

## RESEARCH ARTICLE

# Shading of Ponds Improves the Reproductive Performance of Female Nile Tilapia (*Oreochromis niloticus* L.) Breeders during Warm Months

Xyrra Jeremiah C. Mazo<sup>1\*</sup>  and Emmanuel M. Vera Cruz<sup>2</sup>

<sup>1</sup>Romblon State University-San Agustin Campus, Cabolutan, San Agustin, Romblon, Philippines

<sup>2</sup>College of Fisheries, Central Luzon State University, Science City of Munoz, Nueva Ecija, Philippines

### ABSTRACT

One of the major challenges facing the tilapia farming industry is the production of sufficient amount of quality seeds during warm months. Extreme temperatures brought about by global warming affects the reproduction of fish. Hence, this study evaluated aquashade technology as a possible solution to the problem on low seed production during warm months. The effectiveness of black greenhouse nets with different shading capacities of (i.e. 40% and 80%), as top cover of breeding ponds was investigated on the reproductive performance of female Nile tilapia breeder. Results of the study showed that shading of ponds reduced the average water temperature by 1 - 2 °C at 0700 h and 3-6 °C at 1100 h to 1500 h. The lower water temperature on shaded pond improved the gonadal development and maturation on female breeders, thus promoting better gonadosomatic index value and pattern. No significant differences were recorded on the spawning rate and seed production per female among the shaded treatments, with 40% shading capacity treatment having the numerically higher value. The application of aquashade can eliminate the deleterious effect of extreme water temperature on the reproduction of breeders and is therefore effective in improving the reproductive performance of female Nile tilapia during warm months.

\*Corresponding Author: xyrrajeremiah@gmail.com  
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## 1. INTRODUCTION

Nile tilapia (*Oreochromis niloticus*) is one of the most productive and internationally traded food fish in the world (Yakubu et al. 2014). The ideal attributes of Nile tilapia and its relatively high nutritional value, good taste and low cost make it an excellent candidate for aquaculture (Mjoun et al. 2010; El-Sayed 2013). The tilapia industry has become an important sector in terms of food security and provision of income and employment (Liu et al. 2013). According to the Food and Agriculture Organization (2020), the increasing global production of Nile tilapia makes it the third major finfish species produced in world aquaculture in 2018, following grass carp, and silver carp. However, as opposed to the increasing global production of tilapia, the Philippines reported a declining production trend (Guerrero III 2019). From the fisheries data of the Philippine Statistics Authority

and Bureau of Fisheries and Aquatic Resources, a total of 240% increase in the production of tilapia was recorded from 2001 to 2011, however, from 2007 to 2016, a significantly lower increase of only 7% was reported (Guerrero III 2019). This decline was associated with several factors of which increasing temperature in water bodies is one (Guerrero III 2019).

Over the past few years, rising global temperatures have received much attention because of its worldwide impact on ecosystems. This phenomenon of rising global temperature, which result to warming of water bodies, sea-level rise, ocean acidification, weather pattern changes, and extreme weather events, has directly and indirectly affected the aquaculture sector (Cochrane et al. 2009). Fish being a coldblooded animal, adapts to the temperature of the surrounding water and thus is adversely affected by climate-induced temperature variability which

influences its growth rates (Azaza et al. 2008; Pandit and Nakamura 2010), food intake, food conversion efficiency (Pandit and Nakamura 2010; Khater et al. 2017), disease susceptibility (Karvonen et al. 2010), timing of spawning (Pankhurst and King 2010), egg production (Faruk et al. 2012) and mortality at certain life-cycle stages (Pandit and Nakamura 2010; Pankhurst and Munday 2011). During cold months, a small increase in water temperature produces a positive effect on fish such as enhanced growth, better appetite and protein synthesis, and improved oxygen consumption. However, the same increase during warm seasons affect the fish negatively (Morgan et al. 2001). Long term exposure to high temperature forces fish to respond with stress-related reactions which sacrifices other biological functions such reproduction or immunity (Sadoul and Vijayan 2016; Alfonso et al. 2021). Tilapia seed production during extremely warm months is restricted by thermal-induced stress which affects the reproductive capability of fish (Pankhurst and Munday 2011) and results to lower seed production during this season (Vera Cruz, Jimenez and Bartolome 2023).

