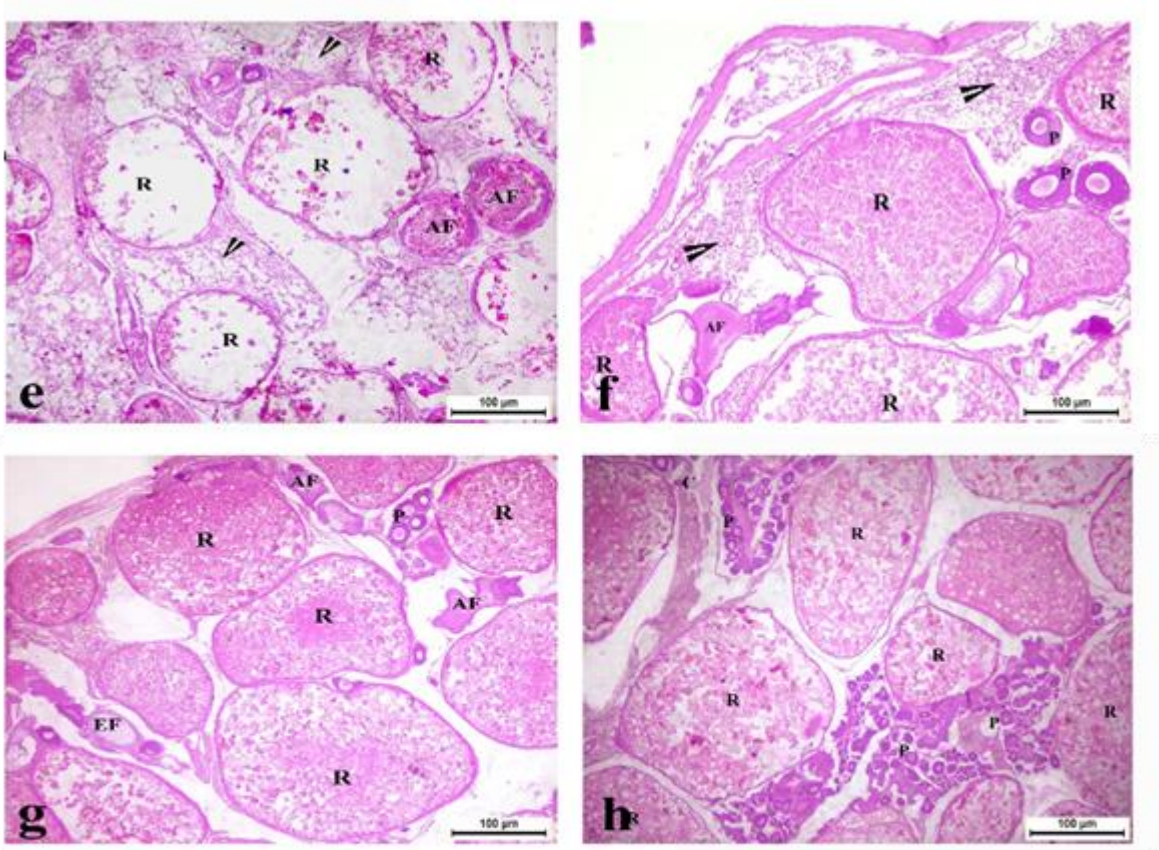


**Fig. (3-II).** Ovary female of hybrid red tilapia with different dietaries of Vit. E. e: oocyte statue of control (0 mg), f: 50mg Vit. E\kg diet, g: 100mg Vit. E\kg diet and h: 200mg Vit. E\kg diet groups at 60$^{th}$ day (post spawning). R: ripped oocyte, P: perinuclear oocyte, EF, empty oocyte, C: chromatin nuclear oocyte, AF: atresia follicle. Arrow: inter-follicular edema.

