

## **Influence of garlic (*Allium sativum*) and vitamin E on semen characteristics, reproductive performance and histopathology of rabbit bucks**

Ekuma, B. O., Amaduruonye, W., Onunkwo., D. N. and Herbert, U.

College of Animal Science and Animal Production,

Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria.



Corresponding Author's E-mail: donunkwo1@gmail.com, 08033388622

### **Abstract**

Researches have proven that garlic has medicinal, antimicrobial properties, speeds up digestion and widely used as preservatives, spice and condiment in many homes. Vitamin E on the hand is a fat soluble vitamin with potent antioxidant properties essential for the stabilization of biological membranes, protecting cells from oxidative stress and inhibits angiogenesis. Thus, a study using thirty-six (36) pre-pubertal New Zealand White rabbit bucks was conducted to investigate the physiological response of garlic and vitamin E supplementations on libido, gonadal and extra-gonadal sperm characteristics, reproductive hormone and testicular histology of rabbit bucks. The bucks were randomly assigned to four dietary treatments and replicated three times in a completely randomized design (CRD). Each dietary treatment consisted of 9 rabbits per treatment with 3 rabbits per replicate. Four dietary treatments were formulated to meet the nutrient requirements of rabbits supplemented at 0% garlic and vitamin E ( $T_1$ ), 3% garlic ( $T_2$ ), Vitamin E ( $T_3$ ), and 3% garlic and Vitamin E ( $T_4$ ) respectively. Data were collected on the libido, seminal characteristics, seminal plasma, testicular morphology and hormones. The testicles were collected and processed for histopathology. Data collected on different parameters were subjected to analysis of variance (ANOVA). Results showed that significantly ( $P < 0.05$ ) differences were observed in all the seminal characteristics parameters, on the testicular weights, volumes and on the reproductive hormones compared with the control. Testicular histology showed increased in number of seminiferous tubules with the supplementation. These results proved that both garlic and Vitamin E improve semen production both in quality and quantity and the overall reproductive performance. Moreover, combination of the garlic and vitamin E gives a better result. Therefore, garlic and vitamin E should be used together for better quantitative and qualitative semen production and reproductive performance in breeding animals

**Keywords:** Semen, testis, hormones, reproduction, histopathology, garlic, vitamin E

### **Introduction**

The nutrient requirement, protein requirement, economic growth and development for many countries have been partially met by animal production and consumption of animal products and by-products. Inadequate supply of proteins from such livestock as cattle, goat, sheep and pig has led to a shift of emphasis towards enhanced productivity and reproductive performance of these animals (Egboet *al.*, 2001). As such, the search for more economical source of animal proteins

makes rabbit production attractive. Topmost in the attributes promoting rabbit production includes its high fecundity, early sexual maturity, short gestation length, its litter size, fast growth rate, high prolificacy and its ability to convert roughages and concentrate into high quality proteins (Biobaku and Dosumo, 2003; Ani, 2006). In spite of all these comparative advantages over other livestock, the efficacy of rabbit production has not been fully elucidated (Herbert and Adejumo, 1995).

Reproductive failure has been a major source

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of economic loss in animal production. These failures have resulted from inefficient reproductive system, poor quality and quantity of semen production and semen characteristics which have been influenced by several factors (Hughes and Varley, 1980; Brown, 1984; Cupps, 1991; Buhr *et al.*, 1993; Kings, 1993; Bearden and Fuquary, 1997). Semen characteristics have been reported to be largely effected by nutrition, drugs, hormones and environmental factors (Rode *et al.*, 1995; Irvine, 1996; Bray *et al.*, 1997; Scot *et al.*, 1998 and Abdel-Rahman *et al.*, 2000). So there is great need to improve the semen quality and characteristics of these animals so as to enhance animal productivity and reproductive performance.

Due to its comparative efficacy, lesser side effect, availability and relatively cheap of these plants products compared to the synthetically produced reproductive enhancers, there has been an increasing interest in the use of plants, herbs, organic and natural products to improve reproductive performances and to ameliorate many reproductive disorders. As a result, plant materials are continuously being scrutinized and explored for their beneficial effects (Ansa *et al.*, 2017). One of such products is *Allium sativum*.

