

Seasonal changes in semen characteristics of pure and crossbred rabbit bucks fed graded levels of vitamin C under heat stress conditions

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ABSTRACT: The experiment was carried out to investigate the effect of season and graded vitamin C levels on semen quality of rabbit bucks in the Northern Guinea Savanna of Nigeria. A total of 55 rabbit bucks consisting of 40 pure New Zealand White (NZW) and 15 New Zealand x California (NZW×CAL) crossbreds were used for the study. The duration of the study spanned the hot dry (March-May) and cool wet (June-August) seasons. The traits investigated were semen motility, concentration, volume, pH, colour, live and dead sperm. Results obtained showed that vitamin C had significant ($p<0.01$) effect on semen characteristics. There was a progressive increase in semen motility, concentration and live sperm and decrease in dead sperm as vitamin C level increased. Semen volume deviated from this pattern while colour was consistent all through. The result on monthly variation indicated that volume increased during the months of March to May ranging between 1.32-1.43 ml while during the months of June to August, it oscillated between 0.41 - 0.95 ml. Sperm concentration significantly ($p<0.05$) increased from the hot dry months ($75.72-129.48 \times 10^6$) to the cool wet months ($98.17-102.78 \times 10^6$) while colour remained consistent with an increase in live sperm cells. There was significant ($p<0.001$) interaction between vitamin C level and season on sperm motility, concentration and live sperm being better in the hot dry (70.9%, 114.5×10^6 , 85.31%) and cool wet (76.3%, 121.6×10^6 , 90.14%) seasons on 400 mg/kg feed vitamin C. Sperm volume was higher on 100 mg/kg feed vitamin C in the hot dry (1.60 ml) and cool wet (0.98 ml) seasons and 400 mg/kg feed in the hot dry (1.57 ml) and cool wet (0.77 ml) than other levels. Based on the results, it is concluded that vitamin C improved the semen characteristics of rabbit bucks at 400 mg/kg feed during the hot dry season which translates to improved semen quality.

Keywords: Months, rabbit bucks, season, semen characteristics, vitamin C.

INTRODUCTION

In the advent of the avian influenza virus in many countries including Nigeria, there has been a reduction in the production of white meat from poultry, this has created an opening for the rabbit industry to play a pivotal role in solving this shortage of animal protein partially from poultry by bridging the gap between animal protein demand and supply (El-Kholy *et al.*, 2007). Reproductive inefficiency is one of the most limiting constraints to efficient rabbit production. The introduction of artificial insemination (AI) in rabbit breeding maximizes the financial profit of this

industry by reducing the number of bucks raised and consequently, the number of non-productive cages (Alvariño, 2000, Khalifa *et al.*, 2000; Seleem and Riad, 2005). Proper integration of AI in rabbit production ensures that one single buck may affect the fertility and prolificacy of about one hundred does (Seleem, 2005). The efficiency of spermatozoa production, libido, and semen quality changes throughout the reproductive life of an animal due to nutrition, environment, health status, drugs, and chemicals (Ifeanyi 2009). It is therefore important for the

success of reproduction and the AI technique that evaluation of both semen and the fertilizing ability of bucks are reliable.

In tropical countries, high temperature hinders the success of rabbit farming (Cervera *et al.*, 1997) as it leads to a significant reduction in daily weight gain, feed intake and feed efficiency (Chiericato *et al.*, 1993). A very important limitation to rabbit production in a hot climatic area is heat stress. Susceptibility to heat stress produces a series of changes in biological functions which in turn lead to impairment of production and reproductive function (Marai *et al.*, 1999). Such detrimental effects are evident during the hot season of the year, which are reflected in limiting the breeding season of rabbits, normally from May to September in the northern hemisphere (Marai *et al.*, 1996).

Vitamin C (ascorbic acid) is widely used to mitigate heat stress in rabbits; moreover, this vitamin promotes growth, reproduction, and counteracts infections by pathogenic bacteria and viruses (Wang *et al.*, 2004). It also serves as a nutritional supplement, as well as reduces metal ions that generate free radicals (Valko *et al.*, 2005); increases body resistance to environmental stress by reducing the synthesis and secretion of corticosteroids, thus alleviating the negative effects of stress (Roth and Kaerberle, 1985). Ascorbic acid is a known antioxidant present in the testis with the precise role of protecting the latter from oxidative damage (Nayanatara *et al.*, 2008). Vitamin C has been shown to improve sperm motility and enhanced semen quality and fertility in rats (Rekha *et al.*, 200).

