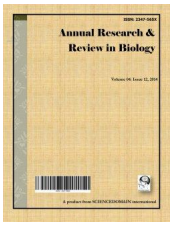

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# Hormonal Assay and Reproductive Performance of Rabbits Fed Pawpaw (*Carica papaya*) Leaves as Feed Supplement

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***Authors' contributions***

*This work was carried out in collaboration between all authors. Author AJH designed the study, wrote the protocol and wrote the first draft of the manuscript. Authors GUU and POO performed the statistical analysis and managed the analyses of the study. Author MIA managed the literature searches. All authors read and approved the final manuscript.*

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## ABSTRACT

**Aims:** To evaluate the effect of *Carica papaya* leaves on some reproductive hormones of male and female rabbits, and monitor their reproductive performance.
**Study Design:** The experimental design used for the study was Completely Randomized Design.
**Place and Duration of Study:** Department of Animal Science, Faculty of Agriculture, University of Calabar and Haematology/Pathology and Microbiology Laboratory, School of Health Technology, Calabar, Cross River State. Feeding trial lasted for ten weeks.
**Methodology:** Twenty-seven sexually matured crossbred (New Zealand White x Chinchilla) rabbits (eighteen does and nine bucks). Rabbits were randomly assigned to three dietary treatment groups made up of three replicates comprising of 1 buck: 2 does (mating ratio/replicate). Dietary treatments: control ($T_1$) - fed concentrate and potato (*Ipomea batatas*) leaves; ($T_2$) - fed concentrate and fresh pawpaw leaves; ($T_3$) - fed concentrate and wilted pawpaw leaves. Blood samples were

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collected from six animals (3 does and 3 bucks) per treatment group, from the marginal ear vein. This was done a day before mating and two weeks post-mating (before feeding in the morning). Test for hormonal parameters in blood serum (testosterone, luteinizing hormone (LH), follicle stimulating hormone (FSH), Progesterone and Estradiol) were carried out with the aid of tube-based enzyme immunoassay (EIA) method.

**Results:** Reproductive parameters evaluated were observed not to be affected by dietary treatments. Follicle stimulating hormone levels increased between pre-mating and post-mating as a result of ovarian factors. Luteinizing hormone was relatively lower during pregnancy. Progesterone levels also increased within the period before mating to post mating for rabbits fed fresh pawpaw leaves (8.25 to 9.14 $ngmL^{-1}$) and wilted pawpaw leaves (7.93 to 8.80 $ngmL^{-1}$).

**Conclusion:** Pawpaw leaves can be served to rabbits either in its fresh or wilted state without affecting reproductive parameters assessed in this study. Results of hormonal assay did not reveal any adverse effects due to dietary treatment on the rabbits.

*Keywords: Reproduction; hormones; rabbit; Carica papaya.*

## 1. INTRODUCTION

Rabbit (*Oryctolagus cuniculus*) is described as a mini livestock specie that can be a major contributor to the diet of both rural and urban settlements. The rabbit's small and flexible nature makes it a suitable livestock that can be handled by women and children [1]. Rabbit has since been identified as an economy livestock that could bridge the wide gap for dietary protein intake in Nigeria. Lyon [2] observed a gradual shift from the production, consumption of conventional animal protein sources (cattle, sheep, swine, goat and poultry) to a class of livestock referred to as non-conventional livestock. The economic potentials of the rabbit which include short generation interval, rapid growth rate, genetic diversity, large litter size, ability to utilize forage and agricultural by-products and adaptation over wide range of ecological environment [3]; qualifies this animal type as being a readily available source of animal protein when properly managed. As an essential, it becomes very important to evaluate other sources of fodder that could be included as feed stuff for rabbits owing to differences in environments where this animal could be raised.

Pawpaw (*Carica papaya*) is a plant which produces edible fruits and it is one of the twenty-two (22) accepted species in the genus *Carica* of the plant family *Caricaceae* [4]. Pawpaw (*Carica papaya*) plant comes in three sexes: "male", "female" and "hermaphrodite". The male produces only pollen, never fruit. The female will produce small, inedible fruits unless pollinated. The hermaphrodite can self- pollinate since its flowers contain both male stamens and female ovaries [4].