Garlic (*Allium sativum*) is a moderately tall annual herbaceous bulbous plant that belongs to the family of *Amaryllidaceae* of the genus *Allium*. The bulb consists of many separate strong flavored cloves which served as the storage organ for the plant (Block, 2010; Gafar *et al.*, 2012). Phytochemical analysis of garlic reveals that it contains enzymes, B-complex vitamins, proteins, minerals, flavonoids and many more sulfur containing compounds which are responsible for its spiciness, smell and taste (Focke *et al.*,

1990; Block, E., 1992; Ankri *et al.*, 1999; Jennifer, 2002). Researches have proved that garlic has medicinal, antimicrobial properties, speeds up digestion and widely used as preservatives, spice and condiment in many homes (Aaro *et al.*, 2001; Hammami and El May, 2012). Documented evidence have shown that garlic has been used in human medicine to treat rheumatoid arthritis, common cold, coughs, diabetes, malaria, diarrhea, cholera and tuberculosis for centuries (Turner, 2004; Gafar *et al.*, 2012). Furthermore, garlic has been reported to be an effective antibiotic, antioxidant, antifungal and antiviral agent and it improves the immune system (Turner, 2004; Andreatta *et al.*, 2005).

Vitamin E (Tocopherol) on the hand is a fat soluble vitamin with potent antioxidant properties essential for the stabilization of biological membranes, protecting cells from oxidative stress, inhibits angiogenesis, has anti-inflammatory activity. It has an anti-ageing property, improve healing process, memory functions, regulating immune functions, maintaining endothelial cell integrity, balancing normal coagulation, enhancing physiological and pathophysiological processes. It is a heart vitamin which has been found to improve circulation, cleans the arteries, inhibits cholesterol oxidation and its deposition in the walls of the arteries thereby prevents atherosclerosis (Wallert *et al.*, 2014; Mathur *et al.*, 2015; Botanical, 2016).

If the chemical and biological properties of garlic, vitamin E or its combinations are proved to enhance the growth and development of the male reproductive organs, semen characteristics, and the overall reproductive performance of rabbits in this study without exhibiting deleterious effects on the physiological characteristics of rabbits, its potential as reproductive supplements for breeding animals will

impact positively on animal production. Therefore, it becomes justifiable to investigate the physiological effects of garlic and vitamin E on the seminal characteristics, testicular developments, morphology and histology and on its overall impacts on the reproductive system of rabbit bucks.

**Materials and methods**

***Experimental Location***

This research was conducted in the Rabbitry Unit of the Teaching and Research Farm of the College of Animal Science and Animal Production, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria. Umudike is located in Abia state, Nigeria; at latitude 05°29' North and longitude 07°31' East; and at an altitude of 122 meters above sea level. It lies within the tropical rainforest zone of South-Eastern Nigeria. The location is characterized by average annual rainfall of 2,177mm in 148-

155 rain days. The average ambient temperature is 25.5°C with minimum and maximum temperature of 22°C and 29°C respectively. Relative humidity ranged from 57-91% (NRCRI, 2004).

***Collection and preparation of garlic meal***

Fresh garlic was purchased from Ubani market, in Umuahia, Abia State, Nigeria. It was cleaned and washed in running tap water to remove adhering debris and contaminants. Also, it was peeled and cut into small sizes, after which they were air-dried under room temperature to a constant weight. The dry garlic was milled using a hammer mill to produce garlic meal. The garlic meal was used in formulating the experimental diets. Four experimental rations were formulated containing 0% (garlic and vitamin E), 3.0% garlic, Vitamin E alone and 3.0% (garlic and vitamin E); representing T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub> respectively. The compositions of the experimental diets are presented in Table 1.

**Table1: Gross composition and calculated nutrients of experimental diets**

<b>Ingredients</b>	<b>Control (T<sub>1</sub>)</b>	<b>Garlic (T<sub>2</sub>)</b>	<b>Vitamin E (T<sub>3</sub>)</b>	<b>Garlic +Vit E (T<sub>4</sub>)</b>
Maize	44.94	44.94	44.94	44.94
Soya bean meal	17.31	17.31	17.31	17.31
Rice husk	32.00	32.00	32.00	32.00
Fishmeal	2.00	2.00	2.00	2.00
Bone meal	1.00	1.00	1.00	1.00
Limestone	2.00	2.00	2.00	2.00
Vit/min Premix*	0.25	0.25	0.25	0.25
Salt	0.50	0.50	0.50	0.50
Total	100.00	100.00	100.00	100.00
Garlic (%)	0.00	3.00	0.00	3.00
Vitamin E (IU/kg)	0.00	0.00	3000.00	3000.00
Calculated nutrients				
Crude Protein (%)	17.00	17.00	17.00	17.00
Metabolizable Energy (ME) (Kcal/kg diet)	2505.42	2505.42	2505.42	2505.42
Crude fiber (%)	11.36	11.36	11.36	11.36
Lysine (%)	0.514	0.514	0.514	0.514
Methionine (%)	0.199	0.199	0.199	0.199

