

**INFLUENCE OF GRAPE SEEDS POWDER AS A NATURAL ANTIOXIDANT ON GROWTH PERFORMANCE, ANTIOXIDANT STATUS AND CARCASS CHARACTERISTICS OF RABBITS UNDER HOT CONDITIONS.**

**Fawzia A. Hassan<sup>1</sup>; Kh. M. Mahrose<sup>2</sup> and M.M. Basyony<sup>3</sup>**

1- Department of By-products Utilization, Animal Production Research Institute, Agricultural Research Center, Dokki, Giza, Egypt. E-mail address: fawzia\_amer@yahoo.com

2- Department of Poultry, Faculty of Agriculture, Zagazig University, Zagazig, Egypt.

3- Department of Poultry Nutrition, Animal Production Research Institute, Agricultural Research Center, Dokki, Giza, Egypt.

**ABSTRACT**

*The present study aimed to investigate the effects of different levels of dietary supplementation of grape seeds powder on growth performance, antioxidant status and carcass characteristics of rabbits under high ambient temperature of Borg-El Arab, Alexandria Governorate. A total number of forty eight weaned New Zealand White (NZW) male rabbits at 6 weeks of age were randomly assigned to four experimental groups in a complete randomized design. The 1<sup>st</sup> group was served as a control group. The 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> groups were received daily pelleted diet supplemented with 0.5, 1.0 and 1.5% grape seeds powder (GSP), respectively. **Results obtained showed that** rabbits received 1.0% of GSP had the heaviest ( $P<0.05$ ) final body weight (2435.83 g) and average daily weight gain (29.88 g/rabbit/day). While rabbits received 1.5% of GSP consumed less ( $P<0.05$ ) feed (87.99 g/rabbit/day) and showed the best ( $P<0.05$ ) value of feed conversion (3.27 g feed/g gain) when compared to the other groups. The highest ( $P<0.05$ ) carcass weights and kidney fat percentage were recorded with rabbits group received 1.0% GSP. Plasma total protein and globulin values were significantly ( $P<0.05$ ) increased with the increase of GSP level used in the diet, while plasma total lipids and ALT values were significantly ( $P<0.05$ ) lower in rabbits group received dietary 1.5% GSP than the other rabbit groups. Total antioxidant capacity (T-AOC), superoxide dismutase, and glutathione peroxidase were gradually ( $P<0.05$ ) elevated with increasing GSP level in the diet. In addition, there was an improvement of the economical efficiency of diets contained 0.5, 1, 1.5% GSP compared to the control one and the best economical efficiency and relative economical efficiency values were recorded with rabbits group fed 1.5% GSP.*

**Conclusively**, it could be concluded that dietary supplementation of grape seeds powder at 0.5, 1, 1.5% levels improved growth performance and increased the resistance of rabbits against oxidative stress by modulation of endogenous antioxidant enzymes under hot conditions, Moreover, 1.5% GSP dietary supplementation was more effective than other levels.

**Key words:** Grape seed powder, Growth performance, Antioxidant status, Carcass characteristics, Rabbits, Hot conditions.

## INTRODUCTION

There are many factors affecting the intensive rabbit production such as environmental and nutritional conditions. The environmental conditions represent an important element in the production cycle. The domestic rabbit has a high metabolic rate, undeveloped sweat glands and slow heat loss. The high temperature in hot climate conditions affects negatively growth, reproductive performance, feed intake and blood constituents (Marai *et al.*, 2000 ; Marai *et al.*, 2006 and Abd El-Monem *et al.*, 2009). Heat stress causes oxidative stress, reflected by increased reactive oxygen species production. In addition, oxidative stress impairs the cell membrane and mitochondrial integrity and causes cell damage through lipid peroxidation, which can be minimized by supplementation of antioxidant (Halliwell and Gutteridge, 1989). The imbalance between reactive oxygen species (ROS) and antioxidant capacity of the organism leads to a condition of oxidative stress (Urso and Clarkson, 2003).

Antioxidants, including vitamins, carotenoids and tannins provide protection against oxidative damage, and increase attention as a potential chemo-preventive agent. Grape seeds are rich in antioxidant compounds, including phenolic compound (predominantly tannins), and it has been demonstrated that these compounds reduce the risk of oxidative stress by protecting against free radical mediated damage (Gorinstein *et al.*, 1994).

Plant-derived antioxidants can greatly reduce the damage due to oxidants by neutralizing the free radicals before they can attack the cells and thereby prevent damage to lipids, proteins, enzymes, carbohydrates and DNA (Satyam *et al.*, 2013). Research for new bio-efficient antioxidants has particularly focused on natural antioxidants to respect consumer concerns over safety and toxicity. Plant extracts rich in polyphenols are good candidates, because they are easily obtained from natural sources and they efficiently prevent lipid oxidation (Brenes *et al.*, 2008 and Pozuelo *et al.*, 2012).

Grape is a widely spread fruit crop in Egypt, it is considered to be the second most important fruit crop after citrus fruit. Grapes are grown from Aswan in the South to Alexandria in the North of Egypt. The majority of grapes in Egypt originate from the species (*Vitis Vinifera L.*) (Bayer Crop Science Egypt, 2012). Currently, there are 66.262 Ha Grapes planted in Egypt that produce 1.37.815 tonnes (FAOSTAT, 2012).

Grape seeds powder is a natural agricultural by-product that has a high concentration of vitamin E, flavonoids, linoleic acid, and oligomeric proanthocyanidins, so it considers a better source of antioxidative constituents than skins of grape juice by-products (Hassan *et al.*, 2014). Grape seeds are rich source of polyphenols such as phenolic acid, anthocyanins, and flavonoids including monomeric phenolic compounds, such as (+) -catechins, (-) -epicatechin, and (-) -epicatechin-3-O-flattened dimeric, trimeric, and turmeric procyanidins (Monagas *et al.*, 2005).

Although polyphenolic compounds may improve animal health, they can also decrease proteolytic activity and thus compromise protein digestion (Oliveira *et al.*, 2010). Studies have shown that the flavonoids had the capacity to act as powerful antioxidants by scavenging free radicals and terminating oxidative reactions (González-Paramás *et al.*, 2004; Yilmaz and Toledo, 2004; Ruberto *et al.*, 2007; Brenes *et al.*, 2008; Dorri *et al.*, 2012).

Therefore, the aim of this study was to evaluate the effect of dietary grape seeds powder as a natural antioxidant on the growth performance, antioxidant status and carcass characteristics of rabbits under high ambient temperature of Borg-El Arab, Alexandria Governorate, Egypt.

## **MATERIALS AND METHODS**

The experimental work of this study was carried out at Borg-El Arab, Alexandria Governorate, Experimental Station of Animal Production, Animal Production Research Institute, Agricultural Research Center, Ministry of Agriculture, Egypt.

