### **EFFECT OF GARLIC** (*Allium sativum*) **ON HEMATOLOGICAL**, **BIOCHEMICAL**, **HORMONAL AND FERTILITY PARAMETERS OF MALE BOUSCAT RABBITS**

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ABSTRACT: The present study was carried out on a flock of Bouscat rabbits belongs to El-Gemaiza Experimental Station, Animal Production Research Institute, Agriculture Research Center, Ministry of Agriculture, Egypt.

The experiment study lasted two months (from 6 up to 8 months of age) to investigate the effects of garlic treatment hematological, on biochemical, hormonal and fertility parameters of male Bouscat rabbits. For this purpose, twenty four male Bouscat rabbits weighted 3150-3300 distributed were into four g with experimental groups equal number (n = 6). The first group was used as a control and subcutaneously injected with saline solution (1 ml of 0.9% NaCl), the second, third and fourth groups were subcutaneously injected once a week for 8 weeks with garlic 3, 9 and 27 mg/kg body weight, respectively.

The results are summarized as follow:

The total numbers of RBCs and WBCs counts as well as Hb concentration in male rabbits treated with different doses of garlic (3, 9 or 27 mg/kg body weight, once a week for 8 weeks) were significantly ( $P \leq$ 0.05 or  $P \leq 0.01$  increased by increasing the dose of garlic. Lower dose of garlic (3 mg/kg body weight) showed slight increases ( $P \le 0.05$ ) in total protein and albumin levels and a slight decrease ( $P \le 0.05$ ) in globulin level, the higher doses (9 and 27 mg/kg body weight) showed marked decreases ( $P \leq 0.05$ ) in total protein, albumin and globulin levels. The total lipid, total cholesterol and triglyceride levels in male rabbits treated with different doses of garlic were decreased by increasing the dose of garlic, this decrease was statistically significant ( $P \le 0.05$  or P  $\leq 0.01$ ) with the higher doses (9 and 27 mg/kg body weight). Garlic treatment to buck rabbits elicited a slight increase in high-density lipoprotein (HDL) level, while it

showed marked decreases in lowdensity lipoprotein (LDL) and very lipoprotein low-density (VLDL) levels. The activity of AST enzyme for buck rabbits treated with different doses of garlic was insignificantly changed by increasing the dose of garlic. While, the lower dose of garlic (3 mg/kg body weight) slightly decreased ( $P \leq 0.05$ ), ALT and ALP enzyme activities, the higher doses (9) and 27 mg/kg body weight) showed increases ( $P \le 0.05$ ) in ALT and ALP enzyme activities of male rabbits. Treatment of buck rabbits with different doses of garlic slightly insignificantly decreased the urea level and slightly insignificantly increased creatinine level. Testosterone levels in rabbits treated with different doses of garlic were increased by increasing the dose of garlic, however, this increase was statistically significant ( $P \le 0.05$ ) with the doses 9 and 27 mg/kg. Lower dose

of garlic (3 mg/kg body weight) elicited increases in the ejaculate volume. concentration, the total output, the wave motions, the motility percentages, the percentages of live spermatozoa and the percentages of abnormal spermatozoa. However, the higher doses (9 and 27 mg/kg body weight) showed decreases in the ejaculate volumes, the wave motions and the motility percentages, but increased concentrations and abnormal spermatozoa.

**Conclusively,** from the present results it can be concluded that the lower doses of garlic can be safely used however, the higher doses may cause problems. The lower dose of garlic improved fertility parameters of buck rabbits.

**Key words**: Blood hematological and biochemical, hormones, semen quality and buck rabbits.

The domestic rabbit is considered a good laboratory animal for production because of its early sexual maturity, high prolificacy, short gestation and generation periods, sizable number of progeny kindled per doe, rapid growth and good meat quality. The potency of garlic has been acknowledged for over 5000 years. Garlic (*Allium sativum*), a member of the family Liliaceae, is a common food spice widely distributed and used all over the world as a condiment in various prepared food. Although there are many garlic supplements commercially available, they fall into one of four categories: fresh garlic, garlic oil, garlic powder and aged garlic extract (Tattelman, 2005).

The health benefits of garlic are likely arising from its wide variety of components. Garlic contains more than 200 chemicals. It contains sulfur compounds (allicin, alliin and agoene), volatile oils, enzymes (allinase, peroxidase and miracynase), carbohydrates (sucrose and glucose), and minerals

(selenium). It also contains amino acids (cysteine, glutamine, isoleucine and methionine), which help to protect cells from the harms of free radicals, bioflavonoids (quercetin and cyanidin, allistatin I and allistatin II and vitamins

C, E and A), which help to protect us from oxidation agents and free radicals (Ayaz and Alposy, 2007).

The traditional medical practitioners have considered garlic as an excellent medicinal plant that has much therapeutic potential. Garlic and its constituents have been widely recognized as agents for prevention and treatment of cardiovascular diseases (Eilat-Adar *et al.*, 2013), as well as for inhibition of tumors development, reduction of tumor mass and decrease the number of mitotic cells within tumors (Wallace *et al.*, 2013). Previous studies have shown that garlic preparations possess many biological activities including antiparasitic (Salama *et al.*, 2014), antifungal (Suleiman and Abdallah, 2014), antidiabetic (Rios *et al.*, 2015), antibacterial (Shaheen *et al.*, 2015) and antioxidative (Asdaq, 2015) activities.