The deficiency of vitamin C leads to a state of oxidative stress in the testes that disrupts both spermatogenesis and the production of testosterone. In recent years, vitamin C supplements have been widely used in rat diets and the levels for enhancing production and reproductive performance have been increased several fold. Supplementation with vitamin C has also been shown to increase total sperm output and sperm concentrations (Nayanatara *et al.*, 2008, Rekha *et al.*, 2009). This present study was therefore designed to evaluate the effects of season, month and vitamin C levels on semen characteristics in pure and crossbred rabbit bucks raised in a tropical environment.

MATERIALS AND METHODS

Experimental site

The study was conducted in the Rabbitry of the National Animal Production Research Institute (NAPRI), Shika, Zaria. Shika is located in the Northern Guinea Savanna, on latitude 11°12'N of the equator and longitude 7°33' with an altitude of about 610 mm, morning temperature ranged between 19°C and 32°C while afternoon temperature ranged between 20°C and 38°C. The cool season or the early dry season falls within the months of November-February while the hot season or late dry season falls within

the months of March to May. A detailed description of the experimental site was given by Olotunmogun *et al.* (2017).

Experimental animals, treatments and management

A total of 55 sexually mature rabbit bucks, made up of 40 pure bred New Zealand White and 15 crossbred (NZW×CAL) rabbit bucks were used for this study. The rabbits were weighed and kept individually in metal cages. This consisted of 10 pure bred and 3 crossbred per treatment in three treatments (100, 200, 300 mg/kg feed) and 10 pure bred and 2 crossbred for the last treatment (400 mg/kg feed). The experiment was conducted in a 2 × 4 factorial arrangement consisting two seasons (hot dry and cool wet) and four vitamin C levels (100, 200, 300 and 400 mg/kg feed) in a completely randomized design. The experimental duration spanned the months of March to August. The dimensions of the metal cages are 60 x 60 x 50 cm, placed in a well-ventilated house; the house is of a dwarf wall of 5ft height, covered with wire netting, and roofed with aluminium sheet. Vitamin C was included in the NAPRI standard concentrate feed (see Table 1) in varying levels of 100, 200, 300 and 400 mg/kg feed. Feed and water were provided *ad libitum* in the morning at 08:00 hours daily in flat bottomed earthen pots.

Data collection

Semen quality parameters

Bucks were trained for semen collection prior to the main collection. A doe was used to tease the bucks and then semen was collected using artificial vagina. Semen was collected once daily from each buck fortnightly between 7.00 am and 9.00 am. This was to ensure that optimum quality semen was obtained. Each ejaculate was examined for volume, concentration, progressive motility, live sperm, dead sperm, pH and colour. Semen colour was observed immediately after semen collection. The semen ejaculate volume was measured using 4 ml calibrated collection tube immediately after the collection of semen. Sperm motility was determined on freshly collected semen placed on a clean slide covered with a coverslip and viewed under a microscope at a magnification of ×400. Sperm concentration was determined using a haemocytometer and Thomas-Zesis cell counter. The haemocytometer has microscopic grids which contained 25 large squares; each of the squares has 16 smaller squares which make a total of 400 squares. Sperm cells were counted diagonally from top left to bottom right, from top right to bottom left and in the middle to make a total of five counts from large squares (Rekwot *et al.*, 1997). Morphology of sperm cells, dead and live sperm was viewed in nigrosine-eosin stained slides under a microscope at a magnification of ×100 according to the procedure of Hackett and Macpherson (1965). pH (hydrogen ion) value was measured using Chemo craft

Table 1. Feed composition.

Ingredient	Percentage
Maize	45
GNC	35
Maize Offal	15
Bone meal	3
Salt	0.25
Vitamin/Minerals premix	0.25
Total	98.5
Calculated Analysis	
Energy(kcal ME/Kg)	2687.74
CP	20
CF	4.7
Ash	2.68

Vitamin/mineral premix content per kilogram ration: vitamin A1251IU, vitamin D32750IU, vitamin E 151IU, vitamin B2 0.006g, Nicotinic acid 0.035g, Calcium Dpantothenate 0.01mg, vitamin B6 0.035g, vitamin B12 0.02g, folic acid 0.001, Biotin 0.0003g, vitamin C 0.0025g, choline choride 0.39, Zinc bactracin 0.002g, methionine 0.2g, Avatee(Lasolocid) 0.09g, manganese 0.1g, Iron 0.05g, Zinc 0.04g, Copper 0.002g, Iodine 0.00153g, cobalt 0.00025g, Selenium 0.0001g.

indicator paper, the pH indicator paper is calibrated from 1 to 14 with various colours that show the readings of hydrogen ions. A part of the indicator paper was immersed in the semen, removed and placed on the colour chart the corresponding colour is recorded as the pH value.