\*Premix composition (per kg of diet): vitamin A, 12,500 IU; vitamin D3, 2500 IU; vitamin E, 50.00mg; vitamin K3, 2.50mg; vitamin B1, 3.00mg; vitamin B2, 6.00mg; vitamin B6, 6.00mg; niacin, 40mg; calcium pantothenate, 10mg; biotin, 0.08mg; vitamin B12, 0.25mg; folic acid, 1.00mg; chlorine chloride, 300mg; manganese, 100mg; iron, 50mg; zinc, 45mg; copper, 2.00mg; iodine, 1.55mg; cobalt, 0.25 mg; selenium, 0.10mg; antioxidant, 200mg.

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### ***Experimental animals and management***

Thirty-six pre-pubertal New Zealand White rabbit bucks were purchased from the Teaching and Research farm of the College of Animal Science and Animal Production, Michael Okpara University of Agriculture Umudike and from Bethel Rabbit Farms in Umuahia, Abia State, Nigeria. Two weeks pre-experimental period were used for quarantine, vaccination against ecto and endo-parasites and to get the rabbits acclimatized to the experimental procedures. The rabbits were randomly assigned to four experimental treatments and replicated three times with three rabbits per replicate in a completely randomized design. The experimental animals were housed in hutches throughout the experimental period. Feed and clean drinking water were provided *ad libitum*. Routine management practices were also carried out appropriately. The study lasted for 14 weeks (November to February).

### ***Experimental design***

The design for the study was a CRD with four treatments consisting of 0% (garlic and vitamin E), 3.0% garlic, Vitamin E alone and 3.0% (garlic and vitamin E); designated T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub> respectively. T<sub>1</sub> served as the control. The vitamin E was supplemented at 3000 IU/kg of feed. Nine rabbits were randomly assigned to each treatment and replicated three times. **The experimental model is as follows:**

$$Y_{ij} = \mu + T_i + e_{ij}$$

**Where** Y<sub>ij</sub> = individual observation on the rabbit characteristics.

$\mu$  = overall mean

T<sub>i</sub> = treatment effect

e<sub>ij</sub> = **random error assumed to be independently, identically and normally distributed with zero means and constant variances.**

### ***Data collection and evaluation***

Data were collected on the libido, seminal

characteristics, plasma and testicular morphology of the rabbit bucks. Blood samples were collected and used for reproductive hormonal assay. All weight measurements were done using an electronic weighing scale (OHAUS Champ II).

### ***Testicular parameters***

The scrotal circumference was taken using a measuring tape calibrated in centimeters. At the end of the 12-week experiment, three bucks from each replicate were slaughtered using a captive bolt and allowed to bleed. The internal organs were incised and weighed. The testis was incised and used to determine testicular dimensions. Weights of the testicles were recorded after the epididymis had been trimmed off. The volume of each testis was recorded using Archimedes' principle of water displacement.

### ***Estimation of semen characteristics***

Semen evaluation involved the estimation of both the macroscopic and microscopic indices. Semen volume was determined in milliliters directly from a calibrated glass collection tube attached to the artificial vagina (AV). Sperm cell concentration was determined using a haemocytometer. Total number of sperm was determined by multiplying semen volume by the sperm cell concentration. Sperm motility was determined subjectively in a drop of fresh semen on a glass slide covered with slip and examined using a microscope. Sperm morphology was determined by performing differential counts of the morphologically normal and abnormal shape of the spermatozoa using an eosin and nigrosin stain. Libido (Reaction time) was determined by observing the time taken (seconds) from exposure of the buck to a doe and the first copulation as recommended by Herbert and Acha (1995).