A number of reports investigated the effect of garlic supplementation or co-administration of garlic and other supplements on male reproductive performance of different animal models. Garlic supplementation increased spermatogenesis and the quality of semen parameters in male rabbits (El-Amary and Abou-Warda, 2007). However, Hosseini and Khaki (2014) reported that garlic consumption decreased the quality of semen parameters in male rats. Concerning the male reproductive hormone (testosterone), dietary garlic increased testosterone level in male rats fed a high fatty diet (Omotoso *et al.*, 2012). Other studies reported that garlic supplementation decreased testosterone level in male rats (Ebomoyi and Ahumibe, 2010).

Due to high nutritional value and medicinal activity of garlic, as well as, the need to improve reproductive performance in rabbits; the present study was designed to evaluate the effect of garlic treatment (3, 9 and 27 mg /kg body weight injected subcutaneously once a week for 8 weeks) on physiological and reproductive parameters of buck Bouscat rabbits.

### MATERIALS AND METHODS

The present study was carried out on a flock of Bouscat rabbits belongs to El-Gemaiza Experimental Station, Animal Production Research Institute, Agriculture Research Center, Ministry of Agriculture, Egypt.

The experiment study lasted two months (from 6 up to 8 months of age) to investigate the effects of garlic treatment on hematological, biochemical, hormonal and fertility parameters of male Bouscat rabbits. For this purpose, twenty four male Bouscat rabbits weighted 3150-3300 g were distributed into four experimental groups with equal number (n = 6). The first group was used

as a control and subcutaneously injected with saline solution (1 ml of 0.9% NaCl), the second, third and fourth groups were subcutaneously injected with garlic 3, 9 and 27 mg/kg body weight, respectively. Garlic powder preparation, Tomex (ATOS Pharma, Cairo, Egypt) was dissolved in physiological saline, and doses of 3, 9 and 27 mg/kg body weight were subcutaneously injected in the neck region of rabbits once a week for 8 weeks.

Blood samples (5 ml/each rabbit) were collected from the ear vein of male rabbits. Each sample was divided into two tubes; the first was heparinized and the second was non-heparinized. The heparinized blood samples were used to test hematological parameters. Non-heparinized blood samples were immediately centrifuged at 3000 r.p.m. for 15 minutes and serum was separated, frozen under -20°C, and kept for biochemical and hormonal assessment. Red blood cells (RBCs), white blood cells (WBCs) counts and hemoglobin (Hb) concentration were determined as described by Emad El-Eslam (1997).

Serum levels of total protein and albumen were determined according to Henry (1964) and Doumas *et al.* (1971), respectively, using commercial kits (Diamond diagnostics). The globulin value was obtained by subtracting the value of albumen from the corresponding value of total protein. Total lipid, high-density lipoprotein, low-density lipoprotein and very low-density lipoprotein levels were determined according to Zöllner and Kirsch (1962), while total cholesterol and triglyceride levels were determined according to Richmond (1973) and Fassati and Prencipe (1982), respectively, using commercial kits (Diamond diagnostics). The activity of aspartate aminotransferase (AST) and alanine amino-transferase (ALT) were assayed according to Reitman and Frankel (1957) and the activity of alkaline phosphate (ALP) was assayed according to Belfield and Goldberg (1971). Urea-N and creatinine levels were determined using commercial kits (Diamond diagnostics) according to the method of Patton and Crouch (1977) and Henry (1974), respectively.

Serum level of testosterone was measured using Coat-A-Count Total Testosterone (PITKTT-5, 2006-12-29) radioimmunoassay kits according to the method of Demetriou (1987). Serum level of tri-iodothyronine (T<sub>3</sub>) was measured using Coat-A-Count Total T<sub>3</sub> (PITKT3-7, 2009-07-16) radioimmunoassay kits according to the method of Hollander and Shenkman (1974). Serum level of thyroxine (T<sub>4</sub>) was measured using Coat-A-Count Total T<sub>4</sub> (PITKT4-4, 2006-03-18) radioimmunoassay kits according to the method of Albertini and Ekins (1982).

After four weeks of injection, semen samples were collected twice a week for four weeks using an artificial vagina device as described by Walton (1958). Ejaculate volume, sperm concentration, total output, wave motion,

sperm motility %, live spermatozoa % and abnormal spermatozoa % were estimated according to the procedures of El-Kelawy (1993).

The bucks were housed separately in individual flat deck batteries (50 x 60 x 40 cm) with universal specification, provided with galvanized feeders and automatic drinkers. All batteries were located in a naturally ventilated room. Buck rabbits were fed *ad libitum* a commercial pelleted ration containing 16.95% crude protein, 12.01 crude fibre and 2.77 fat and 7.95 ash. Clean fresh tape water was available at all the times *ad libitum* throughout the experimental period. All bucks were kept under the same managerial, hygienic and environmental conditions.

