



## Effect of pawpaw (*Carica papaya*) seeds meal on the reproductive performance and histological characters of gonads in Nile tilapia (*Oreochromis niloticus*)

## KEYWORDS

Nile tilapia, pawpaw seeds, antifertility, monosex.

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**ABSTRACT** This study performed to examine the ability of pawpaw (*Carica papaya*) seeds to induce sterility in male and female of Nile tilapia (*Oreochromis niloticus*). Five groups were prepared; negative control group with mixed sex fish (G1) and positive control with monosex fish (G2) fed on basal diet, while G3, G4 and G5 were mixed sex fish fed on basal diet with addition of 60, 90, 120g of *C. papaya* seeds/kg diet, respectively. Ten fish with average weight of 30±1g were stocked at each of fifteen tanks (triplicates/treatment) with dimensions 135×50×60cm. Gonado-somatic index (GSI), egg diameter, fecundity and plasma sexual hormones level were determined and histological examination of gonads were performed. GSI, egg diameter and fecundity of treated females decreased significantly ( $P \leq 0.05$ ) after treated and recovery months, while GSI of males was not significantly different ( $P \geq 0.05$ ) among treatments. The levels of testosterone (T) and estradiol (E2) significantly ( $P \leq 0.05$ ) decreased after treated and recovery months. All values decreased gradually with increasing the dose of *C. papaya* seeds. Histological sections showed several structural changes in testes and ovaries of the treated groups with different degree according to *C. papaya* seeds dose. Sections after recovery month showed that *C. papaya* seeds induced permanent sterility in the high dose while the low and medium doses may have reversible effect. The study concluded that, the seeds of *C. papaya* can be used with adjusted amount as a feed additive to control the problem of precocious maturity and breeding of *O. niloticus* in growing ponds instead of unfavorable and expensive hormone.

### Introduction

*Oreochromis niloticus* is one of the most important fishes in the aquaculture, particularly for the lesser-developed countries in the tropics (FAO, 2001). It is characterized by short reproductive cycles, easy spawning, rapid growth, high feed conversion, high tolerance to environmental changes and eating a wide range of natural food organisms or cheap artificial foods (Coward and Little, 2001; Abdelhadi, 2011). It is also characterized by firm flesh texture, neutral flavor and marketable request (Young and Muir, 2002). Despite all of aforementioned merits, tilapias have a serious problem; precocious maturity and uncontrolled reproduction which often result in the overpopulation of production system with young (stunted) fish where they sexually mature at about 20g weight (Mair and Little, 1991). This uncontrolled reproduction of tilapia in culture system leads to low marketable-sized fish.

For profitable culture, various methods were conducted for the control of prolific breeding in tilapia and reduction of variation in the size of harvested fish. These methods include monosex culture, sex reversal, cage/tank culture, use of predators, high stocking density, sterilization, intermittent/selective harvesting and use of slow maturing tilapia species (Mair and Little, 1991; Beardmore, 1996 and Fagbenro, 2002). The development of hormonal sex-reversal techniques is a major breakthrough allowed male monosex populations to be raised to uniform marketable sizes. However, each of these methods has its own shortcomings, even hormonal sex-reversal techniques. Hence there is a need to search for a better solution to control this undesirable tilapia recruitment in culture system. Using medicinal plants as natu-

ral reproduction inhibitors is a new trend that may offer a solution for this problem. Egypt is one of subtropical-temperate countries where *Carica papaya* available all year round. *C. papaya* seeds contain many active ingredients such as cariacin, carpasemine enzyme, plant growth inhibitor and oleanolic glycoside which had been found to cause sterility in male rats (Das, 1980 and Kobayashi *et al.*, 2008). *C. papaya* was used to control the reproduction of male albino rats (Udoh *et al.*, 2005a and b) and to control the prolific breeding of *O. niloticus* (Ekanem and Okoronkwo, 2003 and Ayotunde and Ofem, 2008). The present study aimed to elucidate the effect of *C. papaya* seeds on the fertility of males and females in *O. niloticus*, by examining the histomorphological alternation of testis and ovaries, estimating gonadosomatic index, ova diameter and absolute fecundity as well as analysis the level plasma testosterone in males and estradiol in females.