Statistical analysis

The data generated from the experiment were statistically analyzed using the General Linear Model procedure of Statistical Analysis (SAS, 2002). Significant differences between means were separated by orthogonal pair wise difference of SAS (2002). Vitamin C, season, vitamin C and season interaction, month effect and month and vitamin C interaction were analyzed. The model used for this design is as follows:

$$Y_{ijkl} = \mu + T_i + S_k + (T_i \times S_k) + e_{ijkl}$$

Where: Y_{ijkl} = observation of the rabbit bucks responses, μ = population mean, T_i = treatment or month effect (vitamin C level or month), S_k = season effect, $T_i \times S_k$ = interaction effect, e_{ijkl} = random error.

RESULTS AND DISCUSSION

Table 2 shows the effect of vitamin C levels on semen characteristics of rabbit bucks. The results show a significant ($p < 0.01$) difference in all the traits with the exception of colour which was consistently milky at all levels. The sperm motility, concentration and live sperm

improved as vitamin C levels increased. Semen volume was highest at 100 mg followed by 400 mg vitamin C and lowest at 300 mg. In this study (data not included), water intake was significantly affected by vitamin C supplementation (Bisong, 2015). It is likely that seminal fluid production was affected by water intake considering the increasing spermatozoa concentration. A function of vitamin C in reproduction under environmental stress is to ameliorate reproductive performance with regards to sperm viability and normality. The significant differences obtained in this study across vitamin C levels do not corroborate the earlier report of Asadpour *et al.* (2011) who reported a non-significant effect of vitamin C levels on semen concentration among other traits. The higher values observed on 400 mg/kg feed, however, agrees with the reports of Metwally *et al.* (2009) and Fazeli *et al.* (2010) which indicated that semen concentration and other traits except for semen volume improved as vitamin C level increased in the diet.

El-Tohamy *et al.* (2012) reported that vitamin C improved the semen volume of rabbit bucks. This finding can be attributed to vitamin C being involved in the synthesis of sex steroids such as testosterone, and peptide hormones, as well as hydroxylation of steroids which is especially vitamin C dependent (Luck *et al.*, 1995; Weber *et al.*, 1996). The depressive effect of the low dose of vitamin C on sperm motility observed in this study is similar to the findings of Sonmez and Demirci (2003) who reported a 2% improvement in sperm motility of rams with vitamin C supplementation. This could further indicate that lower levels of vitamin C were not sufficient under tropical conditions to give the required ameliorative effects on spermatogenesis. The increase in live sperm cells with vitamin C levels in this study is at variance with the report of Fazeli *et al.* (2010) that there is no variation in live spermatozoa among dosage of vitamin C levels.

Table 3 presents the effect of monthly variations in the semen characteristics of rabbit bucks. There was a highly significant ($p < 0.01$) variation between the months. The mean semen volume was highest in the month of April (1.43 ml) while the lowest was recorded in the month of July (0.41 ml) with a downward trend from March to August (1.38 - 0.69 ml). Sperm motility values recorded in June were significantly ($p < 0.01$) higher (75.2%) than the values obtained in other months. Concentration was affected significantly ($p < 0.01$) as it increased between March and April ($75.72 - 129.48 \times 10^6$) while other months had fluctuations ranging between $90.62 - 102.78 \times 10^6$. The semen pH had slight fluctuations across the months while semen colour remained consistently milky. Live sperm cells increased from March (75.96%) to August (91.62%).

The milky colour of sperm observed in this study for all the months indicates the rabbits produced good quality sperm irrespective of the month. Salah *et al.* (1992) reported that semen ejaculate colour whether milky or creamy is independent of season and this is in line with this present study. The significant decrease in sperm motility observed within the months of March to May and increase

Table 2. Effect of vitamin C level on semen characteristics of rabbit bucks.