Hence, mitigating measures on the adverse effect of unusual increase in water temperatures that causes low seed production must be carried out. Aquashade technology or the shading of fish rearing and/or breeding units using greenhouse nets, can be a possible solution. The application of shading reduces the light penetration on the water body and thus reduces its temperature. Studies showed that shading of rearing containment have resulted to a positive effect on the growth of some species of fish (Huse et al. 1990; Buentello et al. 2000). Thus, this study investigated the effectiveness of using greenhouse nets with varying pond shading capacities, on the breeding performance of Nile tilapia during extremely warm months.

## 2. MATERIALS AND METHODS

### 2.1 Experimental fish

Freshwater Aquaculture Center Selected Tilapia (FaST) strain breeders weighing from 110.00 – 170.00 g, or with a mean weight of  $135.00 \pm 26.00$  g., were used in the study. The fish were six months old and were first time spawners. The stocks were acquired from the Freshwater Aquaculture Center (FAC), Central Luzon State University, Science City of Muñoz, Nueva Ecija, Philippines.

### 2.2 Experimental lay-out and design

Three earthen ponds at FAC, each with an area of 200 m<sup>2</sup>, were used in the study. Two ponds were installed with a steel frame where the top portion was covered with black greenhouse shading nets having the shading capacities of 40% and 80%, respectively. Each pond set-up represented each treatment. The treatments were: T1 - unshaded pond (or No S), T2 - 40% shading capacity (40% S), and T3 - 80% shading capacity (80% S). The depth of water in each pond was one meter and the nets were placed three meters above the water surface. Triplicate hapa nets measuring 2 x 1 x 1m<sup>3</sup> were used as breeding enclosures of fish in each pond. Additional three hapa nets were set up for fish samples where reproductive parameters such as gonadosomatic index etc. were recorded.

The tilapia breeders were randomly distributed to each treatment. Conditioning of fish was done under the assigned treatments for 10 days. After conditioning, eight breeders were randomly allotted to each breeding hapa at a ratio of 3 females: 1 male. Fish were fed daily with commercial feeds containing 31% crude protein at 2% of their body weight during the conditioning phase and the and 14 days of the breeding period. The fish were fed thrice daily at 0800h, 1100h and 1500h.

### 2.3 Water quality monitoring

Water temperature and dissolved oxygen were monitored every other day between 0600-0700 h, 1100-1200 h and 1500-1600 h using DO meter.

### 2.4 Gonadosomatic index (GSI) estimation

The The GSI of female Nile tilapia breeders were evaluated at day 0, day 5 and day 10 of the breeding period to determine gonadal changes in the breeders. Since breeders had to be sacrificed for GSI classification, four female breeders per treatment were sampled from the additional hapa nets installed in each pond during the aforementioned days. The following formula was used for GSI determination:

$$\text{GSI} = (\text{gonad mass} / \text{total body mass}) \times 100\%$$

### 2.5 Characterization of stages of gonadal maturation

The The gonadal maturity of each breeder was noted based on the appearance, size and color

of the gonads as viewed under a stereomicroscope. The gonads were characterized using the criteria for different stages of gonadal development of Nile tilapia by Mous et al. (1995) (Table 1).

Table 1. Criteria on the different stages of gonadal development.

Maturity Stage	Description
Stage I (Immature)	Appearance like testes; sexes indistinguishable macroscopically
Stage II (Early Developing)	Ovaries to be recognized by small whitish dots (eggs); caudal part of the ovaries more thickened than the rostral part
Stage III (Developing or Recovering)	Eggs developing inside the ovaries unequal in size
Stage IV (Early Ripening)	Eggs equal in size but not fully grown; all coloured yellow
Stage V (Ripe)	Eggs large and ovaries visible from the ventral side of the cavity
Stage VI (Spent)	Eggs or juveniles in the buccal cavity, ovaries recovering, thin and reddish; eggs unequal in size, often including a few residual stage V eggs

### 2.6 Breeding performance evaluation

Breeding performance was evaluated based on the spawning rate and seed production capacity of each female breeder. Eggs and fry were collected 14 days from the time of stocking. Fry from the hapa were counted while unhatched eggs from the mouth of breeders were removed carefully and were artificially incubated in an improvised downwelling incubation

jar with continuous water supply. After hatching, the fry were counted and added to the previously counted fry. The following computations were used in the study:

$$\text{Spawning rate} = \left( \frac{\text{Number of females that spawned}}{\text{Number of stocked female}} \right) \times 100\%$$

$$\text{Seed production per female} = \frac{\text{Total seed production}}{\text{Number of females that spawned}}$$

