



GROWTH PERFORMANCE AND PHYSIOLOGICAL RESPONSES OF GROWING RABBITS SUPPLEMENTED WITH GREEN COFFEE EXTRACT

Enayat H. Abo EL-Azayem, Mervat N. Ghazal, Safaa A. Barakat,
Hanaa A. Bassiouni

Anim. Prod. Res. Inst., Agric. Res. Center, Dokki, Giza, Egypt

Corresponding author: Safaa A. Barakat Email, safaa_ataya@yahoo.com.

Received: 08/12/2021

Accepted: 23 /12/2021

ABSTRACT: Forty-five weaning male NZW rabbits at 6 weeks of age and an average weight of 655.6 ± 40 g were assigned randomly into 3 equal groups, fifteen rabbits/each for 8 weeks. The study was executed to evaluate the effect of supplementation green coffee extract (GCE) in diets on rabbit growth performance and physiological response. Rabbits in the 1st group were received a basal diet as a control. While, the 2nd and 3rd groups were received a basal diet supplemented with 2 and 4 ml GCE /kg diet, respectively. The results showed an improvement in the treated groups that were fed diets 2 and 4 ml GCE/ kg diets compared with control. Rabbits fed diets with 4 ml GCE/kg diets recorded significantly ($P<0.05$) higher body weight gain and better feed conversion ratio during all growth period than the control. Dietary with 4 ml GCE/kg diet was significantly higher in carcass weight and total edible parts (%) than that in the control; it also significantly reduced shoulder and abdominal fat. Supplementation of different levels of GCE to rabbits' diet achieved significant increase in beneficial bacteria (total bacterial count and lactobacilli counts) and reduction in pathogenic bacteria (*E.Coli clostridium* spp and ureolytic bacteria). Also, Hemoglobin, RBCs, WBCs and Hematocrite were significantly higher in groups supplemented with GCE than control. There were no significant differences in total protein, albumin, globulin, A/G ratio, liver and kidney functions and plasma lipid profile between all groups. Catalase activity was significantly higher in blood as GCE added to the diet. Meanwhile, all treated groups decreased insignificantly hydrogen peroxide (H_2O_2) compared to the control group. In conclusion, supplementation of 4 ml GCE/kg diet to NZW growing rabbits improved growth performance, microbial ecology, hematological and biochemical responses and economical efficiency ratio without adverse effects on liver and kidney functions.

Key words: Green Coffee Extract, Antioxidant, Growth, Blood, Microbial Ecology, Rabbits.

INTRODUCTION

Weaning is a critical period in growing rabbits which associated with change in rabbits feeding, and a lot of stress and disease sensitivity (El-Kholy et al., 2019). The high morbidity and mortality rates have a major economic influence on the production of rabbit meat. Microflora composition and fermentation pattern in the cecum play an important function in preventing digestive disorders in rabbits (Carabaño et al., 2006). The utilization of extract of many plants containing multi-phytochemicals has antioxidant power to enhance the growth performance, intestinal flora and the health status of growing rabbits (Ojo and Adetoyi, 2017).

Coffee arabica (Rubiace) is a common plant consumed in the world, and a major dietary source of many bioactive substances (Ríos-Hoyo and Gutiérrez-Salmeán, 2016). Coffee is a chemical compound including over thousand different molecular constituents, containing carbohydrates, nitrogenous, lipids, polyphenol, 4 major types were identified: lavan-3-ols (procyanidins and monomers) flavanols, hydroxycinnamic acid, anthocyanidins, many minerals, vitamins, alkaloids, diverse phenolic and non-phenolic compounds such as caffeine, kahweol, cafestol, trigonelline and chlorogenic acid that were associated with metabolism of lipids (Ukers, 2010).

The phenolic compounds such as chlorogenic acid are naturally present in many plants, and chlorogenic acid is a major component in green coffee beans, which has many beneficial effects such as anti-inflammatory and antimicrobial (Ding et al., 2014 and Ho et al., 2012), antioxidant (Chaves-Ulate and Esquivel-Rodríguez., 2019), anti-cancer

(Polamuri et al., 2020), regulatory influence of glucose and lipid concentrations, decrease triglyceride, cholesterol, decreases diabetes and obesity by reducing glucose up take in the small intestine (Greenberg et al., 2006). Green coffee is a raw unroasted coffee bean, containing a greater level of chlorogenic acid than coffee beans that have been roasted. Chlorogenic acid is a potent antioxidant effect in human body and health benefits (Moon et al., 2009). Many pharmacological studies observed that green coffee bean extract (GCBE) regulates hypertensive, vasoreactivity and metabolism of glucose (Kozuma et al., 2005 and Blum et al., 2007). Other studies reported that bioactive compounds in GCE may be improving the gut microbiota (Jaquet et al., 2009). Many of the health advantages associated with green coffee beans are due to chlorogenic acids and caffeine (Lukas et al., 2019).

Recently like green tea, extract from green coffee beans has received many studies for its use as health promoting supplementation. But, information on this extract is limited in rabbits.