### ***Experimental Design and Application***

A total number of Forty eight of weaned New Zealand White (NZW) males rabbits at 6 weeks of age and nearly equal average initial live body weight ( $757.21 \pm 29.74$  g) were randomly assigned to four experimental treatment groups (n=12 in each) in a complete simple randomized design, 4 replicates (3 rabbits for each). The first experimental group received daily untreated pelleted diet (control). The second, third and fourth experimental groups received daily pelleted diet supplemented with 0.5, 1.0 and 1.5% of grape seeds powder (GSP), respectively. The experimental period lasted for 8 weeks. Grape pomace was obtained from El-Ahram Henken for beverages (Ganaklise Company) at Ganaklise, El-Behera Governorate. The pomace was obtained in a wet condition with moisture content from 65-70%. The pomace contains grape seeds and grape skin and stalks. The humidity of grape pomace was reduced by sun-drying to 9-10%, and then the seeds were separated from pomace and ground by hammer mill and kept for subsequent processing.

Chemical analysis of red grape (*Vitis vinifera* L.) seeds powder and experimental diets were detected according (AOAC, 2007) as shown in Table 2. The Chemical analysis and the fraction of phenolic and flavonoids compounds of red grape seeds powder were presented in Table (2 and 3). Flavonids and phenolics were determined in Micro analysis Lab., Food Technology Research Institute by A high-performance liquid chromatographic (HPLC). Phenolic compounds were determined according to the method of Goupy *et al.*, (1999). Flavonid compounds were determined according to the method of Mattlia *et al.*, (2000) as shown in Table 3.

### ***Animal and diets***

Rabbits were individually housed in galvanized wire cages (Dimensions of 60×40× 35 cm) until marketing at 14 weeks of age under a 12:12 h light–dark cycle. All rabbits were fed pelletized feed *ad libitum*, fresh water was automatically available all the time by stainless steel nipples fixed in each cage. Feed ingredients and chemical composition of experimental diets (%DM basis) are shown in Table 1. The experimental diets were formulated to meet the recommended nutrient requirements of growing rabbits according to Lebas (2004).

**Table 1:** Feed ingredients and chemical composition of experimental diets (% DM basis).

Feed Ingredients (%)	Control	Experimental diets (%)		
		GSP (0.5)	GSP (1.0)	GSP (1.5)
Soybean meal (44%CP)	20.9	20.4	20.4	20.4
Barley	32.0	32.0	32.0	32.0
Wheat bran	9.20	9.20	9.20	9.20
Clover hay	31.0	31.0	30.5	30.0
Molasses	3.00	3.00	3.00	3.00
Limestone	0.70	0.70	0.70	0.70
Di- Ca- phosphate	2.20	2.20	2.20	2.20
DL-Methionine	0.40	0.40	0.40	0.40
NaCl	0.30	0.30	0.30	0.30
Vit.-Min. premix*	0.30	0.30	0.30	0.30
Grape Seeds powder	0.00	0.50	1.00	1.50
<b>Total</b>	<b>100</b>	<b>100</b>	<b>100</b>	<b>100</b>
Total price L.E/100 Kg	230.0	228.0	227.5	227.0
<b>Chemical composition (%DM basis)</b>				
DM	87.88	88.3	87.68	86.98
OM	90.88	90.77	90.68	90.59
CP	17.56	17.38	17.37	17.35
CF	13.26	13.41	13.45	13.50
EE	1.980	2.030	2.360	2.420
NFE	58.08	57.95	57.50	57.32
Ash	9.120	9.230	9.320	9.410
Methionine	0.670	0.670	0.670	0.670
Methionine+cysteine	0.760	0.750	0.750	0.760
Lysine	0.980	0.970	0.970	0.960
Calcium	1.290	1.300	1.310	1.320
Available Phosphours	0.510	0.510	0.520	0.520
Digestible energy (Kcal/Kg DM)	2790	2771	2761	2751

\*Mineral and vitamin mixture supplied per kg of diet: Vitamin A 10,000 IU, Vitamin D3, 1,800 UI; Vitamin E, 15 mg; vitamin K3, 4.5 mg; Vitamin B1, 0.5 mg; Vitamin B2, 4 mg; Vitamin B12, 0.001 mg; Folic acid, 0.1 mg; Pantothenic acid, 7 mg; Nicotinic acid, 20 mg; I, 1 mg; Mn, 60 mg; Cu, 5.5 mg, Zn, 75 mg; Fe, 40 mg; Co, 0.3 mg; Se, 0.08 mg; Robenidine, 52.8 mg.

All rabbits were kept under the same management, hygienic and environmental conditions. The rabbits were reared in a well-ventilated building. Rabbitry minimum and maximum temperatures, relative humidity and temperature humidity index (THI) during the experimental period (June to July, 2013) were

ranged between 26.5-32.5°C, 62-75% and 87.5-93.5, respectively, which means that the whole experimental periods lay in the severe heat stress as described by Lphsi (1990). Live Body weight was determined weekly throughout the experimental period, and weight gain was calculated. Feed consumption was determined precisely and calculated as grams per rabbit per day (during the all experimental period). Unused feed from each cage was collected daily, weighed and taken into consideration for the calculation of feed consumption, accordingly, feed conversion was also calculated (g feed / g gain).

#### ***Slaughtering and carcass characteristics***

At the end of the experimental period (14 weeks old), five male rabbits from each group were randomly taken, fasted for 12 hours, individually weighed and immediately slaughtered. Slaughter procedure and carcass analysis were carried out as described by Blasco and Ouhayoun (1996). After complete bleeding, pelt, viscera's and tail were removed then the carcass and its components were weighed as edible parts. The non edible parts including Lung, Spleen, Stomach, Large intestine, Small intestine and kidney fat were also weighted as percentage of pre-slaughter weight. Dressing percentage was calculated by dividing the hot dressed carcass weight by pre-slaughter weight and expressed as a percentage according to Steven *et al.*, (1981).

#### ***Blood Samples and determination of biochemical parameters***

Blood samples (5 ml from each rabbit) were collected during slaughtering to determine blood components and centrifuged at 3000 r.p.m. for 15 minutes to separate blood plasma. Blood plasma total protein, albumin, glucose, urea-N, total cholesterol, LDL, HDL-cholesterol, vLDL, triglycerides, total lipids, creatinine and ALT were colorimetrically determined using commercial kits (purchased from Biodiagnostic, Egypt) according to the manufacturers' instructions. Plasma globulin concentration was calculated by difference and Albumin/Globulin ratio was calculated.

Blood antioxidant constituents were assayed by colorimetric technique using commercial kits (Biodiagnostic, Egypt). Blood plasma malondialdehyde (MDA) was determined according to Ohkawa *et al.* (1979) and glutathione peroxidase (GPx) activity assayed using the method of Chiu *et al.*, (1976). Superoxide dismutase (SOD) activity was assayed according to Misra and Fridovich (1972). Total antioxidant capacity (T-AOC) was determined according to Koracevic *et al.* (2001).

#### ***Economical efficiency***

To determine the economical efficiency of the experimental diets for body weight gain, the costs of feed required for producing one kg of body weight gain was calculated. The cost of the experimental diets was calculated according to the price of different ingredients prevailing at local market as well as the price of tested materials at the time of experimentation. Economical efficiency was calculated as a ratio between the return of weight gain and the cost of consumed feed.