### 2.7 Statistical analysis

Data were analyzed using One-way Analysis of Variance (ANOVA) and means were compared using Least Significant Difference (LSD) test. Meanwhile, percentage data on spawning rate and count data on produced fry were transformed first to their arcsine and square root values, respectively before subjecting them to analysis. The relationship of GSI, spawning rate and seed production to water temperature were analyzed using multiple Pearson correlation. All statistical analyses were done using IBM SPSS Statistics 23.

## 3. RESULTS

### 3.1 Water temperature and dissolved oxygen levels

Mean temperature at each treatment were significantly different ( $P < 0.01$ ) at different times of the day (Table 2). Highest water temperature of 35.67 °C ( $\pm 0.49$ ) was recorded in the unshaded treatment

Table 2. Water temperature (°C) and dissolved oxygen (mg/l) in each treatment.

Time	Shading Capacity of Net			
	No S (n=48)	40% S (n=48)	80% S (n=48)	
Temperature	0700h	29.51 $\pm$ 0.51 <sup>a</sup>	28.17 $\pm$ 0.52 <sup>b</sup>	27.29 $\pm$ 0.50 <sup>c</sup>
	1100h	34.38 $\pm$ 0.57 <sup>a</sup>	31.00 $\pm$ 0.49 <sup>b</sup>	29.11 $\pm$ 0.35 <sup>c</sup>
	1500h	35.67 $\pm$ 0.49 <sup>a</sup>	31.69 $\pm$ 0.65 <sup>b</sup>	29.93 $\pm$ 0.40 <sup>c</sup>
Dissolved Oxygen	0700h	1.90 $\pm$ 1.20 <sup>a</sup>	0.96 $\pm$ 0.17 <sup>b</sup>	2.13 $\pm$ 0.69 <sup>a</sup>
	1100h	7.21 $\pm$ 1.94 <sup>a</sup>	3.31 $\pm$ 0.51 <sup>b</sup>	4.40 $\pm$ 1.29 <sup>b</sup>
	1500h	8.28 $\pm$ 1.96 <sup>a</sup>	5.44 $\pm$ 0.76 <sup>b</sup>	5.86 $\pm$ 1.55 <sup>b</sup>

Means in a row superscripted with different letters are significantly different at 5% level ( $P < 0.05$ )

(No S) with an increase of about 6.00 °C from 0700 h to 1500 h. Meanwhile, lower water temperature was recorded in the shaded treatments, with 31.69 °C ( $\pm 0.65$ ) in 40% S and 29.93 °C ( $\pm 0.40$ ) in 80% S at the hottest time of the day. The use of shading nets lowered the water temperature by 3.00 – 4.00 °C under 40% S, and 5.00 – 6.00 °C under 80% S compared to No S.

On the other hand, significantly higher DO levels were recorded in No S and 80% S at 0700 h ( $P < 0.05$ ). DO level in No S continued to rise as the day progressed, obtaining a significantly higher DO level at 1100 h and 1500 h ( $P < 0.01$ ). However, a notable decline in the DO level of more than 6.00 mg/l was observed overnight in this treatment.

Table 3. Length and weight of gonads, and gonadosomatic index of breeders in each treatment.

		Shading Capacity of Net		
		No S (n=4)	40% s (n=4)	80% s (n=4)
Day-0	Length (cm)	3.12	4.33	3.92
	Weight (g)	1.27	4.67	3.70
	GSI (%)	1.49 ± 0.22 <sup>a</sup>	3.88 ± 0.43 <sup>b</sup>	3.28 ± 1.01 <sup>b</sup>
Day-5	Length (cm)	4.33	4.97	4.58
	Weight (g)	3.67	4.77	2.60
	GSI (%)	2.58 ± 0.09 <sup>a</sup>	3.32 ± 1.33 <sup>a</sup>	1.90 ± 0.31 <sup>a</sup>
Day-10	Length (cm)	4.13	4.47	4.17
	Weight (g)	3.57	3.67	3.40
	GSI (%)	3.22 ± 0.96 <sup>a</sup>	2.29 ± 0.50 <sup>a</sup>	2.84 ± 1.37 <sup>a</sup>

Means in a row superscripted with different letters are significantly different at 5% level ( $P < 0.05$ ).