So, the objective of this study was to investigate the efficacy of green coffee extract (GCE) as a natural source of antioxidants on growth performance of growing rabbits and carcass characteristics, blood parameters, oxidative status and caecal microbial activity.

MATERIALS AND METHODS

The current research was conducted in Rabbit Research, Unit at Sakha Research Station, and located in Kafr El-Shiekh governorate, Egypt, which is a part of the Animal Production Research Institute, Agricultural Research Center, Ministry of Agriculture, Dokki, Giza, Egypt.

Green Coffee Extract, Antioxidant, Growth, Blood, Microbial Ecology, Rabbits.

Experimental design and animals

Forty five New Zealand white male rabbits aged 6 weeks and weighed (655.6 ± 40 g) were divided randomly into three groups (n= 15each). The treated groups were: The 1st group as a control diet (without any supplementation). Rabbits in the 2nd and 3rd groups were fed the same basal diet but supplemented with 2 and 4 ml green coffee extract / kg diet, respectively. Rabbits were housed in individual galvanized wire pyramidal batteries (30 x 25 x 35 cm) with feeder and automatic nipple drinkers. The batteries were arranged in rows in a windowed house naturally ventilated. All rabbits were kept under the same management conditions. Feed and water were supplied *ad libitum*. Ingredients and chemical analysis of the experimental diets are shown in Table 1.

Preparation of green coffee extract

Green coffee beans were obtained from Haraz Company, Cairo, Egypt. A sample of 100 gram of green coffee bean was ground to a fine powder and then put in 1000 ml boiling water for 15 min. after centrifugation at 4000 rpm/min for 15 min. Then, filtered using Whatman filter paper (24 cm). The resulting extract was kept in refrigerator (4° C) and stored until being used then it was mixed with feed before pelleting (Gramaza et al., 2004).

Growth performance: The individual body weight gain (BWG) and feed intake (FI) were recorded weekly for each rabbit during the growing period from 6 to 14 week. Accordingly, the average of both daily gain (ADG, g/d) and feed intake (AFI, g/d) were calculated. The feed conversion ratio (FCR) was determined as a ratio of (g feed/g weight gain).

Carcass traits: At the end of the experiment period, 14 weeks of age 4 rabbits from each

experimental group were randomly taken for slaughter after being fasted for 12 hours to determine carcass characteristics according to Biasco and Ouhayoun (1996).

Blood metabolites: Immediately after slaughtering, blood samples of growing rabbits were collected in heparinized glass tubes from four rabbits in each group. Plasma was separated by centrifugation at 3000 rpm for 15 min and kept at -20°C until analyzed. Quantitative colorimetric determination of total protein (TP, g/dl), and albumin (Alb, g/dl) were executed using kits of Stanbio Laboratory Inc, procedure No. 0280. (San Antonio, Texas, USA). Globulin concentration (Glb, g/dl) was calculated by subtracting Alb values from TP values. Albumin/ Globulin ratio (A/G ratio) was calculated.

Quantitative colorimetric determination of triglycerides (TG, mg/dl), cholesterol (TC, mg/dl) and glucose (Glu, mg/dl) were executed by using kits of Spinreact, S.A./S.A.U. Ctra. Santa Coloma, 7 E-17176 SANT ESTEVE DE BAS (GI) SPAIN. Kits from Diamond for Lab Technology (Heliopolis, Cairo - Egypt) were used in determination of concentrations (mg/dl) of urea (BU) and creatinine (CR) as indicators for kidney functions. Activities of aspartate aminotransferase (AST, U/L) and alanine aminotransferase (ALT, U/L) as indicators for liver functions were determined colorimetrically using kits supplied by Q-Slap, ElQasar El Ainy St, Cairo -Egypt). All determinations were performed according to the procedures outlined by the respective manufacturers. Catalase (CAT, U/L) and hydrogen peroxide (H₂O₂, Mm/L) were colorimetrically determined using commercial kits (bought from Bio-Diagnostic, Cairo, Egypt, according to the manufacturers' instructions). Some fresh blood were taken after collection for determination of blood pictures

which included red blood cells count RBCs ($10^6/\mu\text{l}$), white blood cells count WBCs ($10^3/\mu\text{l}$), different leukocyte count %, hemoglobin (g/dl), hematocrite (%), according to Drew et al. (2004).

Microbiological measurements: Cecum contents were collected from slaughtered rabbits immediately to investigate total bacterial counts ($\times 10^6$ CFU/g caecal digesta) Cholsterida spp. ($\times 10^5$ CFU/g caecal digesta), E. Coli spp. ($\times 10^5$ CFU/g caecal digesta), lactobasillis bacteria ($\times 10^5$ CFU/g caecal digesta) and urealtic bacteria ($\times 10^5$ CFU/g caecal digesta) after slaughtering by Pour Plate Count Technique according to British Standard Institution (1991). Also, pH, NH_3 and Total Volatile Fatty Acid (TVFA) were determined in cecum, pH by digital pH-meter, NH_3 (mmol/l) and TVFA (mmol/l) by applying of the method (Conway, 1958).