Fresh green pawpaw leaves have antiseptic properties, while the brown, dried pawpaw leaves are best as tonic and blood purifier [5]. Fresh green pawpaw leaves are rich in vitamins and minerals. The vitamins include thiamine ($B_1$) (0.94 mg/100 g); riboflavin ($B_2$) (0.13 mg/100 g) and ascorbic acid (16.29 mg/100 g). While minerals include Calcium (Ca) (8612.50 mg/kg); Magnesium (Mg) (67.75 mg/kg); Sodium (Na) (1782.00 mg/kg); Potassium (K) (2889.00 mg/kg) and Manganese (Mn) (9.50 mg/kg) [6]. Therefore, the aim of this study was to assess the hormonal assay and reproductive performance of rabbits fed pawpaw leaves as feed supplement.

## 2. MATERIALS AND METHODS

### 2.1 Location of the Study

The study was carried out at the rabbitry unit of the Teaching and Research Farm of the Department of Animal Science, University of Calabar.

### 2.2 Experimental Animals and Management

Twenty seven (27) sexually matured cross bred rabbits were used for this study. This number comprised of eighteen (18) does and nine (9) bucks. Mating was 1 buck: 2 does. Rabbits were managed under a mixed feeding regime and housed individually within their cells. Cells were properly labeled for identification. Animals were fed twice daily (morning and evening) by providing feed (weighed quantity) and drinking water *ad libitum* throughout the experimental period of 60 days. On arrival, anti – stress with

composition (per 1000 g) of 15,000,000 iu (vitamin A), 87,000 mg (potassium), 4,400,000 iu (vitamin D3), 212,000 mg (sodium sulphate), 1,350 iu (vitamin E), 50,000 mg (sodium chloride), 4,350 mg (vitamin K), 12,000 mg (magnesium sulphate), 4,350 mg (vitamin B2), 12,000 mg (copper sulphate), 2,350 mg (vitamin B6), 12,000 mg (zinc), 11,350 mg (vitamin B12), 12,000 mg (manganese sulphate), 1,000 mg (vitamin C, 15,000 mg (lysine hydrochloride), 16,700 mg (nicotinamide), 10,000 mg (methionine), 5,350 mg (calcium pantothenate) and 1,000 mg (excipients q.s) was offered in drinking water to the rabbits to cushion transportation stress. The rabbits were dewormed using piperazine in clean drinking water. The rabbits were further protected against microbial infection using an antibacterial and anti-protozoan preparation (amoxicillin and clavulanic acid) a broad-spectrum antibiotic to curb diarrhea. Ivomectin was also administered at the rate of 0.15 ml per rabbit to control both endo - and ecto - parasites.

## 2.3 Housing and Equipment

The animals were housed in wooden hutches measuring 65 X 65 X 65 cm and raised 75 cm from the floor level. The hutches were thoroughly washed and disinfected with saponated cresol (Izal) and allowed to dry for seven (7) days before the animals were brought. The floor of the hutches was made of wire mesh. Feed and watering troughs were provided in each hutch. The rabbits were placed individually in clearly marked hutches. Prior to the commencement of the study, rabbits were allowed two (2) weeks of acclimatization.

## 2.4 Collection of Fresh Pawpaw (*Carica papaya*) Leaves

A formulated ration was used as basal diet in this study, which was supplemented with pawpaw leaves. Composition of diet is as presented in Table 1. Pawpaw leaves served to the experimental animals were harvested within and outside the University environment. The leaves were served to the animals in two forms – fresh and wilted. The fresh pawpaw leaves were harvested in the mornings and served to the rabbits one hour after to allow for the morning dew to dry off the leaves whereas the wilted pawpaw leaves were cut the day before and served to the rabbits the following morning.