### 3.2 Gonadal development and maturation of female Nile tilapia breeders

After the 10-day conditioning period and before breeding (Day-0), significantly higher GSI values were obtained from female breeders at 40% S and 80% S with 3.88% ( $\pm 0.43$ ) and 3.28% ( $\pm 1.01$ ), than those under No S with 1.49% ( $\pm 0.22$ ) ( $P < 0.01$ ) (Table 3). However, no significant difference among GSI of each treatment was obtained at Day-5 ( $P = 0.17$ ) and Day-10 ( $P = 0.56$ ) of the breeding period.

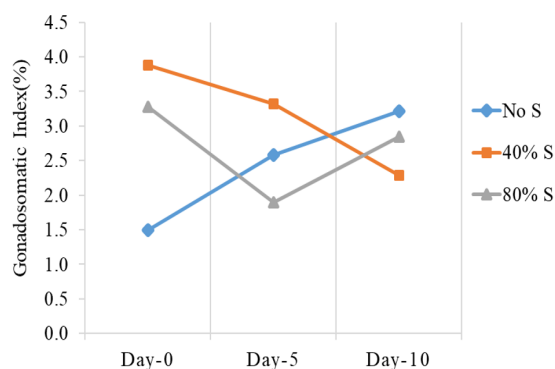


Figure 1. Pattern of gonadosomatic index of breeders in each treatment prior breeding, at Day 5 and Day 10 of breeding period

Moreover, different trends in the GSI values were also observed among treatments (Figure 1). An increasing trend was observed in the GSI of breeders under No S with 1.49% ( $\pm 0.22$ ) at Day-0, 2.58% ( $\pm 0.09$ ) at Day-5 and 3.22% ( $\pm 0.96$ ) at Day-10. This was in contrast to the decreasing pattern of GSI obtained from breeders in the 40% S with 3.88% ( $\pm 0.43$ ) at day 0, 3.32% ( $\pm 1.33$ ) at day 5 and 2.29% ( $\pm 0.50$ ) at day

10. Meanwhile, GSI of breeders in 80% S showed a decreasing trend from day 0 to day 5 with 3.28% ( $\pm 1.01$ ) to 1.90% ( $\pm 0.31$ ) and an increasing pattern at the latter part of the breeding period with 2.84% ( $\pm 1.37$ ).

Results of the gonadal maturation characterization showed the presence of stage III to stage VI gonads (Figure 2) in the stocks examined. Female breeders in No S showed Stages III, IV and V gonads which were light to dark yellow in color, shorter and contained distinctly unequally sized ova. Meanwhile, more advanced stages of IV and V gonads were obtained from breeders in the 40% S treatment through-out the breeding period. These gonads were light to dark yellow, longer and had more uniformly-sized big eggs. Furthermore, breeders under 80% S showed Stages III, IV, V and VI gonads which were also light to dark yellow, and longer than No S but shorter than 40% S. Some gonads have unequal sized ova while some have uniformly sized ova. Stage VI gonads were recorded at day 5 in this treatment.

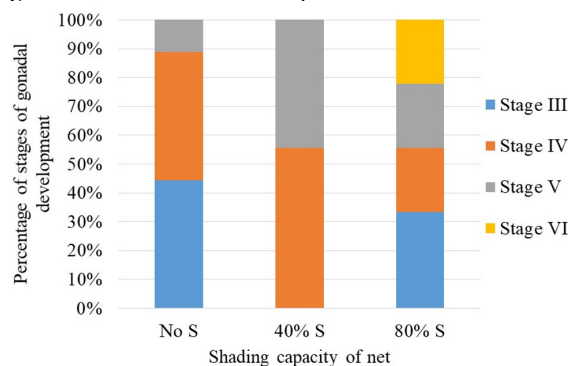


Figure 2. Percentage of stages of gonadal development under the different treatments

### 3.3 Breeding performance of the fish

No spawning occurred in the No S breeders. Meanwhile, no significant differences were obtained on the spawning rate ( $P= 0.74$ ) and seed production per female breeders ( $P= 0.81$ ) under the shaded treatments (Table 4). A higher spawning rate was recorded in 40% S with 75.24% ( $\pm 18.95$ ) than those in 80% S with 63.94% ( $\pm 30.68$ ). In terms of seed production per female breeder, breeders in 40% S had the higher seed production with an average of 560.00 ( $\pm 62.00$ ) fry per female breeder than those in 80% S with an average of 481.00 ( $\pm 160.00$ ) fry per female breeder.