Parameter	Vitamin C level in feed (mg/kg feed)				P-value
	100	200	300	400	
Sperm motility (%)	63.19±2.86 ^c	66.90±2.88 ^{bc}	71.07± 3.09 ^{ab}	73.58±3.35 ^a	0.01
Concentration(x10 ⁶)	80.13±11.79 ^d	100.15±11.87 ^c	114.16±12.74 ^b	118.12±13.80 ^a	0.01
Semen volume (ml)	1.29±0.18 ^a	0.91±0.18 ^c	0.74±0.24 ^d	1.17±0.21 ^b	0.01
Semen pH	6.81±0.18 ^b	7.13±0.18 ^a	6.85±0.20 ^b	6.65±0.21 ^c	0.05
Colour	Milky	Milky	Milky	Milky	0.40
Live sperm (%)	84.79±1.46 ^b	84.05±1.41 ^b	85.69±1.78 ^b	89.63±1.50 ^a	0.01
Dead sperm (%)	15.05±1.47 ^b	15.95±1.43 ^b	14.27±1.79 ^b	10.37±1.60 ^a	0.01

^{abcd}Means within rows with different superscripts are significantly different (p<0.01).

in sperm motility within the months of June to August in this study was also reported by Al-ghalban *et al.* (2004). The steady increase in sperm motility observed from April to July in this study is similar to the report of Boland *et al.* (1985), who also reported a steady increase in sperm motility from April to August.

The high live sperm recorded in this study agrees with the findings of Chemineau *et al.* (1992) who reported variations in both semen quality and quantity due to the changes in daylight length throughout the year. Decreasing daylight in autumn generally causes physiological changes stimulating reproduction functions and increasing the sperm output (Sancho *et al.*, 2004; Chemineau *et al.*, 2007). Semen characteristics were generally better at the end of summer and the two first months of autumn, than during the winter and spring. Though changes in photoperiod in the tropics are not as dramatic as in the temperate regions, the results in this study indicate similar effects of improved semen quality from March to August. Sperm concentration significantly (p<0.01) increased as the months moved from hot dry (75.72 - 129.48 x 10⁶) to cool wet season (98.17 - 102.78 x 10⁶). This result is consistent with an earlier report by Elagib *et al.* (2012) that high temperatures reduced sperm concentration. Higher semen volume observed in the months of the hot dry season than in the cool wet season could be related to the increased spermatogenic activity of the testes. Nkanga and Egbunike (1988) related increases in semen characteristics in the dry season to an increase in day length which activates an increased output of the gonadotropic hormones of the anterior pituitary that leads to an increase in spermatogenic activity.

Table 4 presents the interaction between vitamin C levels and season on semen characteristics. There was a significant (p<0.01) interaction between vitamin C inclusion levels and season on sperm motility, concentration and live sperm. Sperm volume was higher in the hot dry than in the cool wet season for all levels of vitamin C, this is as a result of higher water intake in hot dry season. Live sperm and sperm motility increased in the cool wet than in the dry season for rabbit bucks as the vitamin C level increased while it decreased in the cool wet season for rabbits on 200 mg vitamin C. Semen pH was similar in the hot dry and cool wet season on 100 mg vitamin C, decreased on 200 mg

vitamin C and increased on 300 and 400mg vitamin C in the cool wet season compared to the hot dry season. High values recorded for volume in the hot dry season may be attributed to the level of vitamin C inclusion and favourable environment which supports the report of El-Tohamy *et al.* (2012). Lower values for live sperm recorded at 100 and 200 mg/kg feed vitamin C and higher values at 300 and 400 mg/kg feed in the hot dry season indicates amelioration of the effects of high temperature on spermatogenesis, in contrast, Fazeli *et al.* (2010) reported no variation in live spermatozoa with dosage of vitamin C.

The highly significant difference in live sperm in the cool wet season compared to the hot dry season agrees with El-Tohamy *et al.* (2012), who reported a significant increase in the live sperm in cool wet season than the hot dry season. Supplementation of ascorbic acid (up to 200 mg/kg diet) in the diets of breeder turkey toms has also been shown to improve semen volume and concentration (Dobrescu, 1987; Noll, 1993), an observation in line with what was recorded in the present study. Neuman *et al.* (2002) reported that supplementation of ascorbic acid up to 300 mg/kg in the diet of male turkey breeders did not affect the semen quality indices of volume, sperm concentration, dead spermatozoa, as well as the size of testes and this is contrary to the findings of this study. The improvement observed in sperm motility and live and dead spermatozoa of the bucks could be attributed to the anti-oxidative properties of vitamin C. The antioxidant vitamin C is inherently found in both the spermatozoa and the seminal plasma (Surai *et al.*, 2001). Vitamin C protects the DNA of spermatozoa from oxidative damage by scavenging reactive oxygen species, with resultant preservation of the integrity of the spermatozoa membranes and genes (Fraga *et al.*, 1991; Luck *et al.*, 1995). This phenomenon is thus the likely reason for the observed improvement in percentage sperm motility and live spermatozoa.