Least Square Maximum Likelihood method of analysis (SPSS, Statistics Users Guide, Version 10) was used to analyze the obtained data according to the formula: Yij =  $\mu$  + Ti + eij, where: Yij is any observation,  $\mu$  is the overall mean of observation, Ti is the effect of treatment and eij is the random error. Duncan's New Multiple Range test was used for multiple comparisons (Duncan, 1955).

### **RESULTS AND DISCUSSION**

### Effect of garlic treatment on blood indices:

Data in Table 1 showed that the total numbers of RBCs and WBCs counts as well as Hb concentration in male rabbits treated with different doses of garlic (3, 9 or 27 mg/kg body weight, once a week for 8 weeks) were significantly ( $P \le 0.05$  or  $P \le 0.01$ ) increased by increasing the dose of garlic. The percentages of this increase reached 8.1, 24.6 and 38.9% for RBCs , 30.7, 37.7 and 79.4% for WBCs and 7.2, 24.2 and 38.5% for Hb as compared with that of the corresponding control value, respectively.

Garlic treatment increased the number of RBCs, WBCs counts and Hb concentration in male rabbits (Al-Jowari, 2014). Garlic significantly prevented the reduction of RBCs caused by lead intoxification (Ouarda and Abd-Ennour, 2011) in rabbits. However, Suleria *et al.* (2013b) reported that rabbits treated with garlic showed insignificant reduction in RBCs count. Fazlolahzadeh *et al.* (2011) suggested that garlic contains some constituents that may play a role in the function of organs related to blood cell formation such as thymus, spleen, and bone marrow to stimulate more blood production. In addition, Samson *et al.* (2012) suggested garlic compounds might have a stimulatory effect on some haematopoetic growth factors (cytokines) which interact with specific receptors on the surface of haematopoietic cells, regulating the proliferation and differentiation of progenitor cells and the maturation and functioning of mature cells. Chemical components of garlic seem to act as active oxygen scavenger competes with hemoglobin in the RBCs for oxygen

resulting in tissue hypoxia, which in turn stimulates the kidney to form and secrete erythropoietin. The end-product of metabolism of garlic in the body **Table 1:** Effect of garlic treatment on red blood cells (RBCs), white blood cells

(WBCs) counts and hemoglobin (Hb) concentration of male Bouscat rabbits.

|                 | RBCs                              |                | WBCs                              |                | Hb                       |                |
|-----------------|-----------------------------------|----------------|-----------------------------------|----------------|--------------------------|----------------|
| Treatment       | x10 <sup>6</sup> /mm <sup>3</sup> | % of<br>change | x10 <sup>3</sup> /mm <sup>3</sup> | % of<br>change | concentration            | % of<br>change |
| Control         | $4.91\pm0.5^{b}$                  | -              | $7.19\pm0.3^{c}$                  | -              | $9.90\pm0.9^{b}$         | -              |
| Garlic(3 mg/kg) | $5.31 \pm 0.7^{ab}$               | 8.1            | $9.40\pm0.4^{b}$                  | 30.7           | 10.61 ±0.7 <sup>ab</sup> | 7.2            |
| Garlic(9 mg/kg) | $6.12\pm0.6^{a}$                  | 24.6           | $9.90\pm0.6^{b}$                  | 37.7           | $12.30\pm0.9^{b}$        | 24.2           |
| Garlic(27mg/kg) | $6.82\pm0.5^a$                    | 38.9           | $12.90 \pm 0.8^{a}$               | 79.4           | $13.71\pm0.8^{\rm a}$    | 38.5           |
| Significance    | *                                 | -              | **                                | -              | *                        | -              |

a, b and c means with different super-script in the same column, differ significantly  $P \le 0.05$ . \* =  $P \le 0.05$  and \*\* =  $P \le 0.01$ .

may also, step up Hb synthesis and RBC production by their indirect effect on erythropoietin (Fazlolahzadeh *et al.*, 2011). Also, garlic contain natural sulfur compounds which act as antioxidant active substances that implies the antioxidant action of garlic sulfhydryl groups on RBCs counts (Attia and Ali, 1993). Moreover, William (1999) reported that several vitamins like vitamin  $B_1$ ,  $B_2$ ,  $B_6$ ,  $B_9$ , C and E are present in garlic have a role in RBCs formation, maturation and in hemoglobin biosynthesis, absorption and utilization. Regarding effect of garlic on WBCs count, Onu and Aja (2011) reported garlic might help in boosting the immune system of the rabbits. Iranloye (2002) suggested the anti-infection properties of garlic that stimulate immune functions. Also, garlic possess some important phytochemicals such as flavonoids, steroidal glycosides, alkaloids, saponins, tannins, phenolics, pectin and amino acids, with their biological and physiological roles to stimulate the immune system and organs related to blood cell formation particularly the bone marrow (Jeorg and Lee, 1998).