**Table 1. Gross composition of experimental diet (Basal diet)**

| Ingredient | % Composition |
|---|---|
| Maize | 36.00 |
| SBM | 15.70 |
| Fish Meal | 2.00 |
| PKC | 4.00 |
| Wheat Offal | 10.00 |
| Rice husk | 27.00 |
| Palm Oil | 2.50 |
| Bone meal | 2.00 |
| Methionine | 0.25 |
| Lysine | 0.25 |
| Salt | 0.10 |
| Premix | 0.20 |
| Total | 100.00 |

*Calculated analysis; % CP = 15.83; % CF =10.00: ME (Kcal/Kg) = 2,488.68*

## 2.5 Feeding

The animals were randomly assigned to three treatments groups comprising of nine (9) rabbits each (6 does and 3 bucks). Each treatment had 3 replicates (1 buck: 2 does). Treatment groups were:

Control ($T_1$) = Concentrate + *Ipomea batatas* (Potato) leaves.
Treatment 2 ($T_2$) = Concentrate + Fresh pawpaw leaves.
Treatment 3 ($T_3$) = Concentrate + wilted Pawpaw leaves.

Potato leaves were served to rabbits in the control treatment ($T_1$) to stabilize for forage. Pawpaw leaves served in treatments 2 and 3. Each rabbit was offered a weighed amount of feed at 5 percent body weight daily in the morning hours (07:00 - 08:00am) and water served *ad libitum*. Left over feed was collected into clearly labeled disposable plastic plates with a lid and weighed daily with precision electronic balance; model HK – DC-320AS with minimum and maximum sensitivities of 0.01 and 320 g, respectively. The feed intake was computed by difference.

## 2.6 Determination of Reproductive Performance of Rabbits

### 2.6.1 Breeding and reproduction

Mating across the treatment groups was done at the ratio of 1 buck: 2 does. Post – service pregnancy was monitored by observing body

weight changes in does, in addition to abdominal palpation two (2) weeks post – mating. At the end of the third week kindling boxes were placed in the cells of pregnant does. Non – pregnant does were re-mated.

The following reproductive parameters were determined:

i. Litter size at birth
ii. Litter size at weaning
iii. Litter weight at birth
iv. Litter weight at weaning

## 2.7 Blood Collection and Hormonal Assay

The blood (six per treatment from both sexes) were collected from the marginal ear vein of the experimental rabbits a day before mating and two weeks post-mating between the hours of 7:00 and 8:00am before feeding. The blood samples were collected into a set of sterile plastic bottles and allowed to coagulate to produce sera for hormonal analyses. The test for hormonal parameters in the blood serum (testosterone, luteinizing hormone (LH), follicle stimulating hormone (FSH), progesterone and estradiol were carried out with the aid of the tube-based enzyme immunoassay (EIA) method. The protocol used for the hormonal assay was according the method of [7] as described for the Kit [BioCheck ELISA Assay, USA]. The hormonal assay was carried out before and after mating, respectively.

## 2.8 Experimental Design

The animals were assigned to dietary treatments using the Completely Randomized Design (CRD).

### 2.8.1 Statistical analysis

All data collected were subjected to one - way analysis of variance for Completely Randomized Design (CRD) using Genstat Release 10.3 statistical package. Significance means were separated using the least significance difference (LSD) option of the same statistical software as described by [8].

# 3. RESULTS

## 3.1 Proximate Composition of the Fresh and Wilted Pawpaw Leaves as Well as the Basal Diet

Results of proximate composition of fresh and wilted pawpaw leaves as well basal diets are presented in Table 2. The crude protein content of the fresh pawpaw leaves (FPPL) was 17.50% while that of the wilted pawpaw leaves (WPPL) was 13.12% and the basal diet recorded 14.75%. The ether extract (EE), crude fibre (CF), Ash, nitrogen free extract (NFE) and dry matter (DM) contents of the fresh pawpaw leaves were 7.00, 15.00, 45.50 and 19.61%, respectively. The wilted pawpaw leaves had 5.50, 17.50, 10.00, 53.88 and 21.29 %, respectively. While the basal diet (Concentrate) recorded 2.50% ether extract, 10.00% crude fibre, 10.00% Ash, 62.75% nitrogen free extract and 91.84% dry matter, respectively.