Table 4. Spawning rate and seed production per female breeders among treatments.

	Shading Capacity of Net		
	No S (n=3)	40% S (n=3)	80% S (n=3)
Spawning Rate	0.00 $\pm$ 0.00 <sup>b</sup>	75.24 $\pm$ 18.95 <sup>a</sup>	63.94 $\pm$ 30.68 <sup>a</sup>
Seed Production per Female	0.00 $\pm$ 0.00 <sup>b</sup>	560.00 $\pm$ 62.00 <sup>a</sup>	481.00 $\pm$ 160.00 <sup>a</sup>

### 3.4 Correlation of water temperature with the gonadosomatic index, spawning rate and seed production of female breeders

Results of the analysis revealed a significantly strong positive relationship ( $r= 0.78$ ,  $P= 0.00$ ) between the water temperature and the dissolved oxygen of pond water. Meanwhile, water temperature and gonadosomatic index showed significantly strong negative relationship ( $r= -0.76$ ,  $P= 0.02$ ) at day 0, a positive but not significant relationship at day 5 ( $r= 0.16$ ,  $P= 0.69$ ) and also a positive but not significant relationship ( $r= 0.25$ ,  $P= 0.53$ ) at day 10 of the breeding period.

Furthermore, water temperature showed a significantly strong negative relationship with spawning rate ( $r= -0.73$ ,  $P= 0.02$ ), and seed production ( $r= -0.78$ ,  $P= 0.01$ ), while dissolved oxygen also showed a significantly strong negative relationship with spawning rate ( $r= -0.74$ ,  $P= 0.02$ ), and seed production ( $r= -0.77$ ,  $P= 0.02$ ).

## 4. DISCUSSION

The increasing global temperature has posed a threat to the aquatic environment, increasing the temperature of the water and affecting various physiological processes of fish. Several studies have

shown that increasing water temperatures within the fish's temperature tolerance range improves its growth (Azaza et al. 2008; Pandit and Nakamura 2010; Faruk et al. 2012; Santos et al. 2013; Makori et al. 2017; Koyakoma et al. 2019), but similar increase have different effects on its reproduction (Faruk et al. 2012). Water temperatures of 28.0 – 32.0 °C were found effective for increasing the growth of Nile tilapia (Pandit and Nakamura 2010), while lower temperatures of 25.0 – 28.0 °C were suggested for optimum egg production of Nile tilapia (Faruk et al. 2012). Moreover, Mahmoud et al. (2020) suggested a slightly higher temperature range for reproduction which was up to 30.0 °C. Beyond this point, an inhibitory effect on the reproduction can be observed. Studies have shown that molecular analysis showed inhibition in the expression of certain reproductive genes, such as steroidogenic acute regulatory protein (STAR) and vitellogenin gene (Mahmoud et al., 2020). Moreover, in the same instances, the damaged ovarian tissues were observed (Mahmoud et al., 2020). With the effect of extreme global warming episodes and the projected increase in the coming years, the aquaculture sector is greatly challenged. Thus, to address the negative effect of extremely high temperature on the reproduction of Nile tilapia, the present study evaluated aquashade technology or the shading of ponds using greenhouse nets to lessen the impact of high temperatures, maintain the desirable water temperature for reproduction and improve the seed production of Nile tilapia during warm months. Shading capacities of 40% and 80% were evaluated on its effect on the reproductive performance of female *O. niloticus*.

The present study showed that shading was effective in reducing the water temperature of ponds, however, only the highest shading capacity of 80% maintained the temperature of not greater than 30.0 °C. The black net shade serves as a neutral filter with almost no change in shade factor from 300-1100 nm (Oren-Shamir et al. 2001), so most wavelengths of light were trapped on the surface of the net inhibiting the radiation and heat to be transmitted below. Moreover, water temperature on the unshaded treatment reached almost 36.0 °C, a critical condition for the fish which have resulted to the absence of spawning and reproduction. In addition, extreme temperatures higher than 32.0 °C were found highly stressful to fish, severely affecting their growth performance, altering their haemato-biochemical parameters and increasing mortality in Nile tilapia (Islam et al. 2020; Pandit and Nakamura 2010).