### ***Semen collection***

Artificial vagina (AV) with a calibrated glass

collection tube constructed by Herbert and Adejumo (1993; 1995) was used for semen collection. Two weeks prior to semen collection, the bucks were trained to serve an artificial vagina using a teaser rabbit doe. The semen was collected weekly between 9am and 10am. During semen collection, the rabbit doe was taken to the buck's cage and held in position for service. The AV was lubricated using glycerol and its temperature adjusted to 40-42°C. The artificial vagina was strategically placed under the belly of the doe such that the penis of the buck was introduced into the artificial vagina.

**Histological study**

At the end of the experiment, rabbits sampled from each replicate were slaughtered, its testicles collected and processed for histology. The tissue was embedded, dehydrated in an alcoholic solution of different concentration. Clearing and impregnation was done using xylene and paraffin wax respectively. The tissue was cut (Sectioned) using a microtome (Rotary Kepee Model KD 202A), stained with hematoxylin and eosin; and examined using a light microscope of different magnification according to the procedure described by Majumdar (1980), Ogbuewu *et al.* (2009) and Amao *et al.* (2012) for histological studies. The slides were examined for histological indicators to

observe possible degenerative changes on the testicular structure using a microscope connected to a computer system. A photomicrographic software - Phoenix Micro Image Analysis (2003) version 1.33 was used to project the slides on the computer for clear assessment. The slides were subsequently captured and printed for interpretation and documentation at the Physiology Laboratory of the College of Animal Science and Animal Production, Michael Okpara University of Agriculture, Umudike.

**Reaction time (libido)**

A matured doe (teaser) was introduced to the buck prior to semen collection to monitor their sex drive. The time in seconds it took for the rabbit bucks to sniff, groom and mount the female was recorded with a stop watch, and libido determined as described by Chibundu, 2005.

**Statistical analysis**

Data collected on different parameters were subjected to analysis of variance (ANOVA) in accordance with the methods of Steel *et al* (1980), significance means were separated according to Duncan's Multiple Range Test (Duncan, 1955).

**Results and discussion**

The result on semen characteristics of bucks fed diet supplemented with garlic and vitamin E are presented in Table 3.

**Table 2: Semen characteristics of buck rabbits fed diet supplemented with garlic and vitamin E.**

Parameter	Control (T <sub>1</sub> )	Garlic (T <sub>2</sub> )	Vitamin (T <sub>3</sub> )	Garlic +Vit E (T <sub>4</sub> )	SEM
Semen volume (ml)	0.56	0.56	0.53	0.60	0.17
Spermatozoa mass motility (%)	76.33 <sup>d</sup>	83.66 <sup>b</sup>	79.53 <sup>c</sup>	87.26 <sup>a</sup>	1.28
Spermatozoa proportion (%)	84.00 <sup>c</sup>	90.66 <sup>a</sup>	80.33 <sup>c</sup>	92.33 <sup>b</sup>	1.49
Sperm concentration (x10 <sup>6</sup> /mm <sup>3</sup> )	118.33 <sup>c</sup>	120.33 <sup>b</sup>	117.50 <sup>c</sup>	135.66 <sup>a</sup>	2.24
Total n o. of S perm/ejaculate (x10 <sup>9</sup> /mm <sup>3</sup> )	66.26 <sup>ab</sup>	68.23 <sup>ab</sup>	62.70 <sup>b</sup>	81.40 <sup>a</sup>	2.87
Normal spermatozoa proportion (%)	95.40 <sup>c</sup>	98.60 <sup>a</sup>	96.93 <sup>ac</sup>	97.60 <sup>a</sup>	0.43
Abnormal sperm proportion (%)	3.60 <sup>a</sup>	1.40 <sup>c</sup>	2.83 <sup>ab</sup>	2.00 <sup>ab</sup>	0.39
Total viable spermatozoa (x10 <sup>9</sup> /mm <sup>3</sup> )	515.03 <sup>b</sup>	569.96 <sup>b</sup>	474.83 <sup>b</sup>	693.83 <sup>a</sup>	29.23
Libido (s)	4.66	4.33	4.66	4.66	0.14

<sup>abcd</sup>; means with different superscripts along row s are significantly different (P <0.05). NS=Non-significant difference (P >0.05). SEM= Standard error of means