## 3.2 Hormonal Characteristics of Rabbits Fed Fresh and Wilted Pawpaw Leaves

Results of the hormonal characteristics of rabbits fed fresh and wilted pawpaw leaves as feed supplements are presented in Table 3.

### 3.2.1 Follicle stimulating hormone (FSH)

Results for FSH before mating were 4.05, 6.00 and 6.96 $mIUmL^{-l}$, respectively for the control, fresh and wilted pawpaw leave dietary treatments; while results for FSH post-mating in the female rabbits recorded no significant difference ($P > 0.05$) between dietary treatments. The values were 4.13, 6.09 and 6.29 $mIUmL^{-l}$, respectively for the control, fresh and wilted pawpaw leave dietary treatments.

**Table 2. Proximate composition of fresh, wilted pawpaw leaves and basal diet**

| Parameters (%) | Fresh pawpaw leaves | Wilted pawpaw leaves | Concentrate |
|---|---|---|---|
| Dry matter | 19.61 | 21.29 | 91.84 |
| Crude protein | 17.5 | 13.12 | 14.75 |
| Crude fibre | 15.0 | 17.5 | 10.0 |
| Ether extract | 7.0 | 5.5 | 2.5 |
| Ash | 15.0 | 10.0 | 10.0 |
| Nitrogen free extract | 45.5 | 53.88 | 62.75 |

#### 3.2.2 Luteinizing hormone (LH)

Results for LH before mating were 12.29, 11.88 and 12.37 mIUmL$^{-l}$, respectively, for the control, fresh and wilted pawpaw leave dietary treatment. While results for LH post-mating in the female rabbits recorded no significant difference (P > 0.05) between dietary treatments, the values were 8.50, 15.10 and 10.30 mIUmL$^{-l}$ respectively, for the control fresh and wilted pawpaw leave diets.

#### 3.2.3 Estradiol ($E_2$)

Result for $E_2$ in the female rabbits before and after mating recorded no significant difference (P >0.05) between dietary treatments. The results before mating were 24.36, 22.50 and 20.14 pg mL$^{-1}$ and post-mating results were 25.00, 25.52 and 22.01 pg mL$^{-1}$ respectively for the control, fresh and wilted pawpaw leave dietary treatments.

#### 3.2.4 Progesterone

Results for progesterone before and after mating in the female rabbits were 7.98, 8.25 and 7.93 pg mL$^{-1}$ and 7.88, 9.14 and 8.80 pg mL$^{-1}$ respectively in the control, fresh and wilted pawpaw leave dietary treatment. The results at both phases showed no significant difference (P > 0.05) between dietary treatments.

#### 3.2.5 Testosterone

Results for testosterone in the male rabbits before and after mating recorded no significant difference (P > 0.05) between dietary treatments. The values before mating were 7.98, 8.25 and 7.93 pg mL$^{-1}$. While result after mating were 7.88, 9.14 and 8.80 pg mL$^{-1}$, respectively for the control, fresh and wilted pawpaw leave dietary treatment.

### 3.3 Reproductive Performance of Rabbits Fed Pawpaw Leaves as Feed Supplements

Results for reproductive performance of rabbits fed pawpaw leaves as feed supplement are presented in Table 4. From analyses of data obtained in this study, a non-significant (P > 0.05) treatment effect was observed for all parameters monitored. Average litter size at birth obtained in were 5.25($T_1$), 4.75($T_2$) and 6.80($T_3$) while average litter weights obtained in the control, fresh and wilted pawpaw leaves dietary treatment groups were 120.00, 164.00 and 192.00 g, respectively.

Average birth weight of kit within the experimental groups were 22.86 g (control), 34.53 g (fresh pawpaw leaves) and 28.24 g (wilted pawpaw leaves).

### 3.4 Average Litter Size at Weaning

The results obtained were 1.00, 4.25 and 5.00 for average litter size at weaning for the control, fresh and wilted pawpaw leaves' groups, respectively. Average litter weight at weaning for the control, fresh and wilted pawpaw leaves dietary treatments were 450.00, 825.00 and 826.50 g, respectively.

