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Effect of vitamin E or propolis supplementation on semen quality, oxidative status and hemato-biochemical changes of rabbit bucks during hot season

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ABSTRACT

This study was designed to determine the effects of supplementation of vitamin E or propolis to bucks' diets on semen quality, oxidative status and hemato-biochemical changes during summer season. Thirty adult male New Zealand white rabbits were randomly allocated into three equal groups, 10 bucks each. Bucks received a maintenance diet supplemented by vitamin E (150 mg/kg diet, E-group) or propolis (150 mg/kg diet, P-group) or not supplemented (control, C-group) for ten consecutive weeks. Semen and blood samples were collected to analyze semen quality traits and hemato-biochemical parameters, respectively. Ambient temperature and relative humidity were recorded daily and used to estimate the temperature humidity index (THI). Results indicated that bucks in the E- and P-groups had better libido (lower reaction time, $P<0.05$) and higher sperm concentration and viability than the control group ($P<0.01$). These enhancements were parallel to an increase in blood plasma testosterone concentrations in the vitamin E and propolis-treated groups ($P<0.01$). Additionally, concentrations of seminal plasma total protein and initial fructose were significantly increased in the semen of both treated-groups. For blood components, either vitamin E or propolis supplementation enhanced hematopoiesis including the number of red blood cells, hematocrite value and hemoglobin concentration ($P<0.01$). Also, concentrations of blood plasma glucose and high density lipoprotein (HDL) were increased, while those of blood plasma cholesterol and triglycerides were decreased ($P<0.01$) in both treated-groups. Supplementation with vitamin E or propolis enhanced ($P<0.01$) oxidative status of the blood plasma, whereas propolis-treated bucks had the highest value of total antioxidant capacity followed by vitamin E treated-bucks. Also, malondialdehyde activity was lower ($P<0.05$) in the propolis-treated bucks compared with the control, while bucks of the E-group were intermediate. In summary, inclusion of vitamin E or propolis in male rabbits' diets during the hot season could be used effectively to mitigate negative impacts of elevated temperature on semen quality, oxidative status and hemato-biochemical features.

1. Introduction

Under subtropical conditions, ambient temperature is the major constraining factor controlling animal productivity. In Egypt, the summer and early autumn seasons (from May to September) are not within the thermal neutral zone (Attia et al., 2011). The comfort zone temperature is around 21 °C and their productive and reproductive performances could be impaired, which in turn prevents continuation of the breeding season for about 5 months (García-Tomás et al., 2008). Under such environmental conditions, several physiological and reproductive disorders are caused by

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disturbances in blood metabolites, oxidative status, enzymatic reactions and hormonal secretions (Alvariño, 2000; Marai et al., 2002). Rising free radicals, reactive oxygen species (ROS), seems to be one of the most harmful effects of elevated temperature that initiates peroxidation of polyunsaturated fatty acids (PUFA). These are important for sperm plasma membrane integrity and subsequent fertility (Altan et al., 2003; Sahin et al., 2001). In the testes, low concentrations of ROS are normally produced during testicular spermatogenesis and steroidogenesis, and they are necessary for different sperm functions such as capacitation, hyperactivation, acrosome integrity and sperm–oocyte fusion (Mathur and D'Cruz, 2011). Overproduction of ROS can impair sperm integrity causing male infertility (Paul et al., 2009). Since cooling of the housing buildings is expensive, a number of alternative methods are used to modulate the harmful effects of high environmental temperature; focusing mostly on dietary manipulation. Vitamin E is believed to be the primary component of the antioxidant system of the spermatozoa, and is one of the major membrane protectors against ROS and lipid peroxidation (Surai et al., 1998). Additionally, vitamin E has been shown to increase total sperm output and sperm concentration in male rabbits (Castellini et al., 2003).

There is an international interest concerning application of natural medical sources in animal production fields. Propolis is a natural resinous substance collected by bees from parts of plants, buds and exudates (Zhou et al., 2008). About 300 components, mainly phenolic compounds, have been identified. Most of these isolated compounds belong to three main groups, flavonoids, phenolic acids and esters (Simoes et al., 2004). Propolis has several therapeutic properties including antioxidant, antimicrobial, anti-parasitic, antiviral, anti-inflammatory and anti-tumoral properties (Kumazawa et al., 2004; Paulino et al., 2008). Therefore, this study was designed to test the efficiency of propolis comparable to vitamin E in mitigating heat stress effects on semen characteristics, oxidative status and hemato-biochemical profile in adult male rabbits.