### Effect of garlic treatment on serum protein fractions:

The present study showed that the lower dose of garlic (3 mg/kg body weight) showed slight increases ( $P \le 0.05$ ) in total protein and albumin levels and a slight decrease( $P \le 0.05$ ) in globulin level, the higher doses (9 and 27 mg/kg body weight) showed marked decreases ( $P \le 0.05$ ) in total protein, albumin and globulin levels (Table 2). The percentages of this change reached 10.8, -22.5 and -25.6% for total protein, 26.3, -13.2 and -7.9% for albumin and -8.5, -34.2 and -47.6% for globulin as compared with that of the corresponding

control value, respectively. The A/G ratios in male rabbits treated with different doses of garlic were insignificantly increased by increasing the dose of garlic. **Table 2:** Effect of garlic treatment on serum total protein, albumin (A) and

|       | Total protein   | A   | Albumin (A)  | G    | obulin(G | 9     |    |      |
|-------|-----------------|-----|--------------|------|----------|-------|----|------|
| Bous  | cat rabbits.    |     |              |      |          |       |    |      |
| globu | ulin (G) levels | and | albumin/glob | ulin | (A/G)    | ratio | of | male |

| Treatment        | Total pro            | otein          | Albumin (A) G  |             | Globulin             | Globulin(G)    |              |
|------------------|----------------------|----------------|----------------|-------------|----------------------|----------------|--------------|
|                  | g/dl                 | % of<br>change | g/dl           | % of change | g/dl                 | % of<br>change | ratio        |
| Control          | $6.87\pm0.5^{\rm a}$ | -              | $3.80\pm0.5$   | -           | $3.07\pm0.4^{\rm a}$ | -              | $1.24\pm0.4$ |
| Garlic(3 mg/kg)  | $7.61 \pm 0.7^{a}$   | 10.8           | $4.80\pm0.6$   | 26.3        | $2.81\pm0.5^{\rm a}$ | - 8.5          | $1.71\pm0.4$ |
| Garlic(9 mg/kg)  | $5.32 \pm 0.5^{b}$   | - 22.5         | $3.30\pm0.2$   | - 13.2      | $2.02\pm0.3^{\rm b}$ | - 34.2         | $1.63\pm0.2$ |
| Garlic(27 mg/kg) | $5.11 \pm 0.2^{b}$   | - 25.6         | $3.50 \pm 0.3$ | - 7.9       | $1.61\pm0.2^{\rm b}$ | - 47.6         | $2.17\pm0.6$ |
| Significance     | *                    | -              | NS             | -           | *                    | -              | NS           |

a and b means with different super-script in the same column, differ significantly ( $P \le 0.05$ ). \* =  $P \le 0.05$  and NS = Not significant.

Albumins and globulins are two key components of serum proteins. As albumin synthesizes in the liver, it can be used as a biomarker to monitor liver function (Friedman *et al.*, 1980). Hussein *et al.* (2007) suggested that garlic has ability to stimulate the regeneration of hepatic tissue, which increases protein synthesis in damage liver, improves the functional status of the liver cells and prevent protein oxidation. In addition, it is believed that garlic affect whole body protein metabolism through hormonal regulation by stimulating adrenaline and nor-adrenaline hormonal secretion (Srivastava and Pathak, 2012b). On the other hand, the decrease in protein levels, observed with higher doses in the present investigation may be due to their degradation and possible utilization for metabolic purposes and may attributed to the destruction or necrosis of cells and their consequent impairment in protein synthesis machinery (Srivastava and Pathak, 2012b)

# Effect of garlic treatment on total lipid, total cholesterol and triglyceride levels:

The present data in Table 3 showed that the total lipid, total cholesterol and triglyceride levels in male rabbits treated with different doses of garlic were decreased by increasing the dose of garlic, this decrease was statistically significant ( $P \le 0.05$  or  $P \le 0.01$ )with the higher doses (9 and 27 mg/kg body weight). The percentages of this decrease reached -23.4, -51.0 and -58.4% for the total lipid, -3.6, -21.7 and -40.4% for cholesterol total cholesterol and 9.5, -29.8 and -37.1% for triglyceride as compared with that of the corresponding control value, respectively.

Addition of garlic extract to oxidated vegetable oil caused a significant reduction in total cholesterol and triglyceride levels in rabbits (Fiolka *et al.*, 2004). Garlic supplementation also elicited a decrease in total cholesterol and **Table 3:** Effect of garlic treatment on serum total lipid, total cholesterol and triglyceride levels of male Bouscat rabbits.

|                 | Total lipid              |                | Total cholesterol              |                | Triglyceride         |                |
|-----------------|--------------------------|----------------|--------------------------------|----------------|----------------------|----------------|
| Treatment       | mg/dl                    | % of<br>change | mg/dl                          | % of<br>change | mg/dl                | % of<br>change |
| Control         | $195.64 \pm 19.1^{a}$    | -              | $151.65 \pm 28.6^{\mathrm{a}}$ | -              | $127.34\pm8.2^a$     | -              |
| Garlic(3mg/kg)  | $149.81\pm28.4^{b}$      | - 23.4         | $146.14 \pm 13.6^{a}$          | - 3.6          | $115.28\pm12.4^a$    | - 9.5          |
| Garlic(9mg/kg)  | $95.82 \pm 16.1^{\circ}$ | - 51.0         | $118.72 \pm 17.9^{b}$          | - 21.7         | $89.34 \pm 15.6^{b}$ | - 29.8         |
| Garlic(27mg/kg) | $81.43 \pm 15.3^{\circ}$ | - 58.4         | $90.44 \pm 12.5^{\circ}$       | - 40.4         | $80.14 \pm 17.4^{b}$ | - 37.1         |
| Significance    | **                       | -              | **                             | -              | *                    | -              |

a, b and c means with different super-script in the same column, differ significantly ( $P \le 0.05$ ). \* =  $P \le 0.05$  and \*\* =  $P \le 0.01$ .