## 4. DISCUSSION

### 4.1 Proximate Composition of Test Materials

The proximate composition of the test ingredients (fresh and wilted pawpaw leaves) as well

**Table 3. Hormonal characteristics of rabbits fed pawpaw leaves as feed supplement**

| Hormones | $T_1$ Control | $T_2$ Fresh pawpaw leaves | $T_3$ Wilted pawpaw leaves | ±S.E.M |
|---|---|---|---|---|
| **Before mating** | | | | |
| FSH (mIUmL$^{-1}$) | 4.05$^b$ | 6.00$^a$ | 6.96$^a$ | 0.67 |
| LH(mIUmL$^{-1}$) | 12.29 | 11.88 | 12.37 | 0.26 |
| Estradiol($E_2$) (pgmL$^{-l}$) | 24.36 | 22.50 | 20.14 | 1.02 |
| Progesterone (ngmL$^{-l}$) | 7.98 | 8.25 | 7.93 | 0.67 |
| Testosterone (ngmL$^{-l}$) | 4.58 | 4.58 | 4.54 | 0.23 |
| **Post-mating** | | | | |
| FSH (mIUmL$^{-1}$) | 4.13 | 6.09 | 6.29 | 0.90 |
| LH(mIUmL$^{-1}$) | 8.50 | 15.10 | 10.30 | 2.81 |
| Estradiol($E_2$) (pgmL$^{-l}$) | 25.00 | 25.52 | 22.01 | 1.62 |
| Progesterone (ngmL$^{-l}$) | 7.88 | 9.14 | 8.80 | 0.96 |

*$^{a,b}$: means on the same row with different superscripts are significantly different (P<0.05)*

**Table 4. Reproductive performance of rabbits fed pawpaw leaves as feed supplements**

| Parameters | $T_1$ Control | $T_2$ Fresh pawpaw leaves | $T_3$ wilted pawpaw leaves | S.E.M |
|---|---|---|---|---|
| Av. litter size at birth | 5.25 | 4.75 | 6.80 | 0.62 |
| Av. litter weight at birth (g) | 120.00 | 164.00 | 192.00 | 20.95 |
| Av. birth weight of kitten (g) | 22.86 | 34.53 | 28.24 | 3.37 |
| Av. litter size at weaning | 1.00 | 4.25 | 5.00 | 1.27 |
| Av. litter weight at weaning (g) | 450.00 | 825.00 | 862.50 | 131.7 |

*AV. = Average*

as the basal diet (concentrate) are presented in Table 2. Proximate composition revealed that crude protein content of the basal diet (14.75 percent) was in line with recommended level for growing rabbits [9]. Also, the crude protein content of fresh and wilted pawpaw leaves (17.5 and 13.12 percent) respectively were within the recommended levels for rabbits generally. The crude protein levels of 12, 15, 16 and 17 percent, respectively have been reported as optimum levels for rabbits in terms of maintenance, gestation, growth and lactation [10]. The crude fibre level of the test materials and basal diet were in the range of 10.00 - 17.50 percent and is within the recommended level of crude fibre level of 10 – 14 percent for rabbits [11]. Research findings have reported that crude fibre level in excess of 9 percent is needed for normal growth and effective intestinal motility in rabbits [10]. The ether extract, ash, nitrogen free extract and dry matter content are within the recommended levels required in rabbit ration to support reproductive performance.

## 4.2 Hormonal Characteristics of Rabbits Fed Pawpaw Leaves as Feed Supplement

### 4.2.1 Follicle stimulating hormones (FSH)

FSH before mating recorded significant ($P < 0.05$) difference between dietary treatments (Table 3). The treatment with wilted pawpaw leaves recorded the highest level (6.96 $mIUmL^{-1}$), followed by the fresh pawpaw leaves dietary treatment and least in the control diet. The progressive increase in FSH levels across dietary treatments before and post - mating indicated that feeding pawpaw leaves to the rabbits did not inhibit the functions of ovarian factors, which was evident in the surge. This result however, is contrary to the findings of Ogbuewu et al. [12] who recorded a progressive decline in FSH values due to ginger rhizome powder supplementation. The residual papain content in the fresh and wilted pawpaw leaves obtained in this study affected positively the synthesis of FSH, thereby enhancing the releasing of FSH into the blood, thereby boosting the reproductive potentials of the female rabbits.