2. Materials and methods

The present study was conducted in the Agricultural Experimental Station (31° 20′N, 30°E), Faculty of Agriculture, Alexandria University, Alexandria, Egypt. The procedures imposed on the animals were carried out meeting the Directive 2010/63/EU of the European Parliament and of the Council of 22 September 2010 on the protection of animals used for scientific purposes.

2.1. Animals and their management

Thirty, 12 month old, male, New Zealand White rabbits with a mean body weight of 3.35 ± 0.06 kg were used in the present study. Bucks were individually kept in wire cages ($40 \times 50 \times 35$ cm^3) in a naturally ventilated house. A period of 16 h of light was provided which included 12 h of natural day light and 4 h of supplementary electric light. Clean fresh water was available for rabbits at all times. Bucks were randomly allocated into three equal groups: (1) control (C-group), bucks received the maintenance diet; (2) vitamin E (E-group), bucks received the maintenance diet supplemented with 150 mg of vitamin E/kg diet; and (3) propolis (P-group), bucks received the maintenance diet supplemented with 150 mg of propolis/kg diet. Both additives were mixed with diet components during manufacturing of the pellets. Treatments were applied for 10 weeks. Bucks were individually fed 200 g/d of the maintenance diet (18% barley, 25% wheat bran, 6% yellow corn, 18% soy bean, 28% alfalfa hay, 3% molasses, 1% $CaCo_3$, 1% NaCl; 17.8% CP and 2650 K cal/Kg DE) (NRC, 1994). Vitamin E

Table 1
Chemical composition of propolis detected by gas chromatography/mass spectrometry.

Compounds	Retention time (min)	Area of each component (%)
Trimethylsilyl 3-Phenyl-2-Propenoate	17.72	3.71
Dodecanoic Acid, Trimethylsilyl Ester	20.11	0.69
6,7-Dihydroxycoumarin Di-Tms	21.95	0.51
Tetradecanoic Acid, Trimethylsilyl Ester	24.28	3.58
Hexadecanoic Acid, Ethyl Ester	27.09	1.78
Cinnamic Acid, 3,4-Dimethoxy-, Trimethylsilyl Ester	27.90	2.56
Hexadecanoic Acid, Trimethylsilyl Ester	28.11	24.42
Ferulic Acid	28.84	2.68
Silane,[1-(5-Ethenyltetrahydro-5-Methyl-2-Furanyl)-1-Methylethoxy]Trimethyl-, Cis	29.16	1.06
Octadecanoic Acid, Trimethylsilyl Ester	31.59	2.60
3-Methyl-3-Butenyl Isoferulate-Tms-Derivative	32.66	2.96
2-Propen-1-One, 1-(2,6-Dihydroxy-4-Methoxyphenyl)-3-Phenyl-, (E)	34.44	0.84
1,2,4-Tris(Tert Butyldimethylsiloxy)Naphthalene	35.36	1.57
1h-Imidazole-4-Carboxamide, 5-Amino-, Tetrakis(Trimethylsilyl) Deriv.	35.91	1.94
3-(Hydroxymethyl)-1-Phenyl-1-Heptadecyn-3-Ol	36.22	13.75
2′,4′,6′-Tris(Trimethylsilyloxy)Chalcone	36.33	1.73
4-Quinolinamine, N,3-Diphenyl-2-(Phenylmethyl)	36.63	11.03
Bis-O-Trimethylsilyl-Palmitinic Acid-Glycerin-(1)-Monoester	37.23	2.65
Benzoic Acid, 2-[(Trimethylsilyl)Amino]-3-[(Trimethylsilyl)Oxy]-, Methyl Ester	37.39	0.96
1h-Pyrrole, 5-(3-Methoxyphenyl)-2,3-Diphenyl	39.15	1.61
2-Methyl-1,6- Bis[(Trimethylsilyl)Oxy] Anthra-9,10-Quinone	39.58	5.95
3,5,7-Tris(Trimethylsilyloxy)Flavone	39.74	10.15
Meso-Dimethyl-R-3,C-6-Bis[(TertButyldimethylsilyl)Oxy]–T-4,5-Epoxycyclohex-1-Ene-1,2-Dicarboxylate	41.30	1.27