triglyceride levels in rabbits (Suleria *et al.*, 2013a). In addition, garlic administration reduced serum total cholesterol and triglyceride levels in hypercholesterolemic rabbits (Fyiad and El-Sayed, 2012). Moreover, garlic administration decreased the levels of total cholesterol and triglyceride in diabetic rabbits (Rind *et al.*, 2013). These compounds may exert their anti-cholesterolemic effect by three different mechanisms: by inhibiting hepatic cholesterol biosynthesis (Singh and Porter, 2006); by enhancing cholesterol turnover to bile acids and its excretion through gastrointestinal tract (Srinivasan and Sambaiah, 1991); or by inhibiting cholesterol absorption from intestinal lumen (Slowing *et al.*, 2001). Moreover, other non-sulphur components of garlic, such as steroid saponins, might also be able to reduce lipids and cholesterol biosynthesis (Omojola *et al.*, 2009).

## Effect of garlic treatment on high-density lipoprotein (HDL), low-density lipoprotein (LDL) and very low-density lipoprotein (VLDL) levels:

The present study showed that garlic treatment to male rabbits elicited a slight increase in high-density lipoprotein (HDL) level, while it showed marked decreases in low-density lipoprotein (LDL) and very low-density lipoprotein (VLDL) levels (Table 4). The percentages of this increase reached 12.7, 2.0 and 8.7% for(HDL), -13.1, -31.4 and -66.4% for (LDL) and -9.3, -29.7 and - 37.1% for(VLDL) as compared with that of the corresponding control value, respectively.

The present results are consistent with the results of Fiolka *et al.* (2004) who reported adding garlic extract to oxidized rapeseed oil caused a significant increase the level of HDL in rabbit. Treatment with different doses of aqueous

garlic extracts also caused a reduction in LDL and VLDL levels and increased the level of HDL in rabbits (Suleria *et al.*, 2013a). Different studies reported that garlic and its constituents decreased LDL and increased HDL levels in **Table 4:** Effect of garlic treatment on serum high-density lipoprotein (HDL),

|                  | HDL             |                | LDL                   |                | VLDL                     |                |
|------------------|-----------------|----------------|-----------------------|----------------|--------------------------|----------------|
| Treatment        | mg/dl           | % of<br>change | mg/dl                 | % of<br>change | mg/dl                    | % of<br>change |
| Control          | $42.71\pm5.2$   | -              | $83.47 \pm 6.9^{a}$   | -              | $25.47\pm1.6^a$          | -              |
| Garlic(3 mg/kg)  | $48.15\pm3.3$   | 12.7           | $72.52\pm9.1^{ab}$    | - 13.1         | $23.1\pm4.1^a$           | - 9.3          |
| Garlic (9 mg/kg) | $43.57\pm5.0$   | 2.0            | $57.28 \pm 12.9^{b}$  | - 31.4         | $17.9\pm3.1^{\text{b}}$  | - 29.7         |
| Garlic(27 mg/kg) | $46.41 \pm 4.2$ | 8.7            | $28.02\pm5.1^{\rm c}$ | - 66.4         | $16.03\pm5.5^{\text{b}}$ | - 37.1         |
| Significance     | NS              | -              | **                    | -              | *                        | -              |

low-density lipoprotein (LDL) and very low-density lipoprotein (VLDL) levels of male Bouscat rabbits.

a, b and c means with different super-script in the same column, differ significantly ( $P \le 0.05$ ). \* =  $P \le 0.05$ , \*\* =  $P \le 0.01$  and NS = Not significant.

hyperlipidemic rabbits (Fyiad and El-Sayed, 2012). The protective effects of garlic may be attributed to inhibition of enzymes involved in lipid synthesis, prevention of lipid peroxidation and LDL, and increasing antioxidant activity (Rahman and Lowe, 2006). Dillon *et al.* (2003) reported that garlic inhibits the *in vitro* oxidation of isolated human LDL by scavenging superoxide and inhibiting the formation of lipid peroxides; and protects cellular structures against peroxidation, which act as inhibitors for some enzymes, such as hydroxyl methyl glutaryl CoA reductase (Ashraf *et al.*, 2005). Moreover, garlic appears to inhibit hepatic fatty acid synthesis by lowering key enzymes activities in supplying substrates, and to prevent lipid implantation on the arterial wall (Abramovitz *et al.*, 1999). The hypolipidaemic activities of garlic may be also attributed to other non-sulphur components, like the steroid saponins (Omojola *et al.*, 2009).