### 4.2.2 Luteinizing hormone (LH)

The results of LH before and post - mating recorded no significant ($P < 0.05$) difference between dietary treatments. The LH values (Table 3) before mating were within the range of 11.88 - 12.37 $mIUmL^{-1}$ and far higher in fresh pawpaw leaves than the values obtained in wilted pawpaw leaves after mating (8.50 - 15.10 $mIUmL^{-1}$). The LH values were highest in the wilted pawpaw leaf dietary treatment in the pre-mating phase and least in the fresh pawpaw leaf dietary treatments compared to the control. However, in the post-mating phase, the fresh pawpaw leaves recorded the highest level of LH followed by the wilted pawpaw leaves and least in the control. Results showed that pawpaw leaves and associated papain content had no adverse effect on the level of LH in the circulating blood. The LH values obtained in this study are higher than the results recorded by Ogbuewu et al. [12], who reported the following ranges 0.10 - 0.66 $mIUmL^{-1}$ for male rabbits and 0.90 - 3.15 $mIUmL^{-1}$ in female rabbits, respectively who administered ginger powder to rabbits. The differences in results in the different studies could be attributed to the different test ingredients used in the separate studies. Since LH level in this study was not adversely affected, it showed that the normal plasma estradiol and progesterone concentrations were not adversely affected since, LH in the female rabbits is responsible for maintaining these hormones [13].

### 4.2.3 Estradiol

The estradiol levels in both phases (before and post-mating) did not show any significant ($P > 0.05$) difference between dietary treatments. The levels in both phases were relatively the same with a gradual declining trend across dietary

treatments. The wilted pawpaw leaves had the least estradiol levels in both phases. While fresh pawpaw leaves dietary treatment recorded the highest estradiol level compared to the control group. The values (Table 3) obtained in this study could be as a result of medicinal components of pawpaw leaves, according to earlier research findings by Hsia et al. [14] who stated that the administration of some medicinal plant extracts in rabbits would have an increase effect in circulating estradiol as observed in the fresh leaves dietary treatment. The marginal decrease in estradiol level across dietary treatments contradicted the report of Ogbuewu et al. [12] who recorded increase in the concentration of blood estradiol (166.35 - 175.05 $pgmL^{-1}$) with the supplementary ginger rhizome powder. These authors attributed such increase in estradiol level to increase metabolic activities in the supplemented animals due to increased uptake and assimilation of the ginger powder of these animals [15].

#### 4.2.4 Progesterone

The levels of progesterone across dietary treatments in the two phases (before and post-mating) recorded no significant ($P > 0.05$) difference. The values (Table 3) were relatively the same in both phases in a fluctuating trend. However, the fresh pawpaw leaf dietary treatments in both phases recorded the highest levels of progesterone compared to the other two treatments. The values of progesterone (15.45, 16.80, 16.55, 15.57) obtained in this study are slightly lower than the values earlier reported by Ogbuewu et al. [12]. The difference in results could be attributed to the difference in the test materials used in the separate studies. According to Osinowo [13], progesterone helps to regulate the oestrus cycle, prepare the animals for conception and pregnancy. Research findings in this study showed that pawpaw leaves either fresh or wilted did not have any adverse effect on the circulating progesterone level as reproductive activities (conception to parturition) were observed to be successful for females among the treatment groups.

#### 4.2.5 Testosterone

The serum testosterone level recorded no significant ($P > 0.05$) difference across dietary treatments in both phases. The testosterone values (Table 3) obtained in this study are higher than the values (2.51 – 2.88 ng $mL^{-1}$) reported by Iwu [16] and (3.93 – 6.28ng $mL^{-1}$) Ogbuewu et al. [12] respectively, who fed coconut ash supplement and ginger powder, respectively to rabbits. According to Osinowo [13] and Ogbuewu et al. [12], circulating testosterone in the male is produced primarily in the gonads under the influence of FSH and LH. The observed marginal increase in the testosterone level at post-weaning phase is an indication that pawpaw leaves contain some androgenic analogue which acts on the hypothalamus to stimulate testosterone released from the testis through the action of gonadotropin releasing hormone (GnRH) and Luteinizing hormone. This observed increased in testosterone level is in agreement with the findings of Ogbuewu et al. [12] in which ginger rhizome powder was administered on rabbits.