### Materials and Methods

#### Fish aquaria:

*O. niloticus* were stocked in 15 rectangular glass tanks, with dimensions of 135×50×60 cm (L×W×H), filled with dechlorinated water and equipped with automatic mechanical filtration and pump. Water temperature was adjusted daily at 30±1°C using heater. The criteria of water used in the experiment were measured weekly. pH was measured by a pocket pH meter (Orion 210) after calibration by two standard buffer solutions (pH: 7 and 9). Total alkalinity was measured by titration with 0.02 N H<sub>2</sub>SO<sub>4</sub> (Snoeyink and Jenkins, 1980), and hardness by titration with EDTA (SMEWW, 1998). Total ammonia nitrogen (TAN: NH<sub>3</sub> and NH<sub>4</sub>), nitrite (NO<sub>2</sub>) and nitrate (NO<sub>3</sub>) were determined using colorimetric estimation of com-

mercial kits (API® freshwater master test kit). The unionized ammonia (UIA) concentration was estimated by multiplying the TAN value by the factor that was calculated by Emerson *et al.* (1975).

### Experimental design

The ingredients of basal diet used in this experiment were represented in Table 1. Seeds of *C. papaya*, were shade dried, milled into fine particle size (<250 µm) then added to the basal diet with three different concentrations (60, 90 and 120 g of *C. papaya* seeds/kg). Sufficient amount of water was added to the diet and mixed till obtaining stiff dough which straighter on a tray, cut to pieces and put in oven at 50°C for about 24h to get dry pieces of food, then packed in plastic container at room temperature.

Fish were divided into five groups; G1 was mixed sex fish fed on basal diet (negative control), G2 was monosex fish fed on basal diet (positive control), G3, G4 and G5 were mixed sex fish fed on basal diet with adding 60 (low dose), 90 (medium dose) and 120 g of *C. papaya* seeds/kg (high dose treatment), respectively.

After two weeks of acclimatization on the experimental diets and laboratory conditions, ten apparently healthy fish with average body weight of about (30±1)g were stocked in each tank (triplicate/treatment). The fish fed six days a week on 3% of their body weight. The daily diet was divided into two equal portions given at 10:00 am and 3:00 pm (NRC, 1993). The experiment was conducted for two months; the 1<sup>st</sup> month was to test the effect of *C. papaya* seeds on the fertility (treated period) and the 2nd month to test if fish can recuperation its fertility (recovery period).

**Table (1): Ingredients composition (%) of the experimental basal diet**

Ingredients	(%)
Fish meal (Morocco)	5.5
Soybean meal	43.3
Wheat flour	28.3
Corn gluten	13.8
Soya oil	3.3
Calcium biphosphate,	3.3
Sodium chloride	0.55
Anti mycotoxins	0.3
Active yeast	0.3
Vitamins and Minerals premix	1.35
Total	100

### Reproductive performance

Spawning of fish in all aquaria was observed daily, three males and females were examined in each treatment at the end of treated and recovery months. Specimens were carefully netted and handled, then total body weight was measured to the nearest 0.1gm. Gonads were removed and weighed to the nearest 0.001gm, then preserved in 10% neutralized formalin solution for further investigation.

- Gonado-somatic index (GSI) was calculated following the equation of Berhaut (1973)

$$GSI = (GW/BW) \times 100$$

Where, GW: wet weight of gonad to the nearest 0.001gm

BW: wet total body weight of the fish to the nearest 0.1gm

- The egg diameter of 10 randomly selected eggs per female was measured along two axes using a calibrated eyepiece micrometer on the binuclear microscope at a power magnification of 4X.
- Absolute fecundity was estimated by taking sub-samples from anterior, posterior and middle regions of ovary representing certain weight. All ripening eggs were counted

in sub-samples, then reported to the total weight of the ovary.

### Blood sexual hormones analysis

Blood samples were withdrawn directly from the heart of live *O. niloticus* by a syringe containing sodium citrate as anticoagulant. Blood samples were centrifuged at 5000 rpm for 10 minutes to get plasma for analysis of testosterone (T) and estradiol (E2) which measured by radioimmunoassay (RIA) using methods described by Rajkowski *et al.* (1977) and Wisdom (1976), respectively.

### Histological examinations

The removed gonads (60 subsamples of each testis and ovaries) were quickly fixed in formalin-saline solution for 24h. The specimens dehydrated in ascending series of ethyl alcohol, cleared with xylene, infiltrated, and embedded in wax then sectioned at 5µm thickness using rotary microtome and stained with Haematoxyline and Eosin (Humason, 1972). Sections were photographed with a Leitz Diaplan microscope (5X-10X-20X-40X).