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From the results in Table 3, the libido and volumes of the semen produced in all the treatments were statistically similar with no significant ( $P>0.05$ ) difference with the control. There were significant ( $P<0.05$ ) differences on the Spermatozoa motility, Sperm proportion, Semen concentration, total number of sperm ejaculated, on the proportion of the normal and abnormal spermatozoa and on the total viable spermatozoa. The supplementation of garlic alone, combination of garlic and vitamin E significantly increases the sperm motility and spermatozoa proportion; while vitamin E alone reduces motility. The combination of garlic and vitamin E produced a better result. The supplementation of garlic and its combination with vitamin E significantly increases sperm concentration, while statistically similar results were obtained on the supplementation of vitamin E alone with the control. Total number of sperm and total number of viable spermatozoa are statistically similar in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub>; whereas it significantly increased in T<sub>4</sub>. The proportion of the normal spermatozoa increases significantly on the supplementation of garlic, vitamin E and its combination, whereas the proportion of abnormal sperm reduced significantly when compared with the control. From the foregoing results, it is observed that the combination of garlic and vitamin E gives a better result.

These findings are in line with those of Hammami and El-may (2009; 2012) that reported increase in the reproductive performance, semen constituents and fertilizing ability of the spermatozoa on adult male rats and on rabbit bucks fed diet supplemented with garlic and vitamin E respectively. The semen volume and semen concentration agrees favorably with the findings of Herbert and Acha (1995) and Bracket (2004) who reported that rabbit semen volume ranges between 0.4-0.71 ml and semen concentration range of  $50-350 \times 10^6/\text{mm}^3$  respectively. From these observations, it is inferred that garlic, vitamin E and its combination increases spermatogenesis. The combination of garlic and vitamin E may have complemented each other during spermatogenesis in the testicle thereby improving the quality and quantity of semen produced. The observed increase in sperm output accompanied with reduced proportion of abnormal sperm showed that the supplementations positively affected the structure of the spermatogenic cells and the leydig cells during the process of spermatogenesis and ejaculation. The significant reduction in the percentages of abnormal sperm cells following supplementations showed a good reproductive potential and fertility in either normal mating or artificial insemination. The result of the seminal plasma parameters of bucks fed diet supplemented with garlic and vitamin E are presented in Table 4.

**Table 3: Seminal plasma characteristics of bucks fed diet supplemented garlic and vitamin E**

Parameter (mmol/l)	Control (T <sub>1</sub> )	Garlic (T <sub>2</sub> )	Vitamin E (T <sub>3</sub> )	Garlic +Vit E (T <sub>4</sub> )	SEM
Fructose	0.42 <sup>a</sup>	0.54 <sup>a</sup>	0.40 <sup>ab</sup>	0.29 <sup>a</sup>	0.03
Glucose	52.30 <sup>a</sup>	38.34 <sup>bc</sup>	31.10 <sup>b</sup>	24.07 <sup>b</sup>	3.40
Glutamic acid	2.36	2.62	2.68	2.56	0.06
Potassium	13.43 <sup>a</sup>	12.67 <sup>a</sup>	6.92 <sup>b</sup>	14.90 <sup>a</sup>	0.99
Zinc	2.38	2.30	2.21	2.33	0.08
Chloride	177.88 <sup>a</sup>	134.17 <sup>ab</sup>	86.15 <sup>c</sup>	169.10 <sup>ab</sup>	11.93
Iron	4.05 <sup>a</sup>	0.92 <sup>d</sup>	3.85 <sup>b</sup>	2.75 <sup>c</sup>	0.37
Magnesium	4.38	4.70	4.63	4.45	0.42
Phosphorus	1.26 <sup>a</sup>	1.20 <sup>c</sup>	1.76 <sup>b</sup>	2.72 <sup>a</sup>	0.18

<sup>abcd</sup> means with different superscripts along rows are significantly different ( $P<0.05$ ). NS=Non-significant difference ( $P>0.05$ ). SEM=standard error of means

From the results in Table 4, the seminal glutamic acid, zinc and magnesium was not affected by dietary supplementations. There was equal absorption of these electrolytes in all the treatments compared to the control. There were significant ( $P<0.05$ ) reduction on the seminal glucose and iron in all the dietary supplementation. The level of fructose reduced in  $T_3$  and  $T_4$ , while  $T_2$  are statistically similar with the control. The level of potassium reduced in vitamin E supplementation, while garlic alone and its combination were not

affected. Seminal chlorides were reduced by garlic alone and vitamin E alone; while its combinations were not affected. Supplementation of vitamin E alone and its combination significantly increased the level of phosphorus in the semen; while  $T_2$  are similar with the control. From these results, it could be inferred that garlic alone, vitamin E alone or its combination have little or no effect on the seminal electrolytes. The result on morphology of rabbit bucks fed diet supplemented with garlic and vitamin E are presented in Table 5.