#### ***Statistical Analysis***

The obtained data were statistically analyzed using the general linear model procedure of SAS® Software Statistical Analysis (SAS 1998). Differences among means were tested by Duncan's multiple range test (Duncan, 1955). All results were analyzed using this model:  $Y_{ij} = \mu + T_i + E_{ij}$ ; where:  $Y_{ij}$  = the observation of  $ij$ ;  $\mu$  = the overall mean;  $T_i$  = the effects of  $i$  (treatments) and  $E_{ij}$  = the experimental random error.

## RESULTS AND DISCUSSION

### *Chemical analysis, phenolic and flavonoids compounds of red grape seeds powder:*

The chemical composition of the GSP was examined and presented in Table 2. Crude protein (9.50%) was similar to the value reported by Basalan *et al.*, (2011) who found that CP was 93.3 g/kg DM. However, EE (4.87%) was lower than the value also reported by Basalan *et al.*, (2011) being (62.6 g/kg DM). In addition, GSP had a higher CF content (37.71%). These differences may be due to harvest methods and grape varieties.

Natural antioxidants include phenolic compounds which may act to confer an effective defense system against free radical attack. The amount of total phenols was 3.80% and total Flavonoids was 0.87%. In this connection, Gladine *et al.*, (2007) stated that the polyphenol content of grape seed was 651 GAE mg/g DM. Moreover, Li *et al.*, (2008) reported that total phenolic content was 2.53 GAE g/100 g powder).

**Table 2.** Chemical analysis of red grape (*Vitis vinifera* L.) seeds powder (on DM basis).

Items (%)	DM	OM	CP	CF	EE	NFE	Ash	Total phenols (%)	Total Flavonoids (%)
<b>Grape seeds powder (GSP)</b>	88.74	91.00	9.50	37.71	4.87	38.92	9.00	3.80	0.87

Analyzed according to AOAC (2007).

The extractable polyphenols, flavonoids and concentration of individual phenolic compounds identified by HPLC in GSP are reported in Table 3. A multitude of flavonoids are contained in GSE. The most abundant of these are the proanthocyanidins, which are oligomers of monomeric flavan-3-ol units linked by carbon-carbon bonds. The major flavan-3-ols identified in GSE are catechin, epicatechin and epicatechin-3-O-gallate (Santos-Buelga *et al.*, 1995). Gallic acid, catechin and procyanidins contents were 2.58, 5.35 and 1.70 mg/100g. Among the identified polyphenols was catechin. Procyanidins constitute the major class of phenolic compounds in grape seeds. Nakamura *et al.*, (2003) determined the concentration of gallic acid, catechin, epicatechin and Procyanidin (B1, B2, C) of the grape seed as 2.06, 1.03, 0.61, 0.70, 1.66, and 0.4 % (w/w), respectively. Also, Li *et al.*, (2008) found that Gallic acid was 14.2 mg/100 g powder, 162 mg/100g powder as Catechin and 121 mg/100g as Epicatechin. In addition, Rockenbach *et*

*al.*, (2011) found that there was a greater concentration of phenolic compounds in the seeds (2128 to 16,518 mg of catechin equivalents (CE)/100 g) than in the skins (660 to 1839 mg CE/100 g). These results showed that GSP was a good source of polyphenols and flavonoids as they have considerable antioxidant activity.

**Table 3.** Phenolic and Flavonoids compounds of red grape (*Vitis vinifera* L.) seed powder.

Items	Phenolic compounds mg/100g <sup>(1)</sup>	Items	Flavonoids compounds mg/100g <sup>(2)</sup>
Gallic acid	2.58	Rutin	141.11
Protocatechuic	25.28	Rosmarinic	196.10
Pyrogallol	53.95	Quercitrin	553.53
Chlorogenic	1.21	Quercetin	679.50
Catchol	41.57	Narenginin	921.55
Procyanidin	1.70	Hesperitin	131.17
Vanillic	2.89	Kampferol	9.73
Catechin	5.35	Apignin	44.89
Cinnamic	1.85		
Salicylic	6.26		
Syringic	4.84		
Chrysin	30.68		
Ferulic	29.34		

(1) Calculated according to Goupy *et al.*, (1999).

(2) Calculated according to Mattlia *et al.*, (2000).

### ***Growth performance and Economical efficiency***

Results of growth performance are illustrated in Table 4. The obtained results indicated that rabbits reared under stress summer conditions and received 1.0% of GSP had the heaviest ( $P < 0.05$ ) final body weight (2435.83 g) and average daily weight gain (29.88 g/rabbit/day) followed by those received 0.5, 1.5% and control groups. However, insignificant differences were observed among rabbits fed diets supplemented with 0.5 and 1.5 % of GSP.

The results of the present study clearly indicated that dietary supplementation of different levels of GSP had a positive effect on growth performance of growing rabbits. The improvement in body weight gain was due to drastic decreasing in rabbit daily feed intake and improvements in feed conversion ratio compared with the control group, and may be due to the biological function of GSP which have the monomers catechin and epicatechin (monomeric flavanols) representing the major phenolic compounds in grape seeds that show antioxidant activity as shown in Table 3. For example, (+)-catechin shows antioxidant activity by inhibiting the oxidation of plasma lipids (Yilmaz and Toledo, 2004). Moreover, (-)-epicatechin is able to scavenge hydroxyl radicals, peroxy radicals, superoxide radicals (Yilmaz and Toledo, 2004). Procyanidins are reported to have potent antioxidant activity both *in vitro* and *in vivo* (Simonetti *et al.*, 2002). In this study, (+)-catechin, (-)-epicatechin and procyanidins were also found in GSP which are

consistent with other studies (Oszmianski and Sapis, 1989; Escribano-Bailon *et al.*, 1992 and Fuleki and Ricardo da Silva, 1997). It is possible that these active principles in GSP may increase the activity of antioxidant enzymes, which act as antimicrobial and cause sterilization of gastrointestinal tract (Abdel-Azeem, 2005) or to tannin effect by reducing intestine movement which may be led to better absorption of nutrients, that reflected on body weight gain (Ismail *et al.*, 2003). The present results are in line with those of body weight and daily body weight gain reported by Goni *et al.*, (2007) and Brenes *et al.*, (2008) on broiler chickens. The possible reasons are that natural antioxidants can protect the intestinal mucosa against oxidative damage and pathogens and limit peristaltic activity in digestive disorders preventing diarrhea (Kermauner and Laurenčić, 2008).

**Table (4).** Means of growth performance and economical efficiency of growing rabbits fed experimental diets containing different levels of Grape seed powder (GSP).