### Statistical analysis:

Results were statistically analyzed using the one way Analysis of Variance test (ANOVA). The statistical analysis was performed using the computer software SPSS (Version 18). Duncan's New Multiple Range Test was used to evaluate the differences between means for treatments at the 0.05 significance level.

### Results

As shown in Table 2, all groups of the experiment had very close values for all measured water parameters, except alkalinity and hardness. Averages of pH, dissolved oxygen and unionized ammonia (UIA) were 7.55±0.13, 7.41±0.21 mg/L and 0.0063±0.0026 mg/L, respectively. The same concentration of nitrate (5 mg/L) was estimated for all tanks during the whole period of this study while concentration of nitrite was under detection of the used kits.

However, alkalinity and hardness displayed more variable values among experimental groups. Values of alkalinity ranged between 91.54±12.13 and 106.13±23.91 mg CaCO<sub>3</sub>/L in G3 (low dose) and G5 (high dose), respectively with the average of 100.42±18.03 mg CaCO<sub>3</sub>/L. Hardness had the average of 116.06±10.82 mg CaCO<sub>3</sub>/L, with the lowest value (111.13±10.79 mg CaCO<sub>3</sub>/L) in G3 and the highest value (121.04±6.3 mg CaCO<sub>3</sub>/L) in G5.

**Table (2): Physicochemical characteristics of water used in the experiment**

Parameter	mean ± SD
pH	7.55±0.13
Dissolve oxygen (mg/L)	7.41±0.21
Alkalinity mg(CaCO <sub>3</sub> )/L	100.42±18.03
Hardness mg(CaCO <sub>3</sub> )/L	116.06±10.82
Unionized ammonia (NH <sub>3</sub> )(mg/L)	0.006±0.003
Nitrite (NO <sub>2</sub> <sup>o</sup> ) (mg/L)	0
Nitrate (NO <sub>3</sub> <sup>o</sup> ) (mg/L)	5

### Reproductive biology

Results of GSI, egg diameter and absolute fecundity were summarized in Table 4. There were significant differences ( $P \leq 0.05$ ) among experimental groups after both treated and recovery months for all parameters except that of GSI in males. The treated groups showed lower values of GSI, egg diameter and absolute fecundity than those recorded in G1 after both treated and recovery months. However, values of all parameters in treated groups decreased gradually with increasing the dose of *C. papaya* seeds, reaching the lowest value in G5 (high dose). This was more obvious in fecundity when values in treated females were less than half that recorded in G1.

**Table (4): GSI (%), egg diameter and absolute fecundity of *O. niloticus* after treated and recovery months**

Groups	GSI (%)				Egg diameter (mm)		Absolute Fecundity (egg)	
	GSI male		GSI female		Treated month	Recovery month	Treated month	Recovery month
	Treated month	Recovery month	Treated month	Recovery month				
G1	1.45±0.23	1.38±0.31	6.75 <sup>a</sup> ±0.89	6.54 <sup>a</sup> ±0.24	1.9 <sup>a</sup> ±0.04	2.12 <sup>a</sup> ±0.26	861 <sup>a</sup> ±223.60	858 <sup>a</sup> ±63.24
G2	0.99±0.34	1.22±0.27	---	---	---	---	---	---
G3	1.13±0.10	1.36±0.04	5.66 <sup>b</sup> ±0.14	5.75 <sup>b</sup> ±0.09	1.68 <sup>b</sup> ±0.09	1.72 <sup>b</sup> ±0.06	399 <sup>b</sup> ±22.70	420 <sup>b</sup> ±9.90
G4	1.16±0.15	1.27±0.04	5.45 <sup>b</sup> ±0.44	5.68 <sup>bc</sup> ±0.19	1.64 <sup>bc</sup> ±0.1	1.69 <sup>b</sup> ±0.06	363 <sup>b</sup> ±7.60	387 <sup>b</sup> ±11.00
G5	1.02±0.12	1.08±0.12	5.39 <sup>b</sup> ±0.17	5.41 <sup>c</sup> ±0.12	1.51 <sup>c</sup> ±0.08	1.5 <sup>b</sup> ±0.09	322 <sup>b</sup> ±11.10	328 <sup>c</sup> ±5.90
P-value	0.131	0.182	0.035	0.000	0.003	0.005	0.001	0.000