Economical efficiency

To calculate the economical efficiency (EE) in the experimental diets for BWG, costs of feed needed for producing 1 kg of BWG were determined. The cost of the experimental diets was calculated to the price of ingredients at local market through all period. Economical efficiency was determined as a ratio among the return of weight gain and the cost of consumed feed.

Statistical analysis

The differences between experimental groups were statistically analyzed by using the general linear model (GLM) procedures of SAS (2004), by applying one-way analysis of variance (ANOVA). All results were analyzed using the following statistical model:

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

Where: Y_{ij} = the observation of ij ; μ = overall mean; T_i = effects of treatments, ($i = 1, 2$ and 3) and e_{ij} = experimental random error. The

significant differences between groups' means were separated by Duncan's multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

Growth performance

Growth performance of growing NZW rabbits from 6-14 weeks of age as affected by GCE supplementation is shown in Table 2. The average of final live body weight (FBW) of the treated experimental groups increased with increasing concentration of GCE in diets. The huge increase in weight was recorded in the group fed 4 ml GCE/ kg diets, which was significantly higher than the control group by 11.4% followed by group fed 2 ml GCE /kg diets by 3.7% at 14 weeks of age. The same trend was noted in the average daily weight gain (ADG), where the highest value was recorded in the 4 ml GCE /kg diet group compared to the control group during (6-10) , (10-14) and (6-14) weeks by 18.4%, 24.4% and 21.7% respectively. It is normal that feed intake increase with increasing age, but a significant increase was found for group 4 ml GCE/ kg diet with increasing age.

Concerning to FCR in the 3rd group (4 ml GCE/kg), it was improved ($P < 0.05$) during all growth periods. While, the worst value was found in rabbits of control group. The efficiency of production is greatly influenced by FCR as it is indicator for evaluating the health and productive performance. It is of interest to note in this investigation that FI and ADG were significantly increased when GCE was used at 4 ml/ kg diet. This increase in BWG could be owing to the presence of bioactive chemicals in GCE that improved digestion, boost nutrient absorption, and hence improve feed utilization efficiency. This result is confirmed by Ashour et al. (2020) who indicated that adding 2.5 g/kg of

Green Coffee Extract, Antioxidant, Growth, Blood, Microbial Ecology, Rabbits.

green coffee powder (GCP) to the diet in broilers diets recorded a higher BW compared than those consuming 1.25 g GCP/kg in the diet and the control at 5 weeks of age, also it enhanced FCR between the age of 3-5 and 1-5 weeks. Scheuermann et al. (2009) indicated that supplementation of phytogetic additives' could improve the feed consumption in broilers. Kohlert et al. (2000) found that enterocytes absorb phytochemicals in natural additives in the intestine, which are then promptly digested by the animal's body. These substances increase the production of cytoskeletal proteins, and then improved the absorption of the small intestine (Khajuria et al., 2002). Caro-Gómez et al. (2019) found that supplementation phenolic compound of GCE contributed to changes in gut microbiota, prompting increase in beneficial microbes, decrease the inflammatory effects, improved the metabolic pathways and nutrients digestibility in mice. Ilmiawati et al. (2020) concluded that low level of GCE has a useful effect on body weight in high-fat diet of rats. In contrast, Fathy and Rezq (2009) reported that oral administration of CBE in a green form caused significant decrease in body weight gain, feed efficiency ratio, but using orally extract of dark brown coffee beans in treated rats recorded the highest body weight gain and feed efficiency.

Carcass traits

The effect of experimental treatments on carcass weight and total edible part (%) are presented in Table 3. It could be noticed that there was a significant increase ($p < 0.05$) in pre-slaughter weight in rabbit fed with GCE at levels 2 and 4 ml / kg diet compared to rabbits fed a control diet. There was a significant difference in carcass weight and total edible parts (%) in growing rabbits treated with 4 ml

GCE/kg which recorded the highest weight than other groups Also, there were significant differences in dressing (%), among groups treated with 4 and 2 ml GCE/kg. These results may be due to addition proportionally to increase in carcass weight. Although, there were no significant differences in liver (%) between all treatments, but the increase in GCE level resulted in greater liver (%) compared to the control group.