## DISCUSSION

The current findings indicate that the measurements of biological and performance of reproductive VSI, GSI, as well as K-factor, showed significant improvement with the supplementation of 50mg/ kg of Vit. E in comparison with $E_0$. Nevertheless, insignificant HSI changes between all groups were observed. The GSI values in this study agree with the observations of **Zhang *et al.* (2007)**, who showed that the VSI and GSI significantly increased in groups receiving 50 and 125mg/ kg of vitamin E. Both values rising with 50mg $kg^{-1}$ of Vit. E may be due to the positive effect of Vit. E in the process of gonadal development and reproductive performance in fish **(Arfah *et al.*, 2013)**. Additionally, the appropriate dosage of Vit. E in feed can elevate GSI values in tilapia, as recorded by **Tarigan *et al.* (2021)**. Variations in GSI values are likely influenced by the differing vitamin E doses of feed administered. Performance and reproductive can be enhanced through vitamin E supplementation. Vit. E is a crucial micronutrient since it enhances growth, reproduction, overall fish health **(Hunt, 2004)**. The rise in GSI values is believed to result from vitamin E's role in gonad development, particularly in the biosynthesis of vitellogenin and the vitellogenesis process in the liver. An optimal level of vitamin E in feed accelerates vitellogenesis, leading to faster gonad maturation. Consequently, higher vitellogenin levels during oocyte development increase gonad weight, thereby raising the GSI value **(Tang *et al.*, 2004; Arfah & Setiawat, 2013; Ashari *et al.*, 2021; Tarigan *et al.*, 2021)**. Furthermore, the K-factor values in this study are in line with **Khara *et al*. (2016)**, who found that the highest k-factor values were achieved in Caspian brown trout fed 30 and 40mg.kg of Vit E.

Our results of spawning parameters such as oocyte diameter, egg diameter, larval length, fecundity, hatching rate and spawning percentage increased significantly with varying vitamin E levels (50, 100, and 200mg. $kg^{-1}$). However, 50mg. $kg^{-1}$ dose proved most effective. In fish eggs Vitamin E is typically high, but after spawning in broodstock tissues is low **(Mukhopadhyay *et al.*, 2003)**. This phenomenon is due to Vit. E mobilization, and muscles of adult females to the ovaries during vitellogenesis, as observed in turbot and Atlantic salmon. When fish are fed diets rich in Vit. E, the nutrients accumulate in all tissues. However, a significant portion to ovaries prior to spawning, leads to a higher gonadosomatic index **(Gupta *et al.*, 1987; Hamre *et al.*, 1994; Lie *et al.*, 1994)**. **Sutjaritvongsanon (1987)** demonstrated enhanced development of gonad and spawning in goldfish fed with vitamin E, while **Palace and Werner (2006)** exhibited that tocopherol (Vit. E) stimulated events of multiple spawning. Specifically, supplemented females maintained the index of gonadosomatic from the peak to the post-spawn phase in addition produced about 1.5 times more eggs than supplemented. These findings underscore the critical vitamin E role in enhancing the performance of reproduction as well as spawning success in fish.

Vit. E improves spawning quality in fish **(Hamre, 2011)**. The optimum Vit. E dietary level of broodstock studied in various species is reported to be typically around

150 to 190mg. $kg^{-1}$ **(Fernández-Palacios *et al.,* 2005)**. The improvements in egg diameter and hatching rate in current study are attributed to the properties of Vit. E antioxidant, which helps protect cell membranes from lipid peroxidation, thereby maintaining oocyte viability **(Nascimento *et al.,* 2014)**. This study denoted oocyte diameter, and larval length increased with different vitamin E supplementation at 50, 100, and 200mg. $kg^{-1}$. Egg diameter, fecundity, and hatching rate increased with a dosage of 50mg of Vit. E, while spawning percentage increased with both 50 and 200mg of Vit. E. **Gupta *et al.* (1987)** pointed that Vit. E led to larger ova, and increased egg production in common carp than a control. Similarly, the fecundity and hatchability rate in this study are in line with the observations of **Zhang *et al.* (2007)**, who showed that fecundity and hatchability groups fed 50mg/kg of Vit. E were significantly increased than all groups. Additionally, 200mg of vitamin E in feed significantly improved spawning rate and hatching rate of *Perca Flavescens* **(Lee & Dabrowski, 2004)**. Comparable results have also been documented in studies on other fish species such as *Monopterus albus* **(Zhang *et al.*, 2007)**.