Sperm motility was high for bucks on all the vitamin C levels in all the months of study (Figure 1). This indicates a good ability of vitamin C in ameliorating the effects of heat stress on sperm motility despite changes in ambient temperatures from high (36-40°C) during March to May and moderate (32-36°C) during June to August when the rains result in the cooling effect. Figures 2 and 3 show variation

Table 3. Effect of monthly variation on semen quality of rabbit bucks.

Parameter	March	April	May	June	July	August	P-value
Semen vol. (ml)	1.38±0.32 ^b	1.43±0.24 ^a	1.32±0.20 ^b	0.95±0.18 ^c	0.41±0.25 ^e	0.69±0.32 ^d	0.02
Sperm motility (%)	67.45±5.0 ^c	60.01±3.7 ^d	70.96±3.3 ^b	75.2±3.0 ^a	70.05±3.8 ^b	61.97±5.0 ^d	0.01
Sperm conc. (x10 ⁶)	75.72±20.87 ^d	129.48±15.63 ^a	90.62±13.85 ^c	102.78±12.2 ^b	100.26±16.20 ^b	98.17±20.87 ^b	0.04
Semen pH	7.23±0.32 ^{ab}	6.45±0.24 ^c	6.65±0.21 ^c	6.89±0.18 ^b	7.04±0.25 ^a	6.93±0.32 ^a	0.03
Semen colour	Milky	Milky	Milky	Milky	Milky	Milky	-
Live sperm (%)	75.96±1.47 ^c	84.78±1.35 ^b	89.29±1.24 ^a	90.19±1.73 ^a	92.55±2.08 ^a	91.62±2.11 ^a	0.001
Dead sperm (%)	24.18±1.48 ^f	14.79±1.36 ^e	10.71±1.26 ^d	9.94±1.74 ^c	7.45±2.1 ^b	8.05±2.0 ^a	0.001

^{abcdefg} Means within rows with different superscripts are significantly different (p<0.01), vol = volume, conc = concentration.

Table 4. Effect of Interaction between Vitamin C level and season on semen quality of bucks.

Parameters	Season								p-value
	Hot dry	Cool wet	Hot dry	Cool wet	Hot dry	Cool wet	Hot dry	Cool wet	
Vitamin C level	100mg/kg feed	100mg/kg feed	200mg/kg feed	200mg/kg feed	300mg/kg feed	300mg/kg feed	400mg/kg feed	400mg/kg feed	
Semen vol. (ml)	1.60±0.2 ^a	0.98±0.3 ^c	1.10±0.2 ^b	0.76±0.3 ^d	1.10±0.3 ^b	0.43±0.3 ^e	1.57±0.3 ^{ab}	0.77±0.3 ^d	0.04
Sperm motility (%)	56.38±3.8 ^d	70.0±4.22 ^b	68.46±3.76 ^b	65.34±4.4 ^c	67.78±4.5 ^c	74.36±4.2 ^{ab}	70.90±5.2 ^b	76.25±4.15 ^a	0.01
Semen conc. (x10 ⁶)	68.9±15.9 ^e	91.3±17.4 ^c	114.9±15.5 ^b	85.4±17.2 ^d	111.5±18.6 ^{bc}	116.0±17.4 ^{ab}	114.5±21.6 ^b	121.6±17.1 ^a	0.01
Semen pH	6.8±0.24 ^c	6.81±0.27 ^c	7.19±0.24 ^a	7.07±0.28 ^b	6.63±0.29 ^c	7.07±0.3 ^b	6.40±0.33 ^d	6.91±0.26 ^c	0.6
Semen colour	Milky	Milky	Milky	Milky	Milky	Milky	Milky	Milky	-
Live sperm (%)	75.8±1.67 ^{de}	90.31±1.7 ^{ab}	79.688±1.7 ^d	89.32±1.8 ^b	81.32±1.9 ^{cd}	88.72±1.8 ^b	85.31±2.4 ^c	90.44±1.6 ^a	0.02
Dead sperm (%)	24.2±1.7 ^a	9.69±1.7 ^g	20.313±1.7 ^b	10.59±1.8 ^f	18.32±1.9 ^c	11.48±1.8 ^e	14.69±2.4 ^d	9.6±1.64 ^g	0.06