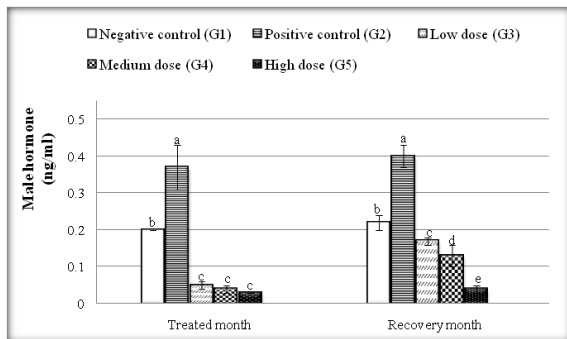
All results are mean±SD of three tanks. Values in the same column having a different superscript letters (a, b and c) differ significantly with each other ( $P \leq 0.05$ ; Duncan's Multiple Range Test).

G1 (Negative control group), G2 (Positive control group), G3 (Low dose treatment), G4 (Medium dose treatment) and G5 (High dose treatment).

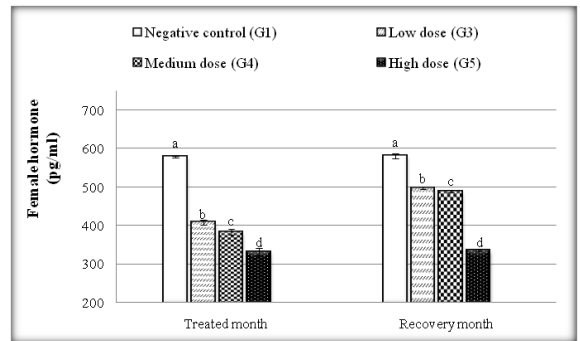
**Analysis of blood sexual hormones**

Blood sexual hormones for male (testosterone, T) and female (estradiol, E2) of *O. niloticus* showed high significant differences ( $P \leq 0.05$ ) after treated month (Fig. 1a and b). The treated groups displayed close values ranged between 0.03±0.00 and 0.05 ±0.01 ng/ml in G5 (high dose) and G3 (low dose), respectively, which were significantly much lower than that recorded (0.37±0.06 ng/ml) in G2 (positive control) or even G1 (0.2±0 ng/ml). On the other hand, the level of E2 decreased gradually with increasing the dose of *C. papaya* seeds, ranging between 331.33±9.87 and 408.67±5.86 pg/ ml in G5 and G3, respectively while the highest value of E2 (579.67±2.08pg/ml) was observed in G1.

After recovery period, little ineffective increase was observed in G1, G2 and G5 while an effective increase occurred in G3 and G4. In spite of that, the results still have high significant difference ( $P \leq 0.05$ ) in sex hormones (T and E2) following the same trends of difference as those obtained after treated pe-riod.



(a)



(b)

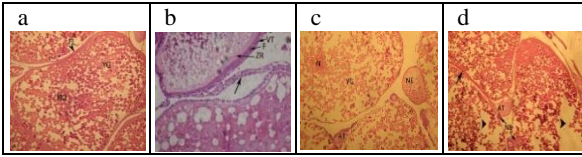
**Figure 1:** Effect of *C. papaya* on blood sexual hormones in male (a) and female (b) of *O. niloticus* after treated and recovery months

**Histological Examination**

Normal structure was noticed after treated month in the ovary of the negative control fish (G1) (Fig. 2a). While separation of follicular layer was noticed in the ovary of fish fed on low dose (G3) (Fig. 2b). Necrosis, atresia (Fig. 2c) and ova fusion (fig. 2d) were observed in the ovaries of medium and high dose treated fish (G4 and G5), respectively. After recovery month, atresia was visible in G1 (Fig. 3a), in addition to vacuolated areas in the ova of G3 (Fig. 3b). Degeneration stroma was seen in the ovary of both G4 and G5 (Figs. 3c and d, respectively).

Examination of testis after treated month revealed a normal structure in G1 (Fig. 4a). However, testis in G2 showed deformed interlobular (Fig. 4b). Deformation in seminiferous lobule (Fig. 4c) and in the interlobular tissue (Fig. 4d) were observed in G3 and G4, respectively. Figure 4e showed deformation of both seminiferous lobule and interlobular tissue in G5. No recognizable changes were observed in the testis of G1 (Fig. 5a). Deformed interlobular tissue was observed in G2 (Fig. 5b). G3 and G4 showed deformation in seminiferous lobule (Figs. 5c and d, respectively). The same as shown after treated month, deformed seminiferous lobule and interlobular tissue was observed in G5 (Fig. 5e).