Concerning to spleen (%), it was higher ($P < 0.05$) by supplementation 4 ml GCE /kg diet than other groups. Spleen plays an important part in immune system which helps the body fight infection by detect faults red blood cells, viruses, bacteria or other germs in blood, and produces white blood cells called lymphocytes to fight this infection (Mark, 2006). While, Edible giblet (%) was significantly higher in rabbit's fed 2 ml GCE/kg diets than the control group. Increasing of GCE level in diet resulted in a slight decrease in shoulder fat and abdominal fat as compared to control group. At the same time, Yang et al. (2003) and Guray et al. (2011) stated that antioxidants reduce the cytosolic malic enzyme activity which leads to reduction in abdominal fat deposition, and increased levels of green tea by-product in broiler diets lead to a reduction in the abdominal fat percentage. This result was confirmed by Ashour et al. (2020) who found that broilers fed diets with 2.5 g GCP/kg had the lowest abdominal fat percentage ($P < 0.05$) compared to the control at 5 weeks of age, while those fed 1.25 g GCP/kg diets recorded significant increase in abdominal fat (%) as compared to control group. Choi et al. (2016) reported that the active compound Caffeoylquinic acids (3-CQA) found in GCBE, decreased body fat deposition by the

modulation of adipogenesis and lipogenesis in mice.

Microbial examination

The pH value showed insignificant differences between the groups fed experimental diets and control group Table 4. It was found that increased GCE supplementation resulted in a significant increase in beneficial bacteria (total bacterial count and lactobacilli bacteria) and, in contrast, a significant decrease in pathogenic bacteria (population of urolytic bacteria, *clostridium spp* and *E.coli spp*) in ceacum digesta. The highest value of beneficial bacteria was recorded in the group of rabbits fed with GCE at level 4 ml/kg and also, recorded the lowest value in pathogenic bacteria followed by GCE at level 2 ml/kg then the control group. That is GCE addition enhancing the natural host defenses. The same results were obtained for TVFA level and NH₃ concentration which increased and decreased significantly, respectively. Ammonia is quickly absorbed whereas urea is poorly used, which may contribute to rising nitrogen retention and body weight gain, and it observed that treated rabbits with GCE lead to the reduction of pathogenic bacteria count and reflected on increasing count of beneficial bacteria. These results are consistent with, Ashour et al. (2020) who reported that the addition of GCP 2.5 g/kg in the diet can be applied to increase the beneficial bacteria count and decrease the pathogenic populations as *E.coli* in the broiler's gut. TVFA production from lactose and lactate by bacteria fermentation in large intestine and cecum (Szylite et al., 1988), this increase due to increased in lactobacillus bacteria count in GCE groups. Lactobacillus in animal intestines is useful to the host, where

coliforms are harmful as reported by Santos et al. (2006). Also, Nishitsuji et al. (2018) recorded that coffee and its potent antibacterial components have an effect on the bacteria in mice's guts, as caffeine (a component of green coffee) which improved immunological tolerance to harmful microorganisms by increasing the focus of certain immune-competent cells and enhancing activity of lysozyme.

Hematological parameters

The results of hematological analysis are shown in Table 5. It can be noticed that there were significant increase ($P<0.05$) in Hb, RBCs, WBCs and hematocrit % of GCE supplemented growing rabbits as compared to the control group, especially the group 4 ml GCE/kg which recorded the highest value compared to the other groups. However, these values are still within the normal ranges (Hewitt et al., 1989). The addition of GCE caused an excess in total RBC's which in turn caused an excess in Hb values, due to the positive relationship between RBC's and Hb (Sturkie, 1986). The same trend was recorded for lymphocytes in GCE groups which significantly increased than the control group. While, neutrophil, monocytes and eosinophils (%) were significantly decreased in treated groups compared to the control group. The present results suggest that the experimental treatments with GCE may be enhancing immune system activity, and a positive effect was observed on liver and spleen where RBCs and lymphocytes are synthesized (Feldman et al., 2000).

Blood constituents

The results in Table 6. Showed that the most of blood constituents not significantly differed among all groups except for Glb and triglycerides (TG). For Glb level, there was a

Green Coffee Extract, Antioxidant, Growth, Blood, Microbial Ecology, Rabbits.

significant ($P < 0.05$) increase in rabbits supplemented with GCE at level 4 ml/kg diets compared to the other two groups. Also, the best value of A/G ratio was recorded in group fed 4 ml GCE /kg diets than other experimental groups. This improvement may be attributed to feed on GCE and to the increasing animal resistance to any physical stress. Furthermore, it is known that the level of TP is an indication of immune status of rabbits; globulins are transporter proteins for thyroid and steroid hormones which play important role in gaining natural immunity to infection (Ganong, 2005). High level of Glb is an important protein to antibodies production and immunological response. Furthermore it, may be indicated to increased the liver to synthesize globulins for immunological functions (Sunmonu and Oloyede, 2007 and Scanes, 2015). Meanwhile, the lower level for AST and ALT activity was recorded in group fed 4 ml GCE /kg diets than other groups. Plasma of BU and CR concentration were recorded insignificant differences between treated and control groups. This result agree with Ashour et al. (2020), it could be noticed that diets supplemented with GCE at 1.25 and 2.5g/kg in broiler chicks had no significant changes in levels of TP and Alb, BU and CR and activities of both AST, ALT compared to the control group. This indicated to the improvement in kidney functions, protein metabolism and then rise of nitrogen retention and utilization (Abou-Sekken et al., 2012). Triglycerides was decreased significantly in group 4 ml GCE/kg as compared to control group. This result is in agreement with Caro-Gómez et al. (2019) who found significant reduced in ALT, AST and TG. Concerning the effect of GCE on Glu, it can be shown that there was an increase in the

level of Glu in treated group with GCE as compared to control group. In this concern, Van Dam et al. (2004) observed that the increase in Glu level, may be related to supplementation of Coffee that improved glucose metabolism. So, the high level of Glu indicated the improvement of metabolism and utilizing feed nutrients. In contrast, Fathy and Rezq (2009) reported that GCBE in mice diets caused significant increase in level of TG and TC. AST, ALT, BU and CR, and decreased Glu level in group with orally GCE compared to the other groups.