^{abcdefg} Means within rows with different superscripts are significantly different (p<0.01), vol = volume, conc. = concentration.

with a decreasing trend in semen volume, and an increasing trend in sperm concentration of bucks on the vitamin C levels from March to August. Al-ghalban *et al.* (2004) reported a significant decrease in semen volume within the months of March to May and a highly significant increase in semen volume from the months of June to August. Similar trends of monthly differences in semen volume have also been reported by Boland *et al.* (1985). Similarly, Obidi *et al.* (2008) reported semen volume decreased during the hot dry period of the year which normally occurs within the months of March to May; and a rapid increase in semen volume from the months of June to August has also

been reported by Tharwat *et al.* (1994).

Semen pH varied for the vitamin C levels in the months of April and May being similar in the months of March, July and August (Figure 4). El-Tohamy *et al.* (2012) reported a significant decrease in hydrogen ion of semen during increase in ambient temperature than in low ambient temperature. Salah *et al.* (1992) also reported a significant seasonal variation in semen pH during the hot dry and cool wet period of the year. The results of Fazeli *et al.* (2010) show that vitamin C reduces semen pH in animals administered vitamin C compared to un-administered animals which expressed high pH values. The values for semen colour, however,

varied widely in the month of April but not in the months of March, June to August being similar in May (Figure 5). Ambient temperature influences changes or variations in semen colour from creamy to milky and watery; this occurs from the hot dry season to the cool wet season of the year (Rai *et al.*, 1997). The variations in semen colour in different seasons of the year could be attributed to the variations in spermatozoa concentration and semen consistency (Zeidan *et al.*, 2000). Salah *et al.* (1992) reported on the contrary, that the ranges of semen ejaculate colour from light milky to creamy with the majority being milky white they observed, were not seasonal dependent.

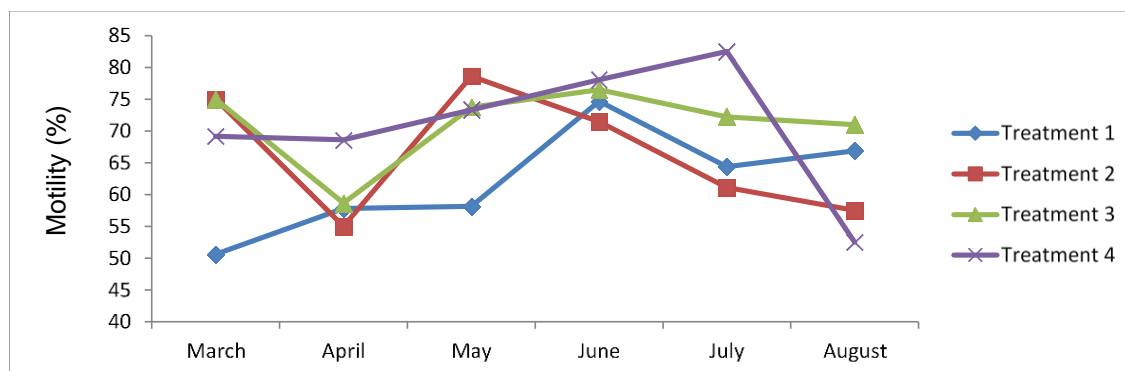


Figure 1. The interaction between monthly effect and vitamin C levels on semen motility. Treatment 1= 100mg; treatment 2= 200mg; treatment 3=300mg and treatment 4= 400 mg/kg diet vitamin C.