Similar results showed that, supplementing with 150mg/kg of Vit. E significantly enhanced egg, and larva quality in the Nile tilapia, resulting in a better reproduction performance **(Darwisito *et al.,* 2013)**. Additionally, adequate levels of α-tocopherol (Vit. E) supplementation in maternal diets has been shown to improve hatching and rates of larval survival in the Nile tilapia, although no significant effect on egg diameter was observed **(Nascimento *et al.,* 2014)**. Vit. E is also believed to decrease the abnormal eggs in addition, arising the fecundity overall **(Izquierdo *et al.*, 2001)**.

Findings of **Pamungkas *et al.* (2014)** revealed that high yield and values were recorded when *Oreochromis niloticus* feed on Vit. E at 225mg/ kg for maturation percentage and spawning frequency. Similar results were obtained by **Gómez *et al.* (2003)**, who determined that the Vit. E dietary requirement for *Oreochromis niloticus* broodstock ranged between 100 and 200mg per kg to enhance reproductive performance. Vitamin E plays a crucial fertility role when there is no synthesis for vitamin E in fish; the intake dietary maternal oogenesis is a key factor influencing reproductive fitness **(NRC, 1993)**. High levels of Vitamin E are evident in eggs, while broodstock tissue levels decrease after spawning. This suggests a crucial physiological role of Vitamin E in fertilization and hatching processes **(Watanabe, 1985)**.

The current results of the hatching rate were highest with 50mg/. kg of Vit. E, which closely aligns with **Takeuchi *et al.* (1981)**, who noticed improved hatchability of brood fish of Ayu, *Plecoglossus altivelis* with 3.4mg of Vit. E per 100g of feed. Similarly, **Mollah *et al.* (2003)** reported significant Vit. E impact on fertilization and hatching rates in *H. fossilis*, at dose of 200mg per kg.

The antioxidant effect of vitamin E is significant in maintaining oocytes viability **(Nascimento *et al.,* 2014)**. For instance, vitamin E at the highest level was detected in eggs and the European sea bass seminal fluid before and after fertilization, as well as in

developing embryos and at hatching. In contrast, dead eggs and embryos with low survival rates showed significantly lower vitamin E levels **(Rudneva-Titova, 1995)**. A linear correlation has been established between dietary Vit. E and its concentration in ovarian tissue, underscoring its importance in reproductive health. Over the past two decades, the role of dietary vitamin E in supporting gonadal development, reproductive processes, and spawning performance has been widely studied **(Farris *et al.*, 2020)**. Conversely, diets deficient in vitamin E can lead to immature gonads, reduced egg fertilization and hatchability rates, and lower larval survival **(Canyurt & Akhan, 2008; Miller *et al.*, 2012)**. Histological sections confirmed vitellogenesis process in which oocytes undergoing final oocyte maturation (FOM), accumulation of yolk, yolk granules and yolk plates in cortical alveoli all supplemented groups of Vit. E. at $45^{th}$ and $60^{th}$ days (post spawning). In the present study, histological analysis confirmed the vitellogenesis process, with oocytes undergoing FOM and the accumulation of yolk, yolk granules, and yolk plates in cortical alveoli observed in all vitamin E-supplemented groups at 45 and 60 days (post-spawning). These findings further emphasize the critical role of vitamin E in ensuring proper reproductive development and successful spawning outcomes and in enhancing reproductive performance and spawning success in fish.

## CONCLUSION

This study evaluated the function of Vit. E on reproductive performance and histological sections of ovaries of females of the hybrid red tilapia. Adding Vit. E with different doses (50, 100, 200mg/ kg of feed) had an effective influence on the reproductive performance and histological sections of ovaries of females of the hybrid red tilapia. Giving Vit. E with dose (50mg. $kg^{-1}$) sufficient and better execution for the females of the hybrid red tilapia hatchery.