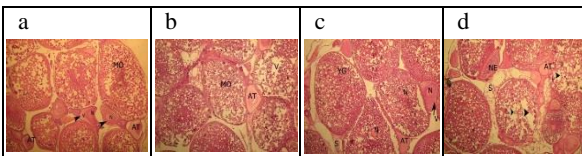
**Figure 2-(a-d):** Cross-sections of ovary tissue of *O. niloticus* showing changes caused by feeding different doses of *C. papaya* seeds.

**Figure 2- (a)** Normal ovary of negative control, showing the follicular layer (▶) of a mature oocyte (MO) which contains yolk granules (YG). H&E, (400X)

**(b)** Ovary of low dose treated fish showing, oocyte surrounded by a layer of vascular connective tissue (VT) - the follicle covered by outer follicular layer (F) and middle zone radiate (ZR). Notice the separation of the follicular layer (→). H&E, (400X)

**(c)** Ovary of medium dose treated fish showing oocyte containing nucleus (N) and yolk granules (YG), atresia (AT) and necrosis (NE). H&E, (50X)

**(d)** Ovary of high dose treated fish showing necrosis (NE), atresia (AT), many vacuolated areas (→) and fusion of ova (▶). H&E, (50X)



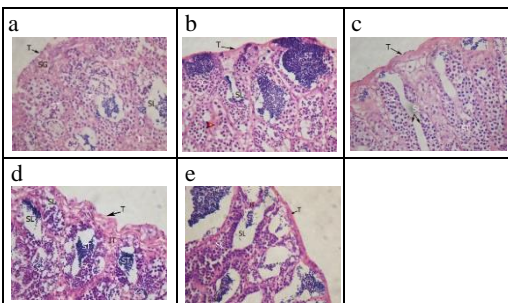
**Figure 3-(a-d):** Cross-sections of ovary tissue of *O. niloticus* showing changes after recovery period

**Figure 3- (a)** Ovary of negative control fish after recovery period showing mature oocyte (MO), vacuolated oocyte stage (V), nucleus (N) and atresia (AT). H&E, (50X)

**(b)** Ovary of low dose treated fish after recovery period showing mature oocyte (MO), vacuolated area (V) and atresia (AT). H&E, (50X)

**(c)** Ovary of medium dose treated fish after recovery period showing nucleus (N) migration to the periphery of the oocyte (vitellogenic stage), atresia (AT) and degenerated stroma (S). H&E, (50X)

**(d)** Ovary of high dose treated fish after recovery period showing necrosis (NE), atresia (AT), degenerated stroma (S) and vacuolated areas (▶). H&E, (50X)



**Figure 4-(a-e):** Cross-sections of testis tissue of *O. niloticus* showing changes caused by feeding different doses of *C. papaya* seeds.

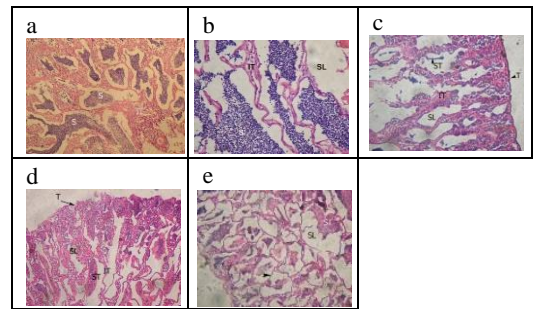
**Figure 4- (a)** Testis of negative control fish showing normal structure of cortex region of testis; spermatozoa (S), spermatocytes (SC), spermatid (ST), spermatogonia (SG), seminiferous lobule (SL), tunica albuginea (T) and interlobular tissue (IT). H&E, (400X)

**(b)** Testis of positive control fish showing structure of cortex region of testis; spermatoocytes (SC), spermatid (ST), seminiferous lobule (SL), tunica albuginea (T) and deformed interlobular tissue (▶). H&E (400X)

**(c)** Testis of low dose treated fish showing spermatoocytes (SC), spermatid (ST), tunica albuginea (T) and deformation in seminiferous lobule (SL). H&E, (200X)