Antioxidative status in blood

The effect of GCE on blood antioxidant of growing rabbits are illustrated in Table 7. Catalase enzyme activity was increased significantly in rabbits fed 4 ml GCE/kg diets by 17.7% as compared with control group. This increase in CAT activity could be attributed to GCE in diets which it enhanced the antioxidant system via increasing the activity of CAT, which converts 6 million H_2O_2 molecules into H_2O and O_2 in 1 minute (Sies, 2015). Antioxidant enzymes are the first line of defense to protect the organism from harmful peroxidation (Mao et al., 2014). This increase in CAT activity caused insignificantly reduction in H_2O_2 by 33% for 4 ml GCE/kg diet group compared to control group. Therefore, the current study confirmed that the addition of GCE success to reduce the oxidative stress (OS) that occurs during the growing periods of rabbits. These finding agree with Zain et al. (2018) who showed that the supplement coffee in diet as a source of polyphenols have antioxidant activity that reduced (OS). In contrast, Ashour et al. (2020) found that adding GCP in broilers diets did not affect significantly in serum (OS) between the control and the treated groups.

Enayat H. Abo EL-Azayem et al.

Economical efficiency

Effect of green coffee extract supplementation at different levels are shown in Table 8. The present results indicated that rabbits fed diets with 4 ml GCE/kg diets had improvement in economical efficiency by 44.5% compared with the control group, and the best relative economical efficiency which had higher net revenue than other groups.

CONCLUSION

The present results concluded that supplementation of green coffee extract

(GCE) as a natural antioxidant improves the growth performance, hematological and plasma biochemical responses. Moreover, it improved the microbial environment resulting in an increase in beneficial bacteria and decrease in pathogenic bacteria, and improved the economic efficiency. From the practical point of view, the best supplementation of GCE is a level of 4 ml/kg diet, so we recommend adding it to the diet of growing rabbits.

Table (1): Ingredients and chemical analyses of the basal diet.

Ingredients %		Chemical analysis ⁽²⁾ %	
Alfalfa hay (12%)	28.55	Crude protein	17.10
Barley	34.40	Crude fiber	12.74
Wheat bran	11.00	Crude fat	1.99
Soybean meal (44%)	17.00	Digestible energy(kcal/kg)	2500
Molasses	5.00	Calcium	1.30
Limestone	0.85	Total Phosphorus	0.80
Di-calcium phosphate	2.10	Methionine	0.63
Sodium chloride	0.40	Lysine	0.86
		Meth.+Cyc.	0.91
Mineral and vitamin premix ⁽¹⁾	0.30		
DL-Methionine	0.40		
Total	100		

⁽¹⁾ Each 3 kg contain: 6000000 IU Vit. A; 900000 IU Vit. D3; 40000 mg Vit. E; 2000 mg Vit. K3; 2000 mg Vit. B1; 4000 mg Vit. B2; 2000 mg Vit. B6; 10 mg Vit. B12; 50 mg Biotin; 10000 mg Pantothenic acid; 50000 Niacin; 3000 mg Folic acid; 250000 mg Choline; 8500 mg Mn; 50000 mg Zn; 50000 mg Fe; 200 mg I; 100 mg Se, 5000 mg Cu, and 100 mg Co.

⁽²⁾ According to CLFF, 2001.

Green Coffee Extract, Antioxidant, Growth, Blood, Microbial Ecology, Rabbits.

Table (2): Growth performance of growing NZW rabbits as affected by dietary supplementation of green coffee extract.

Item	Control	Green coffee extract (ml/kg Diet)		±SE
		2ml	4ml	
Average body weight (g)				
Initial body weight (g)	675.2	679.3	612.3	41.3
Final body weight (g)	2069.6 ^b	2146.2 ^b	2305.0 ^a	34.2
Average daily weight gain (g/day)				
Weeks 6-10	27.23 ^b	27.61 ^b	32.23 ^a	0.73
Weeks 10-14	22.69 ^b	24.76 ^{ab}	28.23 ^a	1.38
Weeks 6-14	23.69 ^b	24.69 ^b	28.84 ^a	0.80
Average daily feed intake (g/day)				
Weeks 6-10	89.61 ^a	91.38 ^a	82.76 ^b	1.60
Weeks 10-14	116.15 ^b	107.53 ^c	124.23 ^a	1.80
Weeks 6-14	102.84 ^{ab}	99.61 ^b	103.46 ^a	1.20
Feed conversion ratio (g feed/ g gain)				
Weeks 6-10	3.30 ^a	3.34 ^a	2.58 ^b	0.10
Weeks 10-14	5.42 ^a	4.62 ^{ab}	4.42 ^b	0.28
Weeks 6-14	4.43 ^a	4.06 ^b	3.62 ^b	0.14

a, b and c Means in the same row with different superscripts are significantly different at (P<0.05). SE = Standard error of means.