### 4.3 Reproductive Performance of Rabbits Fed Pawpaw Leaves as Feed Supplements

The reproductive performance of rabbits is an important feature that determines the efficiency of production [17]. The reproductive performance characteristics of rabbits fed fresh and wilted pawpaw leaves are presented in Table 4.

#### 4.3.1 Average litter size at birth

Average litter size at birth did not differ ($P > 0.05$) statistically across treatment groups (Table 4). Recorded values in this study were 5.25 (Control), 4.75 (Fresh pawpaw leaves) and 6.80 (Wilted pawpaw leaves). The lowest average litter size was recorded for rabbits fed fresh pawpaw leaves. These findings confirm the results of EFSA [18] that certain plant materials contain toxic factors that affect fertility in rabbits. The average litter size in this study ranged from 6.80 – 7.45 is slightly higher than the average values of 4.27 - 5.33 as reported for New Zealand and Dutch rabbits [19]; the values however, are comparable with the average litter size of 6.5 - 7.25 in rabbits as reported by Ewuola and Egbunike [20]. Differences in litter size in rabbits could be attributed to breed/strain differences, age at first parity, feed quality and environmental factors [21].

#### 4.3.2 Average litter weight at birth

The average litter weight at birth in this study ranged from 120.00 – 192.00 grams without any significant ($P > 0.05$) difference between dietary treatments. The average litter weight at birth in this study increased across dietary treatments

with the wilted pawpaw leaves dietary treatment having the highest litter weight at birth implying that wilted pawpaw leaves and associated papain has no adverse effect on the litter weight of rabbits at birth. The values however, are within the range of values earlier reported by Omole et al. [22].

#### 4.3.3 Average birth weight per kit

The average birth of kit obtained in this study ranged from 22.86 - 34.53 grams, with a non-significant ($P > 0.05$) difference between dietary treatments. The values are comparable with the range of values (25.48 - 30.00 g/kitten) reported by Omole et al. [22]. The fresh pawpaw leaves dietary treatment recorded the highest average birth weight of kit implying that fresh pawpaw leaves had no adverse effect on the average birth weight of kittens.

#### 4.3.4 Average litter size at weaning

The average litter size at weaning in this study ranged from 1.00 - 5.00 across dietary treatments. The wilted pawpaw leaves dietary treatment recorded the highest average weight at weaning, followed by the fresh pawpaw leave dietary treatment and least in the control diet. The findings of this study therefore shows that the test materials used as feed supplements are capable of sustaining does throughout the period of nursing without adverse effect on its kits at the point of weaning.

#### 4.3.5 Average litter weight at weaning

The average litter weight at weaning (4 weeks) of rabbits obtained in this study ranged from 450.00 - 862.50 g across dietary treatments. The highest average litter weight at weaning was recorded in the wilted pawpaw leaves dietary treatment, followed by fresh pawpaw leaves dietary treatment and least in the control group. This implies that feeding pawpaw leaves either as fresh or wilted will not adversely affect the weaning weight of kits. This agrees with the report of Orunmuyi et al. [23] that litter size at birth does not exert significant effect on weaning weight of an animal.

## 5. CONCLUSION

From the results of this study, it can be concluded that rabbit does can tolerate pawpaw leaves as feed supplement without adverse effects on reproductive performance. In addition, pawpaw leaves fed to rabbits showed no negative implication on the reproductive hormones of rabbits. However, further investigation could be carried out on reproductive performance of rabbits exposed to pawpaw leaves over long periods.

## ETHICAL APPROVAL

As per international standard or university standard written ethical approval has been collected and preserved by the authors.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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