### Effect of garlic treatment on serum enzyme activities of the liver:

The activity of AST enzyme in male rabbits treated with different doses of garlic was insignificantly changed by increasing the dose of garlic (Table 5). While, the lower dose of garlic (3 mg/kg body weight) slightly decreased ( $P \le$ 0.05), ALT and ALP enzyme activities, the higher doses (9 and 27 mg/kg body weight) showed increases ( $P \le 0.05$ ) in ALT and ALP enzyme activities of male rabbits. The percentages of this change reached -5.9, 1.6 and 5.3% for AST, -12.0, 14.2 and 27.9% for ALT and -28.5, 10.9 and 40.4% for ALP in comparison with that of the control value, respectively.

Abd and Al-Baghdadi (2009) reported that administration of aqueous garlic extract restored AST and ALT enzymes activities induced by carbon tetrachloride in male rabbits. Aletan and Eteng (2013) also reported that oral

**Table 5:** Effect of garlic treatment on serum aspartate amino-transferase(AST), alanine amino-transferase (ALT) and alkaline phosphatase(ALP) enzyme activities of male Bouscat rabbit.

|                   | AST           |                | ALT                 |                | ALP                 |                |
|-------------------|---------------|----------------|---------------------|----------------|---------------------|----------------|
| Treatment         | IU/L          | % of<br>change | IU/L                | % of<br>change | IU/L                | % of<br>change |
| Control           | $79.80\pm7.4$ | -              | $28.07 \pm 3.6^{b}$ | -              | $19.30 \pm 3.4^{b}$ | -              |
| Garlic (3 mg/kg)  | $75.10\pm8.2$ | -5.9           | $24.70 \pm 4.1^{b}$ | -12.0          | $13.80 \pm 3.5^{b}$ | - 28.5         |
| Garlic (9 mg/kg)  | $81.10\pm7.2$ | 1.6            | $32.05\pm4.5^{ab}$  | 14.2           | $21.40 \pm 3.3^{b}$ | 10.9           |
| Garlic (27 mg/kg) | $84.10\pm8.8$ | 5.3            | $35.90 \pm 4.1^{a}$ | 27.9           | $27.09 \pm 3.7^{a}$ | 40.4           |
| Significance      | NS            | -              | *                   | -              | *                   | -              |

a and b means with different super-script in the same column, differ significantly ( $P \le 0.05$ ). \* =  $P \le 0.05$  and NS = Not significant.

administration of garlic extract increased AST enzyme activity by increasing the doses in rats. In addition, Ajayi and Ajayi (2014) reported that AST and ALP enzyme activities were decreased with lower dose of garlic powder, but increased with higher dose in hypercholesterolemic rats.

### Effect of garlic treatment on kidney function:

The present results (Table 6) showed that treatment of male rabbits with different doses of garlic slightly insignificantly decreased the urea level and slightly insignificantly increased creatinine level. The percentages of this increase reached-4.5, -10.9 and -0.4% for urea level and 4.9, 2.4 and 12.2% for creatinine level as compared with that of the corresponding control value, respectively. Administration of garlic extract decreased the levels of urea and creatinine in diabetic rabbits (Rind *et al.*, 2013) or normal rabbits (Suleria *et al.*, 2013b).On the other hand, Mahmoodi *et al.* (2006) found that administration of raw garlic did not change urea and creatinine levels in human. Omurtag *et al.* (2005) reported that garlic and its components provide protection against free radical damage in the body through their antioxidant activities. The antioxidative activities of garlic could also be related to its contents of cysteine-containing bioactive compounds, seleno-compounds and flavonoids (Banerjee and Maulik, 2002).

### Effect of garlic treatment on testosterone hormone level:

Testosterone levels in rabbits treated with different doses of garlic were increased by increasing the dose of garlic(Table 7). However, this increase was statistically significant ( $P \le 0.05$ ) with the doses 9 and 27 mg/kg. The

percentages of this increase reached 12.5, 27.4 and 24.7% as compared with that of the corresponding control value, respectively.

| Treatment         | U               | rea         | Creatinine     |             |  |  |
|-------------------|-----------------|-------------|----------------|-------------|--|--|
|                   | mg/dl           | % of change | mg/dl          | % of change |  |  |
| Control           | $31.20 \pm 3.9$ | -           | $0.41 \pm 0.3$ | -           |  |  |
| Garlic (3 mg/kg)  | $29.80 \pm 2.2$ | - 4.5       | $0.43 \pm 0.3$ | 4.9         |  |  |
| Garlic (9 mg/kg)  | $27.80 \pm 1.9$ | - 10.9      | $0.42 \pm 0.2$ | 2.4         |  |  |
| Garlic (27 mg/kg) | $31.08\pm0.6$   | - 0.4       | $0.46\pm0.1$   | 12.2        |  |  |
| Significance      | NS              | -           | NS             | -           |  |  |

**Table 6:** Effect of garlic treatment on serum urea and creatinine levels of male Bouscat rabbits.

NS = Not significant).