Table (3): Carcass characteristics of growing NZW rabbits as affected by dietary supplementation of green coffee extract.

Item	Control	Green coffee extract (ml/kg Diet)		±SE
		2ml	4ml	
Pre-slaughter weight(g)	2112.50 ^b	2266.25 ^a	2305.00 ^a	44.26
Carcass weight (g)	1340.0 ^b	1246.25 ^b	1472.50 ^a	37.26
Dressing (%)	63.52 ^a	55.02 ^b	63.91 ^a	1.93
Liver (%)	2.48	4.56	4.81	0.91
Heart (%)	0.33	0.54	0.44	0.15
Kidney (%)	0.53 ^b	0.84 ^a	0.71 ^a	0.04
Spleen (%)	0.056 ^b	0.054 ^b	0.085 ^a	0.008
Edible giblets (%) ¹	3.35 ^b	5.95 ^a	4.61 ^{ab}	0.51
Total edible parts (%) ²	66.8 ^b	61.0 ^b	68.5 ^a	1.60
Cecum weight (g)	104.5	139.2	139.1	14.59
Shoulder fat	0.29 ^a	0.22 ^{ab}	0.16 ^b	0.038
Abdominal fat	1.34 ^a	1.14 ^a	0.72 ^b	0.088

a and b Means in the same row with different superscripts are significantly different at (P<0.05). SE = Standard error of means. ¹Edible Giblets %=(liver+ kidney+ heart)/Pre-slaughter weight (g)*100. ²Total edible parts%=(carcass wt.+ edible giblets wt.)/Pre-slaughter weight (g)*100.

Table (4): Caecum count and microbial activity of growing NZW rabbits as affected by dietary Supplementation of green coffee extract.

Item	control	Green coffee extract (ml/kg Diet)		±SE
		2ml	4ml	
PH	6.75	6.72	6.80	0.039
NH ₃ (mmol/l)	10.11 ^a	5.28 ^b	4.65 ^c	0.196
Total bacterial count (10 ⁶)	17.35 ^c	19.73 ^b	20.64 ^a	0.242
Ureolytic bacteria (10 ⁵)	2.87 ^a	1.76 ^b	1.28 ^c	0.104
Lactobacilli (10 ⁵)	6.92 ^b	10.51 ^a	10.93 ^a	0.311
Escherichia coli (10 ⁵)	1.50 ^a	1.19 ^b	1.09 ^b	0.060
Clostridium spp.	5.20 ^a	3.38 ^b	3.19 ^b	0.107
TVFA (mmol/l)	57.07 ^c	63.40 ^b	76.61 ^a	1.203

a,b and c Means in the same row with different superscripts are significantly different at (P<0.05). SE = Standard error of means.

Table (5): Some blood hematological values of growing NZW rabbits as affected by dietary supplementation of green coffee extract.

Item	control	Green coffee extract (ml/kg Diet)		±SE
		2ml	4ml	
Hemoglobin (g/dl)	10.27 ^b	12.97 ^a	13.42 ^a	0.462
RBCs (10 ⁶ / μl)	5.67 ^b	6.20 ^{ab}	6.67 ^a	0.235
Hematocrite %	32.10 ^b	33.77 ^a	34.37 ^a	0.367
WBCs (10 ³ /μl)	5.80 ^b	9.00 ^{ab}	12.00 ^a	1.24
Differential leukocyte count (%)				
Lymphocyte	39.72 ^b	55.70 ^a	62.00 ^a	3.153
Neuterophil	38.95 ^a	28.95 ^b	23.52 ^b	2.195
N/L ratio	1.03 ^a	0.52 ^b	0.37 ^b	0.08
Monocyte	7.32 ^a	6.32 ^b	6.07 ^b	0.245
Eosinophil	2.40 ^a	1.90 ^b	1.40 ^c	0.133

a,b and c Means in the same row with different superscripts are significantly different at (P<0.05). SE = Standard error of means.

Green Coffee Extract, Antioxidant, Growth, Blood, Microbial Ecology, Rabbits.

Table (6): Some blood constituents of growing NZW rabbits as affected by dietary supplementation of green coffee extract.