## REFERENCES

**Abdelhamid, A.F.; Gewida, A.G.A.; El-Sayed, A.F.M. and Badran, M.F.** (2023). Impacts of different levels of vitamin K on the growth performance, hematological parameters, and immunological response of juvenile Nile tilapia (*Oreochromis niloticus*). Aquac. Int. 1–12. https://doi.org/10.1007/s10499-023-01169-6

**Albertine-Berhaut, J.** (1973). Biologie des stades juveniles de téleostéens Mugilidae Mugil auratus Risso 1810, Mugil capito Cuvier 1829 et Mugil saliens Risso 1810: I. Régime alimentaire. Elsevier.

**Arfah, H.; Setiawati Departemen Budidaya Perairan, M.; Perikanan dan Ilmu Kelautan, F. and Pertanian Bogor, I.** (2013). Dietary vitamin E on the reproductive performance of the fantail goldfish Carassius auratus auratus. J. Akuakultur Indones. 12, 14–17. https://doi.org/10.19027/JAI.12.14-17

**Arfah, H.M. and Setiawat, I.** (2013). Vitamin E supplementation with different doses at feed on the reproductive performance of the female brood fish comet (*Carassius auratus auratus*). J. Aquac. Indones. 12: 14-18.

**Ashari, A.; Sinaga, M.; Gde, P.; Julyantotro, S.; Ayu, D.; Pebriani, A.; Kampus Unud, J.; Jimbaran, B. and Selatan, K.** (2021). Effect of Giving Different Doses of Vitamin E in Feed to the Level of Gonad Maturity of Tilapia (*Oreochromis niloticus*). Adv. Trop. Biodivers. Environ. Sci. 5, 64–68. https://doi.org/10.24843/ATBES.2021.V05.I02.P05

**Badran, M.; El-Danasoury, M.; Sharaf, S. and Hassan, A.** (2019). Biological Measurements and Hormonal Profile in Female Gilthead Sea Bream (*Sparus Aurata*) In Response to Human Chorionic Gonadotropin (HCG) Injection. Suez Canal Vet. Med. Journal. SCVMJ 24, 103–112. https://doi.org/10.21608/scvmj.2019.59209

**Canyurt, M.A. and Akhan, S.** (2008). Effect of dietary vitamin E on the sperm quality of rainbow trout (*Onchorynchus mykiss*). Aquac. Res. 39, 1014–1018. https://doi.org/10.1111/J.1365-2109.2008.01952.X

**Coward, K. and Bromage, N.R.** (1999). Spawning periodicity, fecundity and egg size in laboratory-held stocks of a substrate-spawning ž / tilapiine, Tilapia zillii Gervais. Aquaculture 171, 251–267.

**Darwisito S; Zairin, D.; S., Manalu M and Sudrajat O, A.** (2013). Pemberian Pakan Mengandung Vitamin E dan Minyak Ikan Pada Induk Memperbaiki Kualitas Telur dan Larva Ikan Nila (*Oreochromis niloticus*). Anal. pendapatan dan tingkat Kesejaht. rumah tangga petani 53, 1689–1699.

**Duncan, D.B.** (1955). Multiple Range and Multiple F-Test. Biometrics, 11, Sci. Res. Publ. https://doi.org/10.2307/3001478

**Dytham, C.** (2011). Statistics, variables and distribution, Choosing and using statistics: A biologist's guide. Wiley-Blackwell.

**El-Sayed, A.F.M.** (2020). Tilapia Culture, Tilapia Culture: Second Edition. Elsevier. https://doi.org/10.1016/C2017-0-04085-5

**Erdogan, M. and Arslan, T.** (2019). Effects of vitamin E on growth and reproductive performance of pindani (Pseudotropheus socolofi Johnson, 1974). Aquaculture 509, 59–66. https://doi.org/10.1016/j.aquaculture.2019.05.019

**Farris, N.W.; Kim, D.; Hamidoghli, A.; Won, S.; Lee, S.; Bae, J. and Bai, S.C.** (2020). Dietary α-Tocopheryl acetate and arachidonic acid synergistically improves

superoxide dismutase activity in female Japanese eel broodstock, Anguilla japonica. Aquaculture 522, 735100. https://doi.org/10.1016/j.aquaculture.2020.735100

**Fernández-Palacios, B.H.; López, I.; Soledad, M. and Robaina, R.** (2005). Efecto de distintas dietas pasa reproductores de dorada (*Sparus aurata*) sobre la calidad de sus puestas. Informes técnicos del instituto canario de ciencias marinas, accedacris.ulpgc.esM Izquierdo, H Fernandez-Palacios, L RobainaInformes Técnicos del Instituto Canario de Ciencias Marinas, 2005•accedacris.ulpgc.es.