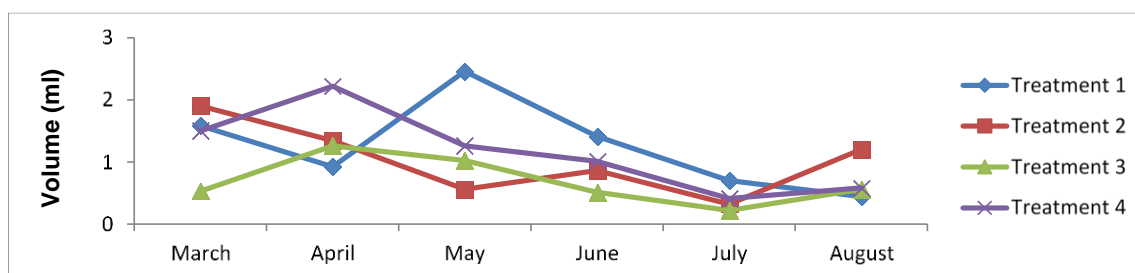


Figure 2. The interaction between monthly effect and vitamin C levels on semen volume. Treatment 1= 100mg; treatment 2= 200mg; treatment 3=300mg and treatment 4= 400 mg/kg diet vitamin C.

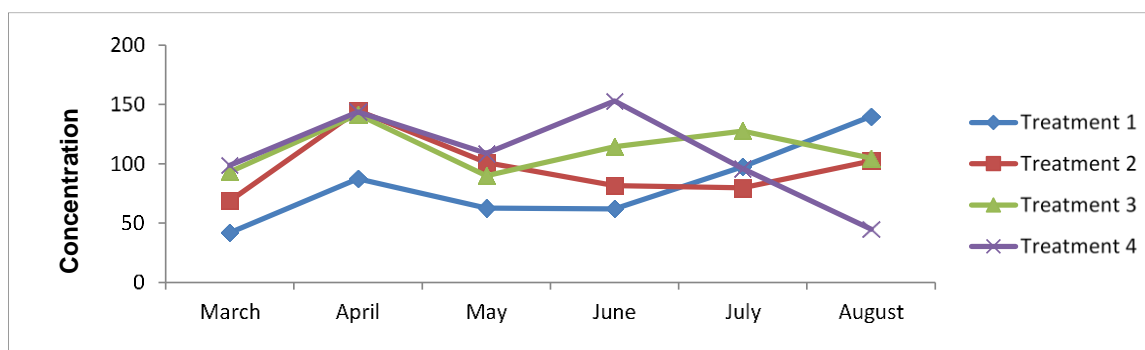


Figure 3. The interaction between monthly effect and vitamin C levels on semen concentration ($\times 10^6$). Treatment 1= 100mg; treatment 2= 200mg; treatment 3=300mg and treatment 4= 400 mg/kg diet vitamin C.

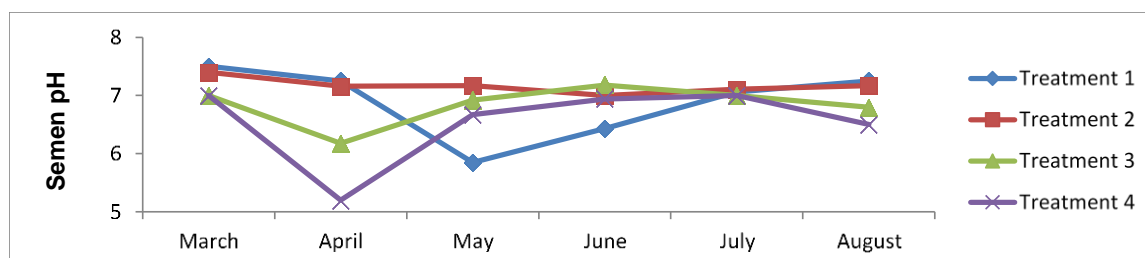


Figure 4. The interaction between monthly effect and vitamin C levels on semen pH. Treatment 1= 100mg; treatment 2= 200mg; treatment 3=300mg and treatment 4= 400 mg/kg diet vitamin C.

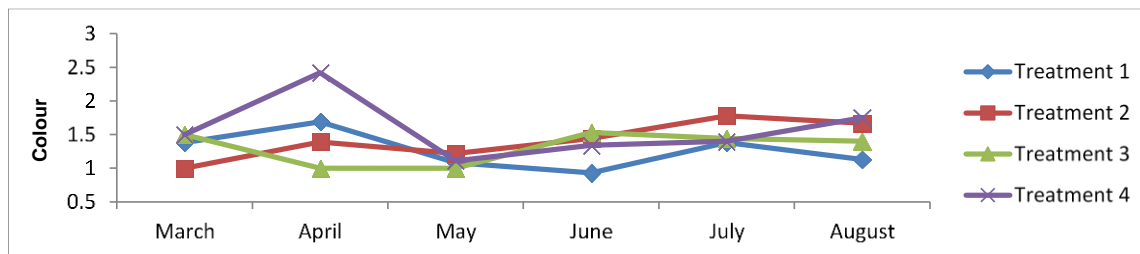


Figure 5. The interaction between monthly effect and vitamin C levels on semen colour. Treatment 1= 100mg; treatment 2= 200mg; treatment 3=300mg and treatment 4= 400 mg/kg diet vitamin C.