Items	Experimental diets				Sig.
	Control	0.5%GSP	1.0%GSP	1.5%GSP	
<b>Growth performance</b>					
Initial body weight (g/rabbit)	760.50 ±38.28	761.67 ±36.15	762.50 ±22.76	744.17 ±21.75	NS
Final body weight (g/rabbit)	2236.25 <sup>b</sup> ±38.34	2374.17 <sup>ab</sup> ±57.20	2435.83 <sup>a</sup> ±70.42	2281.25 <sup>ab</sup> ±56.09	*
Average daily weight gain (g/rabbit/day)	26.35 <sup>b</sup> ±0.73	28.80 <sup>ab</sup> ±0.80	29.88 <sup>a</sup> ±0.92	27.45 <sup>ab</sup> ±1.15	*
Average daily feed intake (g/rabbit/day)	97.37 <sup>a</sup> ±0.62	95.56 <sup>a</sup> ±1.04	98.39 <sup>a</sup> ±1.34	87.99 <sup>b</sup> ±0.59	**
Feed conversion (g feed/g gain)	3.73 <sup>a</sup> ±0.11	3.34 <sup>b</sup> ±0.90	3.33 <sup>b</sup> ±0.11	3.27 <sup>b</sup> ±0.14	*
<b>Economical efficiency</b>					
Average total weight gain/ rabbit, (kg)	1.475	1.612	1.673	1.537	
Total revenue /rabbit, (LE) <sup>(1)</sup>	33.93	37.07	38.50	35.35	
Total feed intake/rabbit, (Kg)	5.45	5.35	5.51	4.93	
Price of feeding/kg, (LE)	2.30	2.28	2.28	2.27	
Total cost of feed/rabbit (LE)	12.54	12.20	12.56	11.19	
Net revenue/rabbit (LE) <sup>(2)</sup>	21.39	24.87	25.94	24.16	
Economical efficiency <sup>(3)</sup>	1.71	2.04	2.07	2.16	
Relative economical efficiency	100.00	119.30	121.05	126.32	

**a,b**, Mean values with the same letter within the same row did not differ significantly ( $P>0.05$ ).

(1) Price of one Kg/ live body weight on selling was 23 LE.

(2) Net revenue = Total revenue (LE) – Total feed cost (LE).

(3) Economic efficiency = Net revenue / Total feed cost (LE).



Rabbits group fed 1.5% GSP diet had least ( $P<0.05$ ) feed intake (87.99 g/rabbit/day) compared to the other tested groups. On the other hand, adding GSE to rabbit diets at each tested level improved ( $P<0.05$ ) the FCR compared to the control group. This improvement may be due to the antioxidative effect of GSE. In this respect, those antioxidants improved feed conversion ratio of heat stressed broilers (Vikili *et al.*, 2010).

It could be suggested that dietary supplementation of (GSP) may be useful to improve health and overall growth performance in growing rabbits. The present results also confirm that polyphenols present in GSP were absorbed at sufficient levels to contribute and modulate the antioxidant activity for rabbits.

The effects of dietary supplementation of GSP on economical efficiency are shown in Table 4. The results showed that the best economical efficiency and relative economic efficiency were recorded by rabbits group fed (1.5% GSP). These results indicated that the total feeding cost reduced by 1.54% by increasing the level of GSP when summer caused stress in rabbits. Grape seed powder at 1.5% of rabbit's diet proved to be more economical than the other treatments included control group. While the rabbit fed 1% GPS achieved the best net revenue followed by 0.5% and 1.5% while the lowest one was the control group.

It could be concluded that there was an improvement of the economical efficiency of diets contained 0.5, 1, 1.5% GSP compared with the control one, due to the improvement of performance of rabbits. Although feed intake and growth performance are reduced through high temperature conditions, the addition of grape seed powder as a natural antioxidant under conditions of oxidative stress where free radical production dramatic increases led to prevent damage to major organs and systems and economically justified, also enhance the antioxidant capacity by optimizing the dietary intake of antioxidants (Ayyat *et al.*, 2004, Abdel-Khalek, 2010, Dogan and Celik, 2012).

### ***Carcass traits***

Results of carcass traits are shown in Table 5, it could be noticed that GSP supplementation in the diet did not significantly affect all of carcass traits studied, except for carcass weights, rabbits group fed 1.0% GSE recorded higher carcass weight than that of 1.5% GSE (1557.25 vs. 1387.0 g) higher carcass weights could be attributed to higher live body weight of rabbits groups in the present study. In this connection, Cavani *et al.*, (1988) claimed that the grape seed meal influenced neither the dressing percentage nor the carcass composition.

**Table 5.** Carcass characteristics of rabbit groups fed the experimental diets.

Items	Experimental diets				Sig.
	Control	0.5%GSP	1.0%GSP	1.5%GSP	
Pre-slaughter weight (g)	2214.00 <sup>ab</sup> ±34.22	2320.00 <sup>ab</sup> ±82.05	2407.00 <sup>a</sup> ±108.97	2200.00 <sup>b</sup> ±35.04	*
Carcass weight (g)	1426.36 <sup>ab</sup> ±30.15	1505.62 <sup>ab</sup> ±63.55	1557.25 <sup>a</sup> ±96.96	1387.00 <sup>b</sup> ±30.21	*
Dressing %	64.41 ±0.61	64.84 ±0.51	64.48 ±1.29	63.05 ±0.42	NS
Edible Giblets % <sup>(1)</sup>	3.74 ±0.07	3.94 ±0.08	3.84 ±0.06	3.96 ±0.07	NS
Total edible parts % <sup>(2)</sup>	68.15 ±0.53	68.77 ±0.48	68.32 ±1.29	67.01 ±0.46	NS
Total Non edible parts %	31.85± 0.55	31.22± 0.46	31.68± 1.21	32.99± 0.46	NS

a, b, Mean values with the same letter within the same row did not differ significantly (P>0.05).

<sup>(1)</sup> Edible Giblets %= (liver+ kidney + heart) / Pre-slaughter weight (g)\*100

<sup>(2)</sup> Total edible parts %= (carcass wt. + edible giblets wt.) / Pre-slaughter weight (g)\*100.

### **Blood plasma biochemical values**

The effects of dietary supplementation of GSP on blood plasma biochemical components are showed in Table 6. Total plasma protein has been reported as an indication of the protein retained in the animal body (Akinola and Abiola, 1991; Esonu *et al.*, 2001). The obtained results revealed that the plasma total protein and globulin of the rabbits group fed 1.5% GSP diet was significantly (P<0.05) higher than those fed 0.5 and 1.0% GSP diets. However, albumin /globulin ratio had the lowest value of 0.95 in rabbits fed diet supplemented with 1.5% GSP compared to the other tested supplemented diets included the control group. The results herein also revealed insignificant differences among experimental groups fed diets containing 0.5, 1.0 and 1.5% GSP and the control group. It is known that globulins are carrier proteins for steroid and thyroid hormones and play a vital role in natural and acquired immunity to infection (Ganong, 2005). The observed increase in globulins could be attributed to the presence of an infection or due to individual differences in the rabbits fed higher levels of GSP diet (1% and 1.5%). However, the present results concerning blood parameters were found within the normal range for the healthy rabbits. However, plasma glucose levels of rabbits fed the GSP levels (0.5, 1.0 and 1.5 %) were lower than those of the control group. The blood plasma glucose level was observed to move up constant with increasing dietary levels of GSP (0.5, 1.0 and 1.5 %), respectively.