**Gammanpila, M. and Yakupitiyage, A.** (2010). Evaluation of the effects of dietary vitamin C, E and Zinc supplementation on reproductive performance of Nile tilapia (*Oreochromis niloticus*). Sri Lanka J. Aquat. Sci. 12, 39. https://doi.org/10.4038/sljas.v12i0.2213

**Gómez-Márquez, J.L.; Peña-Mendoza, B.; Salgado-Ugarte, I.H. and Guzmán-Arroyo, M**. (2003). Reproductive aspects of *Oreochromis niloticus* (Perciformes: Cichlidae) at Coatetelco lake, Morelos, Mexico. Rev. Biol. Trop. 51, 221–228.

**Griesh, A.S.; El-Nahla, A.M.; Aly, S.M. and Badran, M.F.** (2024). Role of Vitamin E Supplementation on the Reproductive and Growth Performance, Hormonal Profile and Biochemical Parameters of Female Hybrid Red Tilapia. Thalassas. https://doi.org/10.1007/s41208-024-00683-5

**Gupta -H.A.; Bhowmick,-R.M., -S.D.; Khan** (1987). Observations on the effect of vitamin E and growth hormone on the gonadal maturity of carps. J.-Inland-Fish.-Soc.-India. 19, 26–31.

**Hamre, K.** (2011). Metabolism, interactions, requirements and functions of vitamin E in fish. Aquac. Nutr. 17, 98–115. https://doi.org/10.1111/j.1365-2095.2010.00806.x

**Hamre, K.; Hjeltnes, B.; Kryvi, H.; Sandberg, S.; Lorentzen, M. and Lie, Ø.** (1994). Decreased concentration of hemoglobin, accumulation of lipid oxidation products and unchanged skeletal muscle in Atlantic salmon (*Salmo salar*) fed low dietary vitamin E. Fish Physiol. Biochem. 12, 421–429. https://doi.org/10.1007/BF00004306

**Haque, M.R.; Islam, M.A.; Wahab, M.A.; Hoq, M.E.; Rahman, M.M. and Azim, M.E**. (2016). Evaluation of production performance and profitability of hybrid red tilapia and genetically improved farmed tilapia (GIFT) strains in the carbon/nitrogen controlled periphyton-based (C/N- CP) on-farm prawn culture system in Bangladesh. Aquac. Reports 4, 101–111. https://doi.org/10.1016/j.aqrep.2016.07.004

Hastings, W.H. and Dickie, L.M., (1972). Feed Formulation And Evaluation, in: Fish

Nutrition. pp. 327–374. https://doi.org/10.1016/b978-0-12-319650-7.50012-2

**Hunt, A.; O., Ozkan, F. and Altun, T.** (2004). Effect of broodstock nutrition on reproductive performance of fish. . Turky. J. Aquat. Life, 2(3), 487-493. https://doi.org/10.1016/S0044-8486(01)00581-6

**Izquierdo, M. and Fernandez- Palacios, H.** (1997). Nutritional requirements of marine fish larvae and broodstock. Feed. tomorrow' s fish. Proc. Work. Ciheam Netw. Technol. Aquac. Mediterr. TECAM, jointly Organ. by Ciheam, FAO IEO, Maz. Spain, 24 26 June 1996. 243–264.