**Table 4: Testicular morphology of buck rabbits fed diet supplemented with garlic and vitamin E**

Parameter	Control (T <sub>1</sub> )	Garlic (T <sub>2</sub> )	Vitamin E (T <sub>3</sub> )	Garlic +Vit E (T <sub>4</sub> )	SEM
Testis weight (g) – Right	3.98 <sup>bc</sup>	4.95 <sup>ab</sup>	4.02 <sup>ab</sup>	5.36 <sup>a</sup>	0.19
Testis weight (g)– left	4.89	4.85	4.83	5.42	0.15
Paired testis weight (g)	8.59 <sup>ab</sup>	9.80 <sup>ab</sup>	8.85 <sup>b</sup>	11.14 <sup>a</sup>	0.37
Epididymis weight (g) –Right	1.87	2.02	1.97	2.08	0.12
Epididymis weight (g)– left	2.00	2.05	1.95	2.05	0.06
Paired weight of epididymis	3.82	4.06	3.97	4.14	0.17
Testes volume (ml) – Right	1.95 <sup>c</sup>	2.87 <sup>b</sup>	2.61 <sup>b</sup>	3.10 <sup>a</sup>	0.31
Tests is volume (ml) – Left	2.27	3.47	2.98	3.16	0.27
Paired testis volume (ml)	4.27 <sup>b</sup>	6.33 <sup>a</sup>	5.59 <sup>a</sup>	6.26 <sup>c</sup>	0.50
Mean Scrotal Circumference (cm)	1.90	1.93	1.90	1.97	0.02
Tunicalalbuginea Weight(g) – Right	0.66	0.88	0.74	0.93	0.09
Tunicalalbuginea Weight(g) – Left	0.67	0.77	0.74	0.79	0.03
Paired Tunicalalbuginea Weight(g)	1.33	1.68	1.48	1.73	0.11

<sup>abcd</sup>: means with different superscripts along rows are significantly different ( $P<0.05$ ). NS=Non-significant difference ( $P>0.05$ ). SEM= Standard error of means.

Apart from the testicular weight and volume, all the other parameters measured were not statistically different ( $P>0.05$ ) although there were numerical increases compared to the control. The supplementation of garlic, vitamin E and its combination significantly increased the weight, volume and the development of the testicles. This is advantageous to an animal breeder. Seminal characteristics, testicular weights, sizes and morphology are good indicator of the present and future spermatozoa production, breeding and fertility potentials of a male animal (Gupta

and Mohanty 2003, Herbert *et al.*, 2005; Togu and Egbunike, 2006). The higher the weight and volume of a testicle without inflammation, the more the number of sertoli cells, spermatogenic cells, the leydig cells, seminiferous tubule and the surface area for spermatogenesis, then the better the breeding and fertility potentials of a male animal. This means that the higher weights and volumes of testis from the supplementations of garlic, vitamin E and its combination compared to the control resulted to a higher number of seminiferous tubules, leydig cells, sertoli

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cells and spermatogenic cells for spermatogenesis. The bigger the testis, the better the reproductive performance. This is inferred that garlic, vitamin E and its combination can be supplemented to the

diets of breeding animals to improve the quantity and quality of semen production. The result of the reproductive hormones of buck rabbits fed diet supplemented with garlic and vitamin E are presented in Table 6.