 Table 7: Effect of garlic treatment on testosterone level of male Bouscat rabbits.

| Treatment         | Testosterone              |             |  |  |  |  |
|-------------------|---------------------------|-------------|--|--|--|--|
|                   | ng/dl                     | % of change |  |  |  |  |
| Control           | $4.01\pm0.3^{b}$          | -           |  |  |  |  |
| Garlic (3 mg/kg)  | $4.51\pm0.3^{b}$          | 12.5        |  |  |  |  |
| Garlic (9 mg/kg)  | $5.11\pm0.4^{\mathrm{a}}$ | 27.4        |  |  |  |  |
| Garlic (27 mg/kg) | $5.00\pm0.4^{\rm a}$      | 24.7        |  |  |  |  |
| Significance      | *                         | -           |  |  |  |  |

a and b means with different super-script in the same column, differ significantly ( $P \le 0.05$ ). \* =  $P \le 0.05$ .

Testosterone hormone was significantly higher in garlic-fed male rabbit (El-Amary and Abou-Warda, 2007). El-Shafey *et al.* (2009) attributed the garlic-induced increase in testosterone level to the elevation of sex hormone binding globulin, which binds more testosterone, and consequently, oblige the testis to excrete more male sex hormone in plasma. Oi *et al.* (2001) suggested that garlic supplementation might enhance protein anabolism and suppress protein catabolism due to hormonal regulation by the stimulation of steroid hormones, leading to greater testis testosterone content and lower plasma corticosterone concentration. Mirfardi and Johari, (2015) suggested that garlic compounds are responsible for the significant increase in testosterone levels by affecting the performance of steroid-generating enzymes, testosterone hormone and its metabolites. They concluded that garlic supplementation likely increases testicular testosterone content due to the stimulation of LH secretion from the

pituitary gland, which stimulate the testes to increase its testosterone production.

### Effect of garlic treatment on physical semen characteristics:

The present study showed that the lower dose of garlic (3 mg/kg body weight) elicited increases in the ejaculate volume, concentration, the total output, the wave motions, the motility percentages, the percentages of live spermatozoa and the percentages of abnormal spermatozoa (Table 8). On the other hand, the higher doses (9 and 27 mg/kg body weight) showed decreases in the ejaculate volumes, the wave motions and the motility percentages, but increased concentrations and abnormal spermatozoa. These results are in good accordance with the results of El-Amary and Abou-Warda (2007) who found that quality of semen parameters was significantly higher after adding garlic to male rabbit diets. The present results are also consistent with the results of Ouarda and Abd-Ennour (2011) who found that treatment of rabbits with row garlic restored lead-induced decrease in sperm speed, motility and viability.

| Table 8: Effect of garlic treatment on | physical semen  | characteristic  | s including |
|--|-----------------|-----------------|-------------|
| ejaculated volume of semen,            | concentration a | and total outpu | it of sperm |
| of male Bouscat rabbits.               |                 |                 |             |

| Treatment            | Ejaculate<br>volume<br>(ml) | Concentration<br>(x10 <sup>6</sup> /ml) | Total output<br>( x10 <sup>6</sup> ) | Wave<br>motion<br>( Score) | Motility<br>(%) | Live sperm<br>(%) | Abnormal<br>sperm<br>(%) |
|----------------------|-----------------------------|---|--------------------------------------|----------------------------|-----------------|-------------------|--------------------------|
| Control              | $0.71\pm0.1$                | $1.99\pm0.5^{b}$                        | $1.41{\pm}0.4^{b}$                   | $3.40\pm0.3$               | $68.32 \pm 4.8$ | $93.83 \pm 1.1$   | $4.01\pm0.5^{b}$         |
| Garlic (3<br>mg/kg)  | $0.83 \pm 0.1$              | $3.55\pm0.7^{a}$                        | $2.57\pm0.5^{a}$                     | $3.90\pm0.2$               | 73.37 ± 5.3     | $94.67\pm0.6$     | $4.11\pm0.3^{b}$         |
| Garlic (9<br>mg/kg)  | $0.61 \pm 0.1$              | $3.29\pm0.4^a$                          | $2.01\pm0.3^{b}$                     | $3.35\pm0.3$               | $67.52 \pm 4.2$ | $93.50\pm1.0$     | $4.51{\pm}0.6^{b}$       |
| Garlic (27<br>mg/kg) | $0.52 \pm 0.1$              | $1.95 \pm 0.5^{b}$                      | $1.03 \pm 0.2^{b}$                   | $2.90 \pm 0.3$             | 59.17 ± 3.6     | $92.17\pm0.8$     | $5.41 \pm 0.4^{a}$       |
| Significance         | NS                          | *                                       | *                                    | NS                         | NS              | NS                | *                        |

a and b means with different super-script in the same column, differ significantly ( $P \le 0.05$ ). \* =  $P \le 0.05$  and NS = Not significant.

*Conclusively*, the present study confirms the health and medicinal benefits of garlic. The dose of garlic plays a key role in determining which effect its administration would have on the physiology and biochemistry of the body organs. While the lower doses of garlic can be safely used, the higher doses may cause problems. The lower dose of garlic improved fertility parameters of buck rabbits.