**Izquierdo, M.S.; Fernández-Palacios, H. and Tacon, A.G.J.** (2001). Effect of broodstock nutrition on reproductive performance of fish. Aquaculture 197, 25–42. https://doi.org/10.1016/S0044-8486(01)00581-6

**Khalil, D.K.; Ali, M.A.M. and Badran, M.F.** (2024). The Impact of Interaction of Photoperiods and Vitamins Supplementation on the Hematological Parameters, Reproductive Hormones, Liver Function, and Reproductive Performance of the Female Nile Tilapia (*Oreochromis niloticus*). Egypt. J. Aquat. Biol. Fish. 28, 825–842. https://doi.org/10.21608/ejabf.2024.351795

**Khara, H.; Sayyadborani, M. and Sayyad Borani, M.** (2016). Effects of α-Tocopherol (vitamin E) and ascorbic acid (Vitamin C) and their Combination on growth, survival and some haematological and immunological parameters of Caspian brown trout, salmo trutta caspius juveniles. Turkish J. Fish. Aquat. Sci. 16, 385–393. https://doi.org/10.4194/1303-2712-v16_2_18

**Lee, K.J. and Dabrowski, K.** (2004). Long-term effects and interactions of dietary vitamins C and E on growth and reproduction of yellow perch, Perca flavescens. Aquaculture 230, 377–389. https://doi.org/10.1016/S0044-8486(03)00421-6

**Lie, Ø.; Sandvin, A. and Waagbø, R.** (1994). Transport of alpha-tocopherol in Atlantic salmon (Salmo salar) during vitellogenesis. Fish Physiol. Biochem. 13, 241–247. https://doi.org/10.1007/BF00004362

**Lovshin, L.L.** (2000). Criteria for Selecting Nile Tilapia and Red Tilapia for Culture, in: Tilapia Aquaculture in the 21st Century, Proceedings of the Fifth International Symposium on Tilapia Aquaculture. pp. 49–57.

**Miller, G.W.; Labut, E.M.; Lebold, K.M.; Floeter, A.; Tanguay, R.L. and Traber, M.G.** (2012). Zebrafish (*Danio rerio*) fed vitamin E-deficient diets produce embryos with increased morphologic abnormalities and mortality. J. Nutr. Biochem. 23, 478–486. https://doi.org/10.1016/J.JNUTBIO.2011.02.002

**Mohamad, S.N.; Noordin, W.N.M.; Ismail, N.F. and Hamzah, A.** (2021). Red hybrid tilapia (Oreochromis spp.) broodstock development programme in malaysia: Status, challenges and prospects for future development. Asian Fish. Sci. 34, 73–81. https://doi.org/10.33997/j.afs.2021.34.1.008

**Mollah, M.F.A.; Sarder, M.R.I. and Begum, T.** (2003). Effects of different dietary levels of vitamin Eon the breeding performance of Heteropneustes fossilis (Bloch). Bangladesh J. Fish. Res. 7, 11–20.

**Mukhopadhyay, P.K.; Chattopadhyay, D.N. and Mitra, G.** (2003). Broodstock nutrition, the key to quality seed production. Infofish International, 3: 25-3. Infofish Int. 25–32.

**Nascimento, T.S.R.; de Stéfani, M.V.; Malheiros, E.B. and Koberstein, T.C.D.** (2014). High levels of dietary vitamin E improve the reproductive performance of female *Oreochromis niloticus*. Acta Sci. - Biol. Sci. 36, 19–26. https://doi.org/10.4025/actascibiolsci.v36i1.19830

**NRC** (1993). Nutrient requirements of Fish. 114.

**Obirikorang, K.A.; Agbo, N.W.; Obirikorang, C.; Adjei-Boateng, D.; Ahiave, S.E. and Skov, P.V**. (2019). Effects of water flow rates on growth and welfare of Nile tilapia (*Oreochromis niloticus*) reared in a recirculating aquaculture system. Aquac. Int. 27, 449–462. https://doi.org/10.1007/S10499-019-00342-0

**Palace, V.P. and Werner, J.** (2006). Vitamins A and E in the maternal diet influence egg quality and early life stage development in fish: A review. Sci. Mar. https://doi.org/10.3989/scimar.2006.70s241

**Pamungkas, W.; Tahapari, E. and Darmawan, J.** (2014). Gonadal Development And Spawning Frequency Of Tilapia (*Oreochromis niloticus*) That Feeded By Vitamin E Supplementation [Perkembangan Gonad dan Performa Pemijahan Induk Ikan Nila (Oreochromis Niloticus) yang diberi Pakan dengan Penambahan Vitamin E Supleme. Ber. Biol. 13, 239–244.