Item	control	Green coffee extract (ml/kg Diet)		±SE
		2ml	4ml	
Total protein (g/dl)	5.83	7.96	6.26	0.73
Albumin (g/dl)	3.96	6.47	3.90	0.75
Globulin (g/dl)	1.86 ^{ab}	1.5 ^b	2.36 ^a	0.19
A/G ratio	2.16	4.83	1.66	0.96
AST(U/L)	29.3	30.6	27.0	2.95
ALT(U/L)	74.3	60.6	53.0	8.48
Urea (mg/dl)	34.3	38.0	39.0	2.91
Creatinine (mg/dl)	0.70	1.03	0.73	0.09
Cholesterol (mg/dl)	63.6	70.3	70.3	3.60
Triglyceride (mg/dl)	86.3 ^a	57.3 ^{ab}	44.0 ^b	10.6
Glucose (mg/dl)	136.6	149.0	147.3	4.74

a and b Means in the same row with different superscripts are significantly different at (P<0.05). SE = Standard error of means.

Table (7): Catalase and H₂O₂ in blood of growing NZW rabbits as affected by dietary supplementation of green coffee extract.

Item	control	Green coffee extract (ml/kg Diet)		±SE
		2ml	4ml	
Catalase (U/L)	478.47 ^b	484.83 ^b	563.53 ^a	19.53
H ₂ O ₂ (Mm/L)	0.359	0.315	0.242	0.035

a and b Means in the same row with different superscripts are significantly different at (P<0.05). SE = Standard error of means.

Table (8): Economical efficiency of growing NZW rabbits as affected by dietary supplementation of green coffee extract.

Item	control	Green coffee extract (ml/kg Diet)	
		2ml	4ml
Average total weight gain/rabbi(kg)	1.39	1.46	1.69
Total revenue/rabbit(LE) (A)	55.6	58.68	67.72
Total feed intake/rabbit(kg)	5.75	5.57	5.79
Price of feeding/kg(LE)	6.5	6.68	6.86
Total cost of feed/rabbit(LE) (B)	37.37	37.21	39.71
Net revenue/rabbit(LE) ¹	18.23	21.47	28.01
Economical efficiency% ²	48.8	57.69	70.53
Relative economical efficiency ³	100	118	144

Price of 1 kg live body weight = 40 LE

(1) Net revenue = A – B.

(2) Economic efficiency = (A-B/B x 100).

(3) Relative Economic Efficiency= Economic efficiency of treatments other than the control/ Economic efficiency of the control group.

REFERENCES

- Abou-Sekken M S, Ali M N, El-Mostafa K M 2012.** Improve utilization of low protein diets using thyme, citric acid or sodium sulphate in Japanese Quails. *Egypt. J. Nutr. Feeds*, 15(1): 113-121.
- Ashour E A, Abd El-Hack M E, Shafi M E, Al-Ghamdi W Y, Taha A E, Swelum A A, Toufarelli V, Mulla Z S, El-Ghareeb W R, El-Saadony M T 2020.** Impacts of green coffee powder supplementation on growth performance, carcass characteristics, blood indices, meat quality and gut microbial load in broilers. *J. Agric.*, 10: 457. doi:10.3390.
- Biasco A, Ouhayoun J 1996.** Harmonization of criteria and terminology in rabbit meat research. *World Rabbit Sci.*, 4: 93-99. DOI:https://doi.org/10.4995/wrs 1996.278.
- Blum J, Lemaire B, Lafay S 2007.** Effect of a green decaffeinated extract on glycemia. *Nutr. Food Res.*, 6 (3):13-17.
- British Standard Institution 1991.** Microbiology Examination of Food and Animal Feeding Stuff. Part 1, 376 p.
- Carabaño R, Badiola I, Licois D, Gidenne T 2006.** The digestive ecosystem and its control through nutritional or feeding strategies. In: Maertens L., Coudert P. (Eds.). *Recent Advances in Rabbit Sci.*, ILVO, Merelbeke, Belgium, 300 p.
- Caro-Gómez E, Jelver A S, Juan S E, Rafael Á, Mauricio N, Sonia M, Eliana P V, Jorge H T, Julio C J, Yudy M L, Katalina M, José R R 2019.** Green coffee extract improves cardiometabolic parameters and modulates gut microbiota in high-fat-diet-fed ApoE^{-/-} Mice. *Nutri.*, 11: 497., doi:10.3390/nu11030497 www

Green Coffee Extract, Antioxidant, Growth, Blood, Microbial Ecology, Rabbits.