There were no significant (P>0.05) differences in the plasma urea nitrogen and creatinine values among the rabbit groups fed the different tested levels of GSE. The presence of constant blood urea level observed in this study as the dietary levels of GSP increased in the diet is an indication that the animal may be in a state of equal nitrogen balance. This could be attributed to the presence of some of bioactive compounds contained in GSP as shown in Table 3. which has the

ability to help the energy metabolic pathway, thus making it easy for the animals to meet their energy requirement. This is in line with Kenneth and Carol (1998) who reported that in a state of equal nitrogen balance, the body protein (mainly muscles and liver proteins) are being built and used as energy. However, increase serum urea concentration may suggest an increase in activities of urea enzymes ornithine, carbonyl transferase and arginase (Ajagbonna *et al.*, 1999). Moreover, Creatinine content has been shown to depend on the quantity and quality of dietary protein (Esonu *et al.*, 2001).

The blood plasma ALT level of growing rabbits fed control diet was significantly ( $P < 0.05$ ) higher compared to those of GSP 0.5, 1.0 and 1.5% diets, respectively. The plasma total lipids levels of growing rabbits fed GSP (0.5, 1.0 and 1.5%) diets were significantly ( $p < 0.05$ ) lower than those of the control group. The blood plasma total lipids level was observed to move down constant with increasing dietary levels of GSP (0.5, 1.0 and 1.5%), respectively. Nevertheless, plasma total cholesterol, triglycerides, HDL, LDL and vLDL levels were insignificant among rabbit groups fed the different tested experimental diets.

Plasma cholesterol and triglycerides levels of rabbits fed GSP 0.5, 1.0 and 1.5% diet were not significant ( $p > 0.05$ ) compared to the control diets. However, the results showed that plasma cholesterol and triglycerides maintained a downward trend as the inclusion rate of GSP in the diet increased.

The blood plasma HDL level of rabbits fed the control diet was not significant ( $P > 0.05$ ) affected relative to those fed GSP 0.5, 1.0 and 1.5% diets, respectively. The blood plasma HDL level was observed to increase constant by supplementing different levels of GSP. The blood plasma LDL and vLDL level of rabbits fed the control diet were not significantly ( $P < 0.05$ ) affected relative to those on GSP 0.5, 1.0 and 1.56% diets, respectively. The blood plasma LDL and vLDL of growing rabbits were decreased slightly by increasing the supplemented GSP at 0.5, 1.0 and 1.5% in diets.

According present data shown in Table 3, it could be concluded that polyphenols and flavonoides of GSP had been able to reduce the total lipids, total cholesterol, triglycerides (TG), low density lipoprotein (LDL), and very low density lipoprotein (vLDL) concentrations of rabbits exposed to a high ambient temperature. In this respect, Yamakoshi *et al.*, (1999) suggested that proanthocyanidins from grape seeds might trap reactive oxygen species in plasma and intestinal fluid of the arterial wall, thereby inhibiting oxidation of LDL. Also, polyphenol fractions from grape seed rich in procyanidins achieved the best compromise between the direct and indirect (i.e. cell-mediated) types of action in protecting LDL against oxidation (Shafiee *et al.*, 2003). These results are in harmony with Attia *et al.*, (2010) who noted that chronic heat stress significantly increased plasma triglycerides. Besides, Teissedre and Waterhouse (2000) noted a high correlation between the total phenol content and low-density lipoprotein oxidation. Furthermore, Akbari and Torki (2013) suggested that the high concentration of antioxidants might decrease the serum concentration of triglycerides. In contrast, Chamorro *et al.*, (2012) found that plasma cholesterol, TG

and lipoproteins (HDL, LDL and vLDL) concentrations were not affected by dietary grape seed extract.

**Table (6).** Effect of grape seed powder (GSP) on blood biochemical of growing rabbits.

Items	Experimental diets				Sig.
	Control	0.5%GSP	1.0%GSP	1.5%GSP	
Total protein (g/dl)	5.43 <sup>c</sup> ±0.17	5.71 <sup>c</sup> ±0.28	6.45 <sup>b</sup> ±0.17	7.25 <sup>a</sup> ±0.10	**
Albumin (g/dl)	3.33 ±0.09	3.51 ±0.15	3.42 ±0.13	3.52 ±0.06	NS
Globulin (g/dl)	2.10 <sup>c</sup> ±0.12	2.20 <sup>c</sup> ±0.13	3.03 <sup>b</sup> ±0.25	3.73 <sup>a</sup> ±0.14	**
Albumin/Globulin ratio	1.61 <sup>a</sup> ±0.09	1.60 <sup>a</sup> ±0.05	1.17 <sup>b</sup> ±0.14	0.95 <sup>b</sup> ±0.05	**
Glucose (mg/dl)	57.25 ±3.57	53.62 ±1.23	56.33 ±0.87	56.51 ±2.54	NS
Urea-N (mg/dl)	64.04 <sup>b</sup> ±2.05	74.62 <sup>a</sup> ±2.68	68.39 <sup>b</sup> ±3.23	64.94 <sup>b</sup> ±3.54	NS
Creatinine (mg/dl)	0.65 ±0.02	0.62 ±0.01	0.64 ±0.02	0.62 ±0.01	NS
ALT(u/l)	9.00 <sup>a</sup> ±1.26	6.20 <sup>b</sup> ±0.20	7.80 <sup>ab</sup> ±0.92	6.00 <sup>b</sup> ±0.01	*
Total lipids (mg/l)	416.60 <sup>a</sup> ±13.41	328.60 <sup>b</sup> ±3.01	305.60 <sup>b</sup> ±2.25	258.20 <sup>c</sup> ±32.89	**
Total cholesterol (mg/dl)	108.25 ±14.28	103.17 ±18.58	99.68 ±10.12	82.86 ±5.55	NS
Triglycerides (mg/dl)	68.92 ±12.70	56.23 ±11.46	44.64 ±5.68	41.37 ±7.07	NS
HDL (mg/dl)	39.26 ±1.24	39.07 ±0.93	39.76 ±0.93	39.96 ±0.20	NS
LDL (mg/dl)	37.42 ±0.35	37.05 ±0.23	36.97 ±0.10	36.60 ±0.47	NS
vLDL (mg/dl)	13.78 ±2.54	11.24 ±2.29	8.93 ±1.13	9.47 ±2.37	NS

**a,b, c** Mean values with the same letter within the same row did not differ significantly (P>0.05).

### ***Blood plasma antioxidant constituents***

Beneficial health effects of edible phytochemicals are now considered to be an inexpensive, readily applicable, acceptable, and accessible approach to control and management a wide variety of effects oxidative stress (Tachibana, 2011). The effects of different levels of GSP on blood plasma biochemical (Lipid peroxide (malondialdehyde) (nmol/l), total antioxidant capacity (T-AOC) (mmol/l), Superoxide dismutase (u/l) and Glutathione peroxidase (u/l) antioxidant enzymes of rabbits are presented in Table 7. The obtained results revealed that the blood plasma lipid peroxide (malondialdehyde (MDA)) level of rabbits fed the control diet did not significantly (p>0.05) differ compared to those fed 0.5, 1.0 and 1.5% GSP treatment diets,

respectively. Niki (2008) found that lipid peroxidation represents oxidative decomposition of lipids and is an indicator of oxidative stress status in tissues and cells. The blood plasma lipid peroxide level was observed to reduce slightly with inclusion levels of GSP. The reduced levels of lipid peroxides following supplementation with grape seeds may have been associated with increased antioxidant enzyme activity and glutathione contents. In this trend, Choi *et al.*, (2010) found that the level of malondialdehyde (MDA) was lower in the serum of rabbits fed grape seed extract or grape peel powder plus cholesterol than in the serum of rabbits fed cholesterol alone. The opposite trend was noticed with plasma T-AOC, SOD and GSH-Px whereas the values of their parameters tended to be higher ( $P < 0.05$ ) with feeding rabbits GSP diets than of the control group. The results herein coincided with those reported by Alía *et al.*, (2003) who found that glutathione peroxidase activity increased after consumption of grape seeds and grape skins.