**Rudneva-Titova, I.I.** (1995). Change in the activity of antioxidant enzymes in the process of early ontogenesis of various species of Black Sea fishes | Izmenenie aktivnost' antioksidantnykh fermentov v protsesse rannego ontogeneza nekotorykh vidov chernomorskikh ryb. Ukr. Biokhim. Zh. 67, 92–95.

**Sallam, G.R.; Fayed, W.A.; El Absawy, M.A.; Ali, H.A. and El Greisy, Z.A.** (2017). Red Tilapia Broodstocks and Larval Production Under Different Water Salinities Without Acclimation. J. Aquac. Res. Dev. 08, 3. https://doi.org/10.4172/2155-9546.1000476

**Sutjaritvongsanon, S.** (1987). Level of vitamin E content suitable for gonad developing and spawning of goldfish, Carassius auratus (Linn.). Master Sci. Theses Fish. Sci. Notes Fac. Fish. Kasetsart Univ., Bangkok,.

**Syed, R.; Masood, Z.; Ul Hassan, H.; Khan, W.; Mushtaq, S.; Ali, A.; Gul, Y.; Jafari, H.; Habib, A.; Ishaq Ali Shah, M.; Gabol, K.; Gul, H. and Ullah, A.** (2021). Growth performance, haematological assessment and chemical composition of Nile tilapia, *Oreochromis niloticus* (Linnaeus, 1758) fed different levels of Aloe vera extract as feed additives in a closed aquaculture system. Saudi J. Biol. Sci. https://doi.org/10.1016/J.SJBS.2021.08.098

**Takeuchi, M.; Ishii, S. and 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. Labratory 104, 111–122.

**Tang, MU and Affandi, R.** (2004). Biology of reproduction fish., in: Research Center for Coastal and Aquatic Areas. Pekanbaru: Riau University. CRC Press, p. 110. https://doi.org/10.1201/9781439856000-12

**Tarigan, G.; Arthana, I.W. and Pebriani, D.A.A.** (2021). Effect of Giving Different Doses of Vitamin E in Feed to the Level of Gonad Maturity of Tilapia (*Oreochromis niloticus*). Adv. Trop. Biodivers. Environ. Sci. 5, 64. https://doi.org/10.24843/atbes.2021.v05.i02.p05

**Tayamen, M.M.; Reyes, R.A.; Danting, M.J.; Mendoza, A.M.; Marquez, E.B.; Salguet, A.C.; Gonzales, R.C.; Abella, T.A. and Vera-Cruz, E.M.** (2002). Tilapia Broodstock Development for Saline Waters in the Philippines, in: The ICLARM Quarterly. The WorldFish Center, pp. 32–38.

**Topic Popovic, N.; Strunjak-Perovic, I.; Coz-Rakovac, R.; Barisic, J.; Jadan, M., Persin Berakovic, A. and Sauerborn Klobucar, R.** (2012). Tricaine methane-sulfonate (MS-222) application in fish anaesthesia. J. Appl. Ichthyol. https://doi.org/10.1111/j.1439-0426.2012.01950.x

**Volkoff, H. and London, S.** (2018). Nutrition and reproduction in fish, in: Encyclopedia of Reproduction. Elsevier, pp. 743–748. https://doi.org/10.1016/B978-0-12-809633-8.20624-9

**Watanabe, T.** (1985). Importance of the study of broodstock nutrition for further development of aquaculture, in: Nutrition and Feeding in Fish. pp. 395–414.

**Zaki, F.M.; Said, M.M.; Tahoun, A.A. and Amer, M.** (2021). Evaluation of Different Sex Reversal Treatments in Red Tilapia Hybrid. Egypt. J. Aquat. Biol. Fish. 25, 279–292. https://doi.org/10.21608/EJABF.2021.140630

**Zhang, G.; He, R.; Zhang, S.; Cao, K. and Gao, H.** (2007). Effect of vitamin E in broodstock diet on reproductive performance of Monopterus albus. Acta Hydrobiol. Sin. 02, 196–200.