Dissolved oxygen (DO) is also an important water quality parameter in the reproduction of fish. Kolding et al. (2008) showed that increasing DO affects the reproduction of Nile tilapia positively. However, in the present study, the lower DO level recorded from the shaded ponds compared to the unshaded pond were in agreement with the results of the studies of Memis et al. (2011) and Vera Cruz, Jimenez and Bartolome (2023). The dissolved oxygen of water is primarily affected by the photosynthetic and respiration activities of phytoplankton in the pond (Steel 1980) and these organisms are temperature-dependent (Boyd 1998). The direct sunlight on the unshaded pond which increased the temperature of water might have increased the number of the phytoplankton and favored higher rate of photosynthetic activity which then resulted to the high increase of DO in this treatment. However, respiration of the relatively high biomass of plankton, fish and bacteria in this treatment caused a greater decline in the DO level at night than in bodies of water with lesser biomass of plankton and bacteria (Boyd and Lichtkoppler 1979; Kunlasak et al. 2013). A high temperature paired with declining oxygen in water is stressful to fish (Cech and Brauner 2011). The shading of pond prevents this condition, with having an optimum DO level during the day and lesser reduction at night, especially in ponds under higher shading capacity of net. On the other hand, Dayrit et al. (2023) noted that average water temperature was considerably decreased in shaded ponds but with no significant decrease in the DO concentrations. The absence of significant decrease in DO concentration in the shaded ponds maybe due to the lesser level of decrease in pond water temperature of only 1.25 °C since the study was done during the rainy season (May to August).

After the conditioning period, the significantly lower GSI value obtained from the breeders in the unshaded pond (1.49%) indicates that high water temperature range of up to 36.0 °C have negatively affected the gonadal development of fish. The highest GSI value of 2.84% obtained in this treatment towards the end of the breeding period were lower than the GSI values obtained from the studies of Babiker and Ibrahim (1979) with 3.58%, Bakhoum et al. (2002) with 3.25%, Teame et al. (2018) with 3.50%, and Shoko et al. (2015) with 3.86 - 4.33%. The higher percentage of Stage III and Stage IV gonads obtained from the breeders in the unshaded treatment also suggests that the very high temperature in this treatment affected the gonadal maturity of breeders. This observation is supported by the findings of

Mahmoud et al. (2020) wherein Nile tilapia subjected to heat treatment of temperatures 34.0 – 37.0 °C revealed a decreased estradiol - an estrogen steroid hormone, lower serum vitellogenin, down-regulated STAR gene and rendered injuries on ovary tissues of female fish. Additionally, germ cells in the developing ovary of fish are heat sensitive and disappear under high temperatures affecting the gonadal growth of fish (Strussmann et al. 1998). On the other hand, the significantly higher GSI value obtained from the breeders in the shading capacities of 40% (3.88%) and 80% (3.84%) after the conditioning period, and the higher percentage of Stage IV, Stage V, and Stage VI gonads on these treatments, indicate that these shading capacities were effective in improving the gonadal development and maturity of fish. These GSI values were also comparable with the GSI values reported from the above-mentioned studies.

Furthermore, the significantly strong negative relationship between water temperature and GSI at Day-0 denotes that water temperatures during the conditioning period of fish has a significant effect on the gonad development of breeders. The stress experienced by the breeders due to the extreme temperature during their reproductive development may have caused delayed ovulation, phase shifting of spawning or complete inhibition of reproduction (Campbell et al. 1992; Contreras-Sánchez et al. 1998). These effects depend on the period of exposure and amplitude of increase in temperature (Pankhurst and Munday 2011). According to Pankhurst and Munday (2011), extreme temperature affects the endocrine system of fish, particularly through the inhibition of ovarian estrogen production. Exposure of fish to temperatures beyond their normal range impairs the gonadal steroid synthesis and hepatic vitellogenin production, alters hepatic estrogen receptor dynamics, which then results to the delay or inhibition of the preovulatory shift from androgen to maturation-inducing steroid production (Pankhurst and King 2010). The inhibitory effect on gonadal development was shown in breeders in the unshaded treatment. This was indicated by the continued increase in GSI value and high proportion of Stages III and IV gonad until the Day-10 of breeding period. A similar result was recorded by Koyokamanda et al. (2019), wherein Mozambique tilapia (*O. mossambicus*) exposed to 34.0 – 36.0 °C caused physical and physiological abnormalities, leading to an inhibitory effect on both gonadal and somatic development on exposed fish.