**Table (7).** Means ( $\pm$  SE) of Malondialdehyde (MDA), Total antioxidant capacity (T-AOC), Superoxide dismutase (SOD) and Glutathione peroxidase (GSH-Px) of growing rabbit's blood plasma as affected by supplementing different levels of Grape seed powder (GSP).

Items	Experimental diets				Sig.
	Control	0.5%GSP	1.0%GSP	1.5%GSP	
Malondialdehyde (MDA) (mmol/l)	14.06 $\pm 0.22$	13.34 $\pm 1.01$	12.94 $\pm 1.89$	9.88 $\pm 0.28$	NS
T-AOC(mmol/l)	130.20 <sup>d</sup> $\pm 2.13$	145.06 <sup>c</sup> $\pm 3.66$	152.32 <sup>b</sup> $\pm 7.28$	169.00 <sup>a</sup> $\pm 1.67$	**
Superoxide dismutase (SOD)(u/l)	24.58 <sup>c</sup> $\pm 0.16$	32.98 <sup>b</sup> $\pm 1.28$	36.22 <sup>a</sup> $\pm 0.68$	36.44 <sup>a</sup> $\pm 0.47$	**
Glutathione peroxidase (GSH-Px) (u/l)	456.80 <sup>c</sup> $\pm 4.99$	561.20 <sup>b</sup> $\pm 8.60$	581.00 <sup>b</sup> $\pm 10.38$	715.60 <sup>a</sup> $\pm 30.33$	**

**a,b,...** Mean values with the same letter within the same row did not differ significantly ( $P > 0.05$ ).

It well known that GSP contains 3.80% total phenols and 0.87% flavonoids, w/w so it could be a valuable source of natural antioxidants. Also, Sgorlon *et al.*, (2005) stated that the effect of grape polyphenols supplementation in New Zealand White rabbit diets on total glutathione (GSx), reduced glutathione (GSH) and oxidized glutathione (GSSG) increased significantly at 0.03 and 0.15 polyphenols mg/kg as a result of heat stress caused by the summer season.

Some studies were conducted on rats and noticed that old rats exposed to a prolonged heat stress, the concentration of total and reduced glutathione showed a significant increase instead of the expected depletion (Ozturk and Gumuslu, 2004; Choi *et al.*, 2012). These results indicated that grape polyphenols and flavonoides fraction are active compounds and act as an effective antioxidants and increases the resistance of plasma against oxidative stress through the activation of antioxidant enzyme system.

**Conclusively**, the results suggested that dietary supplementation of GSP at 0.5, 0.1 and 1.5% levels improved growth performance, economical efficiency and antioxidant status of rabbits compared to the control one, Moreover, grape seeds are rich source of phenolic and flavonoid compounds which activated the antioxidant enzyme system and relived the negative effects of heat stress for growing rabbits.

## REFERENCES

- Abd El-Monem, U.M.; Mahrose, Kh.M. and Khalil, B.A. (2009).** Effects of cage density and climatic conditions on the performance of growing rabbits. *Zagazig Veterinary J.*, **37**(2), 198 – 208.
- Abdel-Azeem, Noha (2005).** Effect of some organic components on rabbit's productive efficiency. Master of Science. Thesis, Faculty of Agriculture, Cairo, Egypt. 134p.
- Abdel-Khalek, A.M. (2010).** Antioxidants in rabbit nutrition: A review, *The 6<sup>th</sup> International Conference on Rabbit Production in Hot Climates*, Assiut, Egypt, 117 – 138.
- Ajagbonna O. P.; Onifade, K. I. and Suleman, U. (1999).** Haematological and biochemical changes in rats given extracts of *Calotropis procera* sokoto. *J. Vet. Sci.*, **1**: 36-42.
- Akbari, M.; and Torki, M. (2013).** Effects of dietary chromium picolinate and peppermint essential oil on growth performance and blood biochemical parameters of broiler chicks reared under heat stress conditions. *Int. J. Biometeorol.*, DoI 10.1007/s 00484 -013-0740-1.
- Akinola, A.O. and Abiola, S.S. (1991).** Blood chemistry and carcass yield of cockerels fed melon husk diets. *Trop. J. Anim. Sci.*, **2**: 39-44.
- Alía, M.; Horcajo, C.; Bravo, L. and Goya L. (2003).** Effect of grape antioxidant dietary fiber on the total antioxidant capacity and the activity of liver antioxidant enzymes in rats. *Nutr Res.*, **23**: 1251-67.
- AOAC (2007).** *Association Of Official Analytical Chemists*. Official Methods of Analysis, 18<sup>th</sup> Ed. AOAC, Washington, DC, USA.
- Attia, Y.A.; Hassan, R.A.; Tag El-Din, A.E.; and Abou-Shehema, B.M. (2010).** Effect of ascorbic acid or increasing metabolizable energy level with or without supplementation of some essential amino acids on productive and physiological traits of slow-growing chicks exposed to chronic heat stress. *J. Anim. Physiol. Anim. Nut.*, **95**: 744–755.
- Ayyat, M.S.; God, H.A.M.; EL-Aasar, T.A. and Abd El-Monem, U.M. (2004).** Alleviation of heat-stressed growing rabbits by using some feed additives under Egyptian condition. *Egyptian. J. of Nutrition and Feeds*, **7** (1): 83-96.
- Basalan, M. ; Gungor, T.; Owens, F.N. and Yalcinkaya, I. (2011).** Nutrient content and in vitro digestibility of Turkish grape pomaces. *Anim. Feed Sci. Technol.*, **169**: 194–198.
- Bayer crop science Egypt (2012)**, <http://www.egypt.cropscience.bayer.com/en/Crops/Grapes.aspx>