**(d)** Testis of medium dose treated fish showing spermatoocytes (SC), spermatid (ST), seminiferous lobule (SL), tunica albuginea (T) and degeneration in the interlobular tissue (IT). H&E, (400X)

**(e)** Testis of high dose treated fish showing spermatoocytes (SC), spermatid (ST), tunica albuginea (T) and deformation in both seminiferous lobule (SL) and the interlobular tissue (IT). H&E, (400X)



**Figure 5-(a-e):** Cross-sections of testis tissue of *O. niloticus* showing changes after recovery period

**Figure 5- (a)** Testis of negative control fish after recovery period showing ripe testis; ripe testis; spermatozoa (S) and interlobular tissue (IT). H&E, (50X)

**(b)** Testis of positive control fish after recovery period showing a ripe testis; seminiferous lobule (SL) spermatozoa (S) and deformed interlobular tissue (IT). H&E (400X)

**(c)** Testis of low dose treated fish after recovery period showing tunica albuginea (T), residual spermatid (ST), interlobular tissue (IT) and deformation in seminiferous lobule (SL). H&E, (200X)

**(d)** Testis of medium dose treated fish after recovery period showing spend stage residual spermatid (ST), tunica albuginea (T), interlobular tissue (IT) and deformed seminiferous lobule (SL). H&E, (200X)

**(e)** Testis of high dose treated fish after recovery period showing spermatid (ST), deformation in both the seminiferous lobule (SL) and interlobular tissue (IT). H&E, (400X)

**Discussion**

In the present study, values of all recorded parameters of water were within the acceptable limits for growth and reproduction of *O. niloticus* as stated by El-Sayed *et al.* (1996) and Milstein and Svirsky (1996), which mean that, the experimental treatments have no influence on water quality.

Insignificant differences obtained in males GSI was in agreement with previous studies conducted on using *C. papaya* seeds as reproductive inhibitor for experimental animals such as albino rats (Maniyannan *et al.*, 2009) and rabbits (Lohiya *et al.*, 1999). They recorded insignificant differences in the testis weight after administration of *C. papaya* seeds with that of negative control. On the other hand, significant decrease occurred in GSI of female agreed with the finding of Jegede and Fagbenro (2008) and Temitope (2010) who reported

significant decrease in GSI of *O. niloticus* females treated with other medicinal plants such as neem (*Azadirachta indica*) and *Hibiscus rosa sinensis* leaf.

The recorded egg diameter (1.9 mm) in negative control group was within the range of egg diameter recorded in previous studies for untreated *O. niloticus* (2-2.25 mm) (Coward and Bromage, 2004 and Temitope, 2011). Significant decrease detected in the egg diameter of all treated fish was in consistency with that recorded for *O. niloticus* treated with *Aloe vera* latex (Temitope, 2011), but contrary with that recorded for *O. niloticus* treated with *Hibiscus rosa sinensis* leaf (Temitope, 2010). This effect persisted even after elimination of *C. papaya* seeds. The results of fecundity was supported by the finding of Temitope (2011) who recorded decreasing in fecundity of *O. niloticus* treated with different doses of *Aloe vera* latex. Also, Udoh et al. (2005b) observed no pregnancy in female rats that administered *C. papaya* for 3 days.

Total plasma testosterone (T) is the most important indicator on the fertility status in males. Costa and Paula (2006) observed positive and significant correlation between (T) and the volume of Leydig cells which found adjacent to the seminiferous tubules in the testis. O'donnell et al. (2001) stated that testosterone produced by Leydig cells is responsible for spermatogenesis as well as for the development and maintenance of male secondary sex characteristics. This means that the amount of plasma testosterone is related to the capacity of the Leydig cells to secrete testosterone in the animal testis (Ewing et al., 1979). On the other hand, estradiol (E2) is the main sex hormone in female which play an important role in sexual development and responsible for estrogenic activity. It is produced mainly by the ovaries and in smaller amounts by the adrenal glands. Estradiol levels are usually used in evaluating ovarian function.