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### تأثير الثوم (Allium sativum) على العوامل الدموية والبيوكيميائية والهرمونية والخصوبة لأرانب البوسكات الذكرية

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أجريت الدراسة الحالية على قطيع من أرانب البوسكات التابعة لمحطة بحوث الجميزة ، معهد بحوث الإنتاج الحيواني ، مركز البحوث الزراعية ، وزارة الزراعة ، مصر ، ومدت الدراسة التجريبية شهرين (من 6 إلى 8 أشهر من العمر) للتحقق من تأثير علاج الثوم على عوامل الدم والكيمياء الحيوية والهرمونية والخصوبة لدى ذكور أرانب البوسكات لهذا الغرض ،وقد تم توزيع أربعة وعشرين ذكر من ذكور الأرانب البوسكات بوزن 3100-3300 جم إلى أربع مجموعات تجريبية بأعداد متساوية (ن= 6ارانب ). وقد تم استخدام المجموعة الأولى كعنصر تحكم وحقنت تحت الجلد بمحلول ملحي (1 مل من 0.9 ٪ كلوريد الصوديوم) ، المجموعة الثانية والثالثة والرابعة تم حقنها تحت الجلد مرة واحدة في الأسبوع لمدة 8 أسابيع مع الثوم 3 و و 27 مجم / كجم من الجسم الوزن ، على التوالي.

كأن العدد الإجمالي لكريّات الدم الحمراء وكرات الدم البيضاء وكذلك تركيز الهيموجلوبين في ذكور الأرانب المعالجة بجر عات مختلفة من الثوم (3 ، 9 أو 27 مجم / كجم من وزن الجسم ، مرة واحدة في الأسبوع لمدة 8 أسابيع) معنويا (P< 0.05 أو P< 0.01) بزيادة جرعة الثوم. أظهرت الجرعات المنخفضة من الثوم (3 مجم / كجم من وزن الجسم) زيادات طفيفة ( ${
m P} \leq 0.05$ ) في مستويات البروتين الكلي والألبومين وانخفاض طفيف في مستوى الجلوبيولين ، والجرعات الأعلى (9 و 27 مجم / كجم من (P  $\leq 0.05)$ الجسم) أظهر الوزن) انخفاضًا ملحوظًا (P< 0.05) في مستويات البروتين الكلي والألبومين والجلوبيولين. انخفض إجمالي مستويات الدهون والكوليسترول الكلي والدهون الثلاثية في ذكور الأرانب المعالجة بجر عات مختلفة من الثوم بزيادة جر عة الثوم ، وكان هذا الانخفاض ذو دلالة إحصائية (P< 0.05 أو P< 0.01) مع الجرعات الأعلى (9 و 27 مجم). / كجم وزن الجسم). أدت معالجة الثوم لأرانب الثوم إلى زيادة طفيفة في مستوى البروتين الدهني عالى الكثافة (HDL) ، بينما أظهرت انخفاضات ملحوظة في مستويات البروتين الدهني منخفض الكثافة (LDL) ومستويات البروتين الدهني منخفض الكثافة (VLDL). تم تغيير نشاط إنزيم AST لأرانب باك المعالجة بجر عات مختلفة من الثوم بشكل طفيف عن طريق زيادة جرعة الثوم. بينما انخفضت الجرعة المنخفضة من الثوم (3 مجم / كجم من وزن الجسم) بشكل طفيف (P< 0.05) وأنشطة إنزيم ALT و ALP ، أظهرت الجرعات الأعلى (9 و 27 مجم / كجم من وزن الجسم) زيادات ( P< 0.05) في أنشطة إنزيم ALT و ALP للأرانب الذكور. أدى علاج الأرانب الباكية بجرعات مختلفة من الثوم إلى انخفاض طفيف في مستوى اليوريا وزيادة طفيفة في مستوى الكرياتينين. تمت زيادة مستويات هرمون التستوستيرون في الأرانب المعالجة بجر عات مختلفة من الثوم بزيادة جرعة الثوم ، لكن هذه الزيادة كانت ذات دلالة إحصائية (P<0.05) بجرعات 9 و 27 مجم / كجم. أدت الجرعات المنخفضة من الثوم (3 مجم / كجم من وزن الجسم) إلى زيادة حجم السائل المنوى ، التركيز ، الناتج الإجمالي ، حركات الموجة ، نسب الحركة ، نسب الحيوانات المنوية الحية ونسب الحيوانات المنوية غير الطبيعية. ومع ذلك ، أظهرت الجرعات الأعلى (9 و 27 ملجم / كجم من وزن الجسم) انخفاضًا في أحجام السائل المنوي ، وحركات الموجات ونسب الحركة ، ولكن زيادة التركيزات وشذوذ الحيوانات المنوية. التوصية: من النتائج الحالية يمكن استنتاج أن الجرعات المنخفضة من الثوم يمكن استخدامها بأمان ومع ذلك ، الجرعات العالية قد تسبب مشاكل. أدت الجرعة المنخفضة من الثوم إلى تحسين معايير الخصوبة لذكور الأرانب.