- Blasco A. and Ouhayoun J. (1996).** Harmonization of criteria and terminology in rabbit meat research. *World Rabbit Sci.*, **4**: 93–99.
- Brenes, A.; Viveros, A.; Goni, I.; Centeno, C.; Sayago-Ayerdy, S.G.; Arija, I. and Saura-Calixto, F. (2008).** Effect of grape pomace concentrate and vitamin E on digestibility of polyphenols and antioxidant activity in chickens. *Poultry Sci.*, **87**: 307-316.
- Cavani, C.; Maiani, A.; Manfredini, M. and Cristina, M. (1988).** The use of dehulled grape seed meal in the fattening of rabbits. *Annales de Zootechnie*, **37** (1), 1 – 12.
- Chamorro, S.; Viveros, A.; Centeno, C.; Romero, C.; Arija, I. and Brenes, A. (2012).** Effects of dietary grape seed extract on growth performance, amino acid digestibility, plasma lipids and mineral content in broiler chicks. *Animal*, **7**(4), 555–561.
- Chiu, D. T. Y.; Stults, F. H. and Tappel, A. L. (1976).** Purification and properties of rat lung soluble glutathione peroxidase. *Biochimica et Biophysica Acta*, **445**: 558–566.
- Choi, C. S.; Chung, H. K.; Choi, M. K. and Kang, M. H. (2010).** Effects of grape pomace on the antioxidant defense system in diet-induced hypercholesterolemic rabbits. *Nutr. Res. and Prac.*, **4** (2), 114-120.
- Choi, S.; Zhang, Z. and Seo, J. (2012).** Suppression of oxidative stress by grape seed supplementation in rats. *Nutr. Res. and Prac.*, **6**(1): 3-8.
- Dogan, A. and Celik, I. (2012).** Hepatoprotective and antioxidant activities of grape seeds against ethanol-induced oxidative stress in rats. *Br. J. Nutr.*, **107**: 45-51.
- Dorri, S.; Tabeidian, S.A.; Toghyani, M.; Jahanian, R. and Behnamnejad, F. (2012).** Effect of different levels of grape pomace on performance of broiler chicks. *The 1<sup>st</sup> International and The 4<sup>th</sup> National Congress on Recycling of Organic Waste in Agriculture*, 26–27 April, 2012, Isfahan, Iran.
- Duncan, D.B. (1955).** Multiple range and multiple F-tests. *Biometrics*, **11**: 1-42.
- Escribano-Bailon, T.; Gutierrez-Fernandez, Y.; Rivas-Gonzalo, J.C.R. and Santos-Buelga, C. (1992).** Characterization of procyanidins of *Vitis vinifera* variety Tinta del Pais grape seeds. *J. Agric. Food Chem.*, **40**: 1794–1799.
- Esonu, B.O.; Emenalom, O.O.; Udedibie, A.B.I.; Herbert, U.; Ekpor, C.F.; Okoli, I.C. and Ihukwumere, F.C. (2001).** Performance and blood chemistry of weaner pigs fed raw *Mucuna* beans (Velvet bean) meal. *Trop. Anim. Prod. Invest.*, **4**: 49-54.
- FAOSTAT, (2012).** <http://faostat3.fao.org/faostat-gateway/go/to/download/Q/QC/E>.
- Fuleki, T. and Ricardo da Silva, J.M. (1997).** Catechin and procyanidin composition of seeds from grape cultivars grown in Ontario. *J. Agr. Food Chem.*, **45**: 1156–1160.
- Ganong, W. F. (2005).** *Review of Medical Physiology*. McGraw-Hill Education, 22<sup>nd</sup> edition.
- Gladine, C.; Rock, E.; Morand, C.; Bauchart, D. and Durand, D. (2007).** Bioavailability and antioxidant capacity of plant extracts rich in polyphenols, given as a single acute dose in sheep made highly susceptible to lipoperoxidation. *Br. J. Nutr.*, **98**: 691–701.
- Goñi, I.; Brenes, A.; Centeno, C.; Viveros, A.; Saura-Calixto, F.; Rebole, A.; Arija, I. and Estevez, R. (2007).** Effect of dietary grape pomace and vitamin E on

- growth performance, nutrient digestibility, and susceptibility to meat lipid oxidation in chickens. *Poult. Sci.*, **86**: 508–516.
- González-Paramás, A. M.; Esteban-Ruano, S.; Santos-Buelga, C.; Pascual-Teresa, S. and Rivas-Gonzalo, J. C. (2004).** Flavanol content and antioxidant activity in winery products. *J. Agric. Food Chem.*, **52**: 234–238.
- Gorinstein, S.; Zemser, M.; Weisz, M.; Halevy, S.; Deutsch, J.; Tilus, K.; Feintuch, D.; Guerra, N.; Fishman, M. and Bartnikowska, E. (1994).** Fluorometric analysis of phenolics in persimmons. *Biosic Biotech Biochem.*, **58**: 1087–92.
- Goupy, P.; Hugues, M.; Boivin, P. and Amiot, M.J. (1999).** Antioxidant composition and activity of barley (*Hordeum vulgare*) and malt extracts and of isolated phenolic compounds. *J. Agric. Food Chem.*, **48**(12), 5834–5841.
- Halliwell, B. E. and Gutteridge, J. M. C. (1989).** *Lipid peroxidation: A radical chain reaction.* in *Free Radicals in Biology and Medicine*. 2<sup>nd</sup> ed. Oxford University Press, New York, NY. pp. 188–218.
- Hassan, Hanaa A.; Edrees, G. M.; El-Gamel, E. M. and El-sayed, E. A. (2014).** Amelioration of cisplatin-induced nephrotoxicity by grape seed extract and fish oil is mediated by lowering oxidative stress and DNA damage. *Cytotechnology*, **66**: 419 – 429.
- Ismail, A. M.; Sedki, A. A. and Abdallah, A. G. (2003).** Influence of black seed, garlic and onion supplementation on reproductive performance and immune functions in rabbits. *Egyptian J. Agric. Res.*, **81**: 1193-1207.
- Kenneth, S. S. and Carol, M. P. (1998).** *Anatomy and physiology: The unity of form and function.* 1<sup>st</sup> edition Mc Graw- Hill.
- Kermauner, A. and Laurenčič, A. (2008).** Supplementation of rabbit diet with chestnut wood extract: Effect on in vitro gas production from two sources of protein. In *Proceedings, 9<sup>th</sup> World Rabbit Congress. Verona, Italy, Jun 10–13*, pp. 689–693.
- Koracevic, D.; Koracevic, G.; Djordjevic, V.; Andrejevic, S.; Cosic V. (2001).** Method for the measurement of antioxidant activity in human fluids. *J. Clin. Pathol.*, **54**, 356-361.
- Lebas, F. (2004).** Reflections on rabbit nutrition with a special emphasis on feed ingredients utilization. In *Proceedings, 8<sup>th</sup> World Rabbit Congress, Puebla, Mexico, September 7-10*, pp. 686-736.
- Li, H.; Wang, X.; Li, P.; Li, Y. and Wang, H. (2008).** Comparative study of antioxidant activity of grape (*Vitis vinifera*) seed powder assessed by different methods. *J. Food Drug Anal.*, **16** (6), 67-73.
- Lphsi, (1990).** *Livestock And Poultry Heat Stress Indices.* Agriculture Engineering Technology. Guide 29634, USA. Clemson University, Clemson, SC.
- Marai, I. F. M.; Askar, A. A. and Bahgat, L. B. (2006).** Tolerance of New Zealand and Californian doe rabbits at first parity to the sub-tropical environment of Egypt. *Livest. Prod. Sci.*, **104**: 165-172.
- Marai, I. F. M.; Ayyat, M. S. and Abd El-Monem, U. M. (2000).** Young doe rabbit performance traits as affected by dietary, Zinc, Copper, Calcium or Magnesium, under winter and summer condition of Egypt. *Proceedings of the 7<sup>th</sup> World Rabbit Conference, Spain*, **8** :313 – 320.