**Table 5: Male reproductive hormone of bucks fed diets supplemented with garlic and vitamin E**

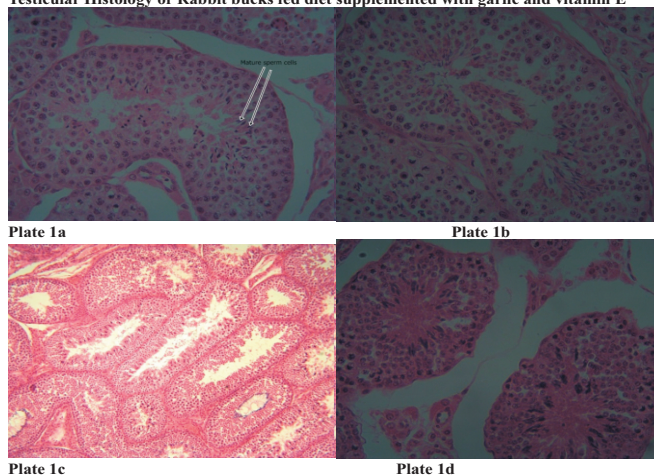
Parameters	Control (T <sub>1</sub> )	Garlic (T <sub>2</sub> )	Vitamin E (T <sub>3</sub> )	Garlic +Vit E (T <sub>4</sub> )	SEM
FSH	8.11 <sup>d</sup>	27.40 <sup>b</sup>	18.47 <sup>c</sup>	29.37 <sup>a</sup>	3.12
Luteinizing Hormone	2.64 <sup>a</sup>	1.58 <sup>c</sup>	1.00 <sup>d</sup>	1.70 <sup>b</sup>	0.61
Testosterone	1.34 <sup>d</sup>	2.03 <sup>b</sup>	1.56 <sup>c</sup>	2.59 <sup>a</sup>	0.14

<sup>abcd</sup>: means with different superscripts along row are significantly different (P <0.05). NS=Non-significant difference (P >0.05). SEM=standard error of means

Bucks in all the treatments had significantly (P<0.05) higher Follicle Stimulating Hormone and testosterone compared to the control. There were significant (P<0.05) reduction on the level of LH compared to the bucks on control. The result indicates that the dietary supplementation of garlic, Vitamin E and its combination affected the hormonal concentrations of the animals. The increase in FSH and testosterone as observed in T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub> had a positive impact on the semen characteristics (Table 2). This result is in line with those obtained by Oi *et al.* (2001) that showed an increase in testosterone levels due to the increase in the luteinizing hormones after administration of 8g of

garlic powder in albino rats. These increases in FSH and testosterone might have been because of the significant increase in the quality and quantity of semen produced. The resultant increases in the level of testosterone and FSH enhanced the activities and responsiveness of the Leydig cells, Sertoli cells and spermatogenic cells during spermatogenesis, thereby increasing semen production. Testosterone is essential for spermatogenesis, induces the male secondary sexual characteristics, while FSH stimulates the growth and development of the interstitial cells (Flaws, 1991). The increases in these hormones might have been responsible for the increase in the weights and volumes of the testis (Table 4).

**Testicular Histology of Rabbit bucks fed diet supplemented with garlic and vitamin E**



**Plate 1:** Light photomicrograph showing histology of rabbit testis with its seminiferous tubules  
 (a) Section of control testis, H&E X400 (b) Section of testis on garlic supplementation, H&E X400  
 (c) Section of testis on vitamin E supplementation, H&E X400 (d) Section of testis on garlic and vitamin E supplementation, H&E X400.

The histological sections of the testis from different treatments revealed the presence of intact seminiferous tubules and interstitial spaces which are highly convoluted lined by germinal epithelium with germ cells compared to the control. The various stages of spermatogenesis and spermeogenesis (spermatogonia, spermatocytes, round spermatids and elongated spermatids) are consistent with normal spermatogenesis. The sertoli cells which support and nourish the spermatozoa, a high activity of spermatogonia and spermatocyte near the base of the lumen were seen. High densely populated area of seminiferous tubules and compact interstitial spaces were observed with the interstitial cells of leydig occurring in clusters of various sizes with a dense population of mature sperm cells simplifying high proliferative activities. The compact arrangement of the inter-tubular spaces is evidence of high proliferative activity on the testis on supplementation of garlic and Vitamin E compare to the control.

### **Conclusion**

The study showed that garlic, vitamin E and its combinations improves semen production, seminal characteristics, promotes sperm concentration, spermatozoa proportion, number of sperm per ejaculate, spermatozoa motility, its viability and reduces abnormal spermatozoa. They stimulate the production of some reproductive hormones, enhanced the growth and development of the male reproductive organs and improve the reproductive performance of the rabbit bucks. Furthermore, the bigger the testicles in the absence of any inflammation the better the reproductive performance and the breeding ability of the rabbit buck. The study revealed that both garlic and

vitamin E improve the quality of semen production and the reproductive performance of the rabbit bucks. However, combination of garlic and vitamin E gave a better result. Based on these findings, both garlic and vitamin E may be combined so as to complement each other for better quantitative and qualitative semen production, hormone production and reproductive performances in breeding rabbits.

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