Some studies have also shown that shading of rearing containment has resulted to positive effects on

some fish species. Enhanced growth was recorded on the shaded pen rearing of Atlantic salmon (*Salmo salar*) during summer and early autumn (Huse et al. 1990) while higher feed conversion ratio was obtained from a Russian sturgeon (*Acipenser gueldenstaedtii*) reared under shaded pond (Memis et al. 2011). In the present study, results on the spawning rate and seed production of fish indicate that shading of pond during warm months improves the breeding performance of fish. However, the nonsignificant difference in spawning rate and seed production of breeders in the shaded treatments indicates that the breeding performance was not affected by the different shading capacities of net. The spawning rates recorded in this study were higher than those noted by Manliclic and Vera Cruz (2017) with 27-50%, and Vera Cruz et al. (2020) with 22-39%, and were comparable with the studies of El-Sayed and Kawanna (2008) with 50-100% spawning rate. Meanwhile the recorded seed production per female fish in the study were comparable with the studies of Manliclic and Vera Cruz (2017) with 366-511 number of fry, and Vera Cruz et al. (2020) with 335-796 number of fry. In the present study, although a slightly higher temperature of 32.0 °C was recorded in 40% S, this shading capacity resulted to higher GSI value, higher spawning rate and higher seed production per female breeder that spawned than the other treatments. Although Mahmoud et al. (2020) suggested 30.0 °C as the most desirable temperature for living and reproduction of Nile tilapia, results of this study revealed that temperatures of up to 32.0 °C still showed some positive results on the reproduction of fish. According to Mahmoud et al. (2020), increase in estradiol, increased levels of serum vitellogenin, and up-regulated STAR gene were recorded in fish reared in water temperatures up to 32.0 °C. However, this temperature may still result to significantly fewer germ cells (Alvarenga and Franca 2009; Pandit et al. 2015; Mahmoud et al. 2020).

Finally, the significantly strong negative relationship between water temperature and spawning rate, and water temperature and seed production indicates that increasing water temperature decreases the capability of female breeders to spawn and produce seed. The extreme temperatures of up to 36.0 °C in the unshaded treatment showed inhibitory effect on the gonadal development of fish that greatly affected the reproduction of fish resulting to no fry being produced. Water temperature in the range of 34.0 – 37.0 °C affects the growth performance and alters haemato-biochemical parameters of Nile tilapia (Islam et al. 2020). An increase in white blood

cells, mediating antibody response to the stressful environment was also observed in the fish (Islam et al. 2020; Shahjahan et al. 2018). Fish exposed to these high temperatures showed damaged ovaries, the follicular atresia showed separated granulosa cells from oocyte membranes, reduction of yolk materials deposited between follicles, and destroyed germline stem cells (Mahmoud et al. 2020; Pandit et al. 2015), posing deleterious effect on the gonadal development and reproduction of fish.

## 5. CONCLUSION

One of the major challenges facing the tilapia farming industry is the production of sufficient amount of quality seeds during warm months. The extreme water temperatures of up to 36.0 °C brought about by global warming adversely affects reproduction of Nile tilapia. Shading of pond using greenhouse net with shading capacities of 40% and 80% reduces the water temperature by 3.0 - 6.0 °C during the hottest time of the day, improves the gonad development and maturation, and eliminates the deleterious effect of extreme temperature on the reproduction of Nile tilapia. For both technical and economic considerations, the use of net with 40% shading capacity is recommended to improve the reproductive performance of Nile tilapia during warm months.

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## AUTHOR CONTRIBUTIONS

**Xyrra Jeremiah C. Mazo:** Conceptualization, Data collection, Data processing, Data interpretation, Manuscript write-up, Editing; **Emmanuel M. Vera Cruz:** Conceptualization, Data interpretation, Manuscript write-up, Review, Editing.

## CONFLICTS OF INTEREST

The authors have no known conflict of interest associated with this publication.

## ETHICS STATEMENT

The study was conducted in accordance with the Animal Care and Ethics Committee, Central Luzon State University, Philippines.

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