- Mattila, P.; Astola, J. and Kumpulainen, J. (2000).** Determination of Flavonoids in Plant Material by HPLC with Diode-Array and Electro-Array Detections. *J. Agric. Food Chem.*, **48** (12), 5834–5841.
- Misra, H. P. and Fridovich, I. (1972).** The role of superoxide anion in the autoxidation of epinephrine and a simple assay for superoxide dismutase. *J. of Biol. Chem.*, **247**: 3170–3175.
- Monagas, M.; Hernández-Ledesman, B.; Garrido, I.G.; Martín-Álvarez, P.J.; Gómez-Cordovés, P. and Bartolomé, B. (2005).** Quality assessment of commercial dietary antioxidant products from *Vitis vinifera L* grape seeds. *Nutrition and Cancer*, **53**: 244-254.
- Nakamura, Y.; Tsuji, S. and Tonogai, Y. (2003).** Analysis of proanthocyanidins in grape seed extracts, Health foods and grape seed oils. *J. of Health Sci.*, **49**(1), 45-54.
- Niki, E. (2008).** Lipid peroxidation products as oxidative stress biomarkers. *Bio-Factors*, **34**(2), 171-180.
- Ohkawa, H.; Ohishi, N., and Yagi, K. (1979).** Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. *Anal. Biochem.*, **95**, 351-358.
- Oliveira, R. A; Narciso, C. D.; Bisinotto, R. S.; Perdomo, M. C.; Ballou, M. A.; Dreher, M. and Santos, J. E. (2010).** Effects of feeding polyphenols from pomegranate extract on health, growth, nutrient digestion, and immunocompetence of calves. *J. Dairy Sci.*, **93**(9), 4280-91.
- Oszmianski, J. and Sapis, J.C. (1989).** Fractionation and identification of some low molecular weight grape seed phenolics. *J. Agric. Food Chem.*, **37**: 1293–1297.
- Ozturk, O. and Gumuslu, S. (2004).** Age-related changes of antioxidant enzyme activities, glutathione status and lipid peroxidation in rat erythrocytes after heat stress. *Life Sci.*, **75**(13), 1551-1565.
- Pozuelo, M. J.; Agis-Torres, A.; Hervert-Hernández, D.; López-Oliva, M. E.; Muñoz-Martínez, E.; Rotger, R. and Goñi, I. (2012).** Grape antioxidant dietary fiber stimulates *Lactobacillus* growth in rat cecum. *J. Food Sci.*, **77** (2), 59 – 62.
- Rockenbach, I. I.; Gonzaga, L. V.; Rizelio, V. M.; Gonçalves, A. E.; Genovese, M. I. and Fett, R. (2011).** Phenolic compounds and antioxidant activity of seed and skin extracts of red grape (*Vitis vinifera* and *Vitis labrusca*) pomace from Brazilian winemaking. *Food Res. Inter.*, **44**: 897–901.
- Ruberto, G.; Renda, A.; Daquino, C.; Amico, V.; Spatafora, C.; Tringali, C. and Tommasi, N. (2007).** Polyphenols constituents and antioxidant activity of grape pomace from five Sicilian red grape cultivars. *Food Chem.*, **100**: 203–210.
- Santos-Buelga, C.; Francia-Aricha, E. M.; and Escribano-Bailon, M. T. (1995).** Comparative flavan-3-ol composition of seeds from different grape varieties. *Food Chem.*, **53**: 197–201.
- SAS (1998).** *SAS Procedure Guide*. Release 6.03 Edition. SAS Institute Inc., Cary NC, USA.
- Satyam, S. M.; Bairy, L. K.; Pirasanthan, R. and Vaihnav, R. L. (2013).** Grape seed extract and zinc containing nutritional food supplement decrease the oxidative

- stress induced by carbon tetrachloride in rats. *Inter. J. of Pharm. and Pharmac. Sci.*, **5**(4), 626 – 631.
- Sgorlon, S.; Stradaoli, G.; Stefanon, B.; Altimer, G. and Della Loggia, R., (2005).** Dietary grape polyphenols modulate oxidative stress in ageing rabbits. *Italian J. Anim. Sci.*, **4** (2), 541-543.
- Shafiee, M.; Carbonneau, M.A.; Urban, N.; Descomps, B.; and Leger, C.L. (2003).** Grape and grape seed extract capacities at protecting LDL against oxidation generated by Cu<sup>2+</sup>, AAPH or SIN-1 and at decreasing superoxide THP-1 cell production A comparison to other extracts or compounds. *Free Radic. Res.*, **37**: 573-584.
- Simonetti, P.; Ciappellano, S.; Gardana, C.; Bramati, L. and Pietta, P. (2002).** Procyanidins from *Vitis vinifera* seeds: in vivo effects on oxidative stress. *J. Agric. Food Chem.*, **50**: 6217-6221.
- Steven Lukefapor, W. D.; Hohenboken, W. D.; Cheeke, P. R.; Patton, N. M. and Kennick, W. H. (1981).** Carcass and meat characteristics of Flemish giant and New Zealand white purebred and terminal cross rabbits. *J. of Applied Rabbit Res.*, **4** (3), 66-72.
- Tachibana, H. (2011).** *Green Tea Polyphenol Sensing*. Proceedings of the Japan Academy, Series B., *Phys. Biol. Sci.*, March 11; **87**(3), 66–80.
- Teissedre, P. L.; and Waterhouse, A. L. (2000).** Inhibition of oxidation of human low - density lipoproteins by phenolic substances in different essential oils varieties. *J. Agric. Food Chem.*, **48**: 3801–3805.
- Urso, M. and Clarkson, P.M., (2003).** Oxidative stress, exercise, and antioxidant supplementation. *Toxicology*, **189**: 41-54.
- Vikili, R.; Rashidi, A.A. and Sobhanirad, S. (2010).** Effects of dietary fat, vitamin E and zinc supplementation on tibia breaking strength in female broilers under heat stress. *African J. Agric. Res.*, **5** (23): 3151-3156.
- Yamakoshi, J.; Kataoka, S.; Koga, T. and Ariga, T. (1999).** Proanthocyanidin-rich extract from grape seeds attenuates the development of aortic atherosclerosis in cholesterol-fed rabbits. *Atherosclerosis*, **142**: 139-149.
- Yilmaz, Y., and Toledo, R. T. (2004).** Major flavonoids in grapeseeds and skins: Antioxidant capacity of catechin, epicatechin, and gallic acid. *J. Agric. Food Chem.*, **52**: 255–260.