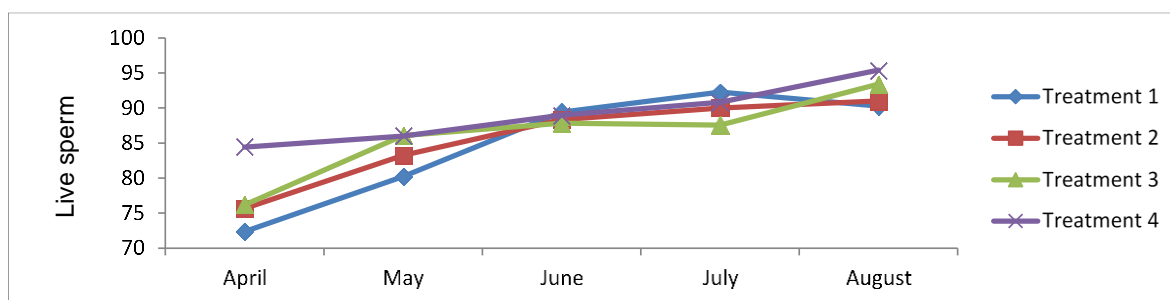


Figure 6. The interaction between monthly effect and vitamin C levels on live sperm cells (%). Treatment 1= 100mg; treatment 2= 200mg; treatment 3=300mg and treatment 4= 400 mg/kg diet vitamin C.

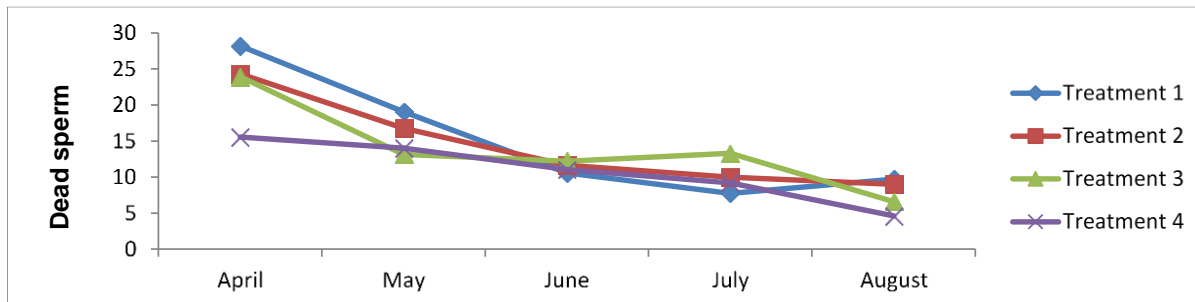


Figure 7. The interaction between monthly effect and vitamin C levels on dead sperm (%). Treatment 1= 100mg; treatment 2= 200mg; treatment 3=300mg and treatment 4= 400 mg/kg diet vitamin C.

Live sperm cells increased (Figure 6), while dead cells decreased (Figure 7) for all vitamin C levels from April to August. Hassan *et al.* (2012) reported that vitamin C decreased dead sperm in rabbit bucks under heat stress, which agrees with the results of this study. Ezzat *et al.* (2011) indicated that oral administration of 1.5 g of vitamin C reduced dead spermatozoa in rabbit bucks. Fazeli *et al.* (2010) also reported an increase in live spermatozoa and a decrease in dead spermatozoa in the vitamin C administered group than the none administered group. This, therefore, buttresses the ameliorative ability of vitamin C in improving sperm quality under heat stress conditions.

Conclusion and Recommendations

Based on the results, it was concluded that vitamin C

improved the semen characteristics of rabbit bucks at 400 mg/kg feed which directly translates to improved fertility potential. It is therefore recommended that especially during the hot dry season (March-May) or in times of heat stress, vitamin C at 400 mg/kg feed should be included in the diet of breeding rabbit bucks in the Northern Guinea Savanna of Nigeria.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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