In the present experiment, increasing of testosterone level in monosex fish was expected as a result of using male hormone. In groups treated with *C. papaya*, the effect of all doses on testosterone and estradiol was markedly obvious when they fall to very low levels after the month of the treatment. The influence of *C. papaya* seeds on testosterone and estradiol levels in low and medium doses groups repaired within a few weeks as shown when their levels increased after stopping treatment with *C. papaya*, but they still significantly lower than negative control. While high dose did not affect, keeping low levels of testosterone and estradiol. This indicates that *C. papaya* can be used as demasculinization agent in fish males and as infertility agent in fish females. Decreasing levels of T and E2 may attribute to the negative impact of *C. papaya* on Leydig cells and ovaries functions respectively. This was in line with results of Maniyannan et al. (2009) and Goyal et al., (2009) who observed similar results on male albino rats administered with methanol subfraction of *C. papaya* seeds. Maniyannan et al., (2009) and Ekanem and Okoronkwo (2003) also were in agreement with the result of recovery month when they recorded reversible effect of *C. papaya* seeds on the male fertility of albino rats and Nile tilapia, respectively.

Sections examination of ovaries and testis tissues in negative control group showed normal histological structure, as expected. Otherwise, the structural alternations such as; separation of the follicular layer that noticed in the ovary of females treated with low dose of *C. papaya* seeds, necrosis

in females treated with medium dose and atresia, necrosis, many vacuolated and fusion of ova in females of high dose treatment, may be attributed to the effect of oleanolic glycoside, the active ingredient in *C. papaya* seeds responsible for sterility process (Das, 1980). This is in agreement with the finding of Temitope and Oyedapo (2008a) who administered *O. niloticus* with different doses of *C. papaya* seeds meal for 60 days. They observed severe atretic follicle in ovaries of fish fed high dose (2.0g *C. papaya* /kg). Similar histological effect was reported on ovaries of *O. niloticus* by using other medicinal plants; *Hibiscus rosa sinensis* leaf meal (Temitope, 2010) and *Aloe Vera* Latex (Temitope, 2011). Temitope and Oyedapo (2008b) and Jegede et al. (2008) obtained the same histological effects on the ovary of *Tilapia zillii* fed on neem (*Azadirachta indica*) leaf meal.

The effect of *C. papaya* seeds on the testis tissues was noticed as deformation in seminiferous lobules of males treated with low dose of *C. papaya* seeds, as degeneration in the interlobular tissue in males treated with medium dose and as deformation in both seminiferous lobule and the interlobular tissue in high dose treated males. Deformed interlobular tissues were also noticed in monosex fish which may be due to 17<sup>0</sup>-methyl testosterone treatment. This results was confirmed by the work of Ekanem and Okoronkwo, (2003) who examined the use of *C. papaya* seeds as a fertility control agent for the male of *O. niloticus* and follow similar concentrations (60g, 120g/kg). They observed swollen nuclei on the sperm cells of fish fed low dose while the high dose of *C. papaya* seed caused disintegration of the sperm cells and the formation of more swollen nuclei in the sperm cells. Temitope and Oyedapo (2008b) recorded the presence of seminiferous tubules in testis *O. niloticus* fed *C. papaya*. Temitope (2010) and Temitope (2011) observed similar histological effect in testis of *O. niloticus* treated with *Hibiscus rosa sinensis* leaf meal and *Aloe Vera* Latex respectively.

After recovery month, sections of both ovaries and testis showed possibility of reversible effects in low and medium doses treatments, while permanent sterility occurred in high dose treatment. Ekanem and Okoronkwo (2003) reported the absence of spawning in aquaria received the high dose treatment of *C. papaya* seed after stopping the treatment for 30 days. Maniyannan et al. (2009) recorded restoration of proper spermatogenesis in male albino rat after 120 days of recovery of *C. papaya* seeds treatment. Lohiya et al. (1999) recorded complete reverse in male rabbits administered *C. papaya* seeds after withdrawal of the treatment.

## Conclusion

The present study comes to introduce *C. papaya* seeds, which are cheap, easy to obtain and non-commercial, as a natural agent to control the fertility of the farmed tilapia and overcome the problem of early maturation, instead of unfavorable and expensive hormone. Reproductive parameters such as gonado-somatic index, egg diameter and fecundity as well as values of plasma estradiol and testosterone and histological observations of testes and ovaries in *O. niloticus* treated with *C. Papaya* seeds at doses of 30 and 60 g/kg revealed temporary sterility while that treated with high dose (120 g/kg) had permanent sterility. This makes *C. papaya* seeds at the dose of 120 g/kg diet are recommendable for use as sterility-inducing agents in males or females of *O. niloticus*.

## REFERENCE

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