- Chaves-Ulate E, Esquivel-Rodríguez P 2019.** Chlorogenic acids present in coffee: antioxidant and antimicrobial capacity. *Agron. Mesoam.*, 30: 299-311.
- Choi B K, BumPark S, RyungLee D I, JinLee H, YuJin Y, HwanYang S, WonSuh J 2016.** Green coffee bean extract improves obesity by decreasing body fat in high-fat diet-induced obese mice. *Asian Pacific J. Trop. Med.*, 9 (7): 635-643.
- CLFF, Central Lab for Food & Feed. Technical Bulletin N. 1 2001.** Feed Composition Tables for Animal and Poultry Feedstuffs Used in Egypt.
- Conway E J 1958.** Micro Diffusion Analysis and Volumetric Error. 4th ed. The McMillan Co., New York, USA, 687p.
- Ding M, Bhupathiraju S N, Chen M, Van Dam R M, Hu FB 2014.** Caffeinated and decaffeinated coffee consumption and risk of type 2 diabetes: a systematic review and a dose-response meta-analysis. *Diabetes Care.*, 37(2): 569-586.
- Drew P, Careles CR, Trevor B, John L 2004.** Determination of total serum protein. *Clin.Chem.*, 2:1642-1643.
- Duncan D B 1955.** Multiple range and multiple F tests. *Biometrics.*, 11: 1- 42.
- El-Kholy K H, Tag El-Deen H T, Abd-El-Lateif A I, Mekawy A I 2019.** Effects of dietary selenium sources on metabolic, enzymatic and immunoglobulin serum profiles in growing rabbits. *Pak. J. Nutr.*, 18: 430-436.
- Fathy N M, Rezq A A 2009.** Effect of green and roasted coffee beans extracts on some physiological parameters and histological structures in rats. *Afr. J. Biol. Sci.*, 5 (2):181-195.
- Feldman B F, Zinkl J G, Jain N C 2000.** Schalm, A *Veterinary Hematology* 5th ed . Lippincott Williams and Wilkins, Philadelphia, USA.1120-1124.
- Ganong W F 2005.** Review of Medical Physiology. McGraw-Hill Education, 22nd ed.
- Gramza A, Korezak J, Hes M, Jedrus K, Golinska A 2004.** Tea extracts influence on catalytical properties of F⁺2 in lipids. *Polish. J. Environ. Stud.*, 13: 143-146.
- Greenberg J A, Boozer C N, Geliebter A 2006.** Coffee: Diabetes and weight control. *Am J. Clin Nutr.*, 84(4): 682-693.
- Guray E, Ocak N, Altop A, Cankaya S, Aksoy H M, Ozturk E 2011.** Growth performance, meat quality and caecal coliform bacteria count of broiler chicks fed diet with green tea extract. *Asian-Australas. J. Anim. Sci.*, 24: 1128-1135.
- Hewitt, C. D.; D. J. Innes; J. Savary and M. R. Wills 1989.** Normal biochemical and hematological values in New Zealand white rabbits. *Clin. Chem.*, 35 (8): 1777 – 1779.
- Ho L, Varghese M, Wang J, Zhao W, Chen F, Knable L A 2012.** Dietary supplementation with decaffeinated green coffee improves diet-induced insulin resistance and brain energy metabolism in mice. *Nutr. Neuro.Sci.*, 15(1): 37- 45.
- Ilmiawati C, Fitri F, Rofnda Z D, Reza M 2020.** Green coffee extract modifies body weight, serum lipids and TNF- α in high-fat diet-induced obese rats. *BMC.Res.Notes.*, 13: 208-212. <https://doi.org/10.1186/s13104-020-05052-y>.
- Jaquet M, Rochat I, Moulin J, Cavin C, Bibiloni, R 2009.** Impact of coffee consumption on the gut microbiota: A

- human volunteer study. *Int. J. Food Microbiol.*, 130: 117–121.
- Khajuria A, Thusu N, Zutshi U 2002.** Piperine modulates permeability characteristics of intestine by inducing alterations in membrane dynamics: Influence on brush border membrane fluidity, ultrastructure and enzyme kinetics. *Phytomedicine.*, 9: 224-231.
- Kohlert C, van Rensen I, März R, Schindler G, Graefe EU, Veit M 2000.** Bioavailability and pharmacokinetics of natural volatile terpenes in animals and humans. *Planta Méd.*, 66: 495-505.
- Kozuma K, Tsuchiya S, Kohori J, Hase T, Tokimitsu I 2005.** Antihypertensive effect of green coffee bean extract on mildly hypertensive subjects. *Hypertens. Res.* 28(9): 711-718.
- Lukas M, Anatol S, Matthias S, Helmut K M 2019.** Green coffee infusion as a source of caffeine and chlorogenic acid, *J. Food Comp. Analy.*, 84:103307.
- Mao X, Lv M, Yu B, He J, Zheng P, Yu J, Wang Q, Chen D 2014.** The effect of dietary tryptophan levels on oxidative stress of liver induced by diquat in weaned piglets. *J. Anim. Sci. Biotechnol.*, 49: 5-11.
- Mark FC 2006.** Normal structure, function, and histology of the spleen. *Toxicol. Pathol.*, 34: 455-465.
- Moon J K, Yoo H S, Shibamoto T 2009.** Role of roasting conditions in the level of chlorogenic acid content in coffee beans: Correlation with coffee acidity. *J. Agric. Food Chem.*, 57: 5365-5369.
- Nishitsuji K, Watanabe S, Xiao J, Nagatomo R, Ogawa H, Tsunematsu T, Umemoto H, Morimoto Y, Akatsu H, Inoue K 2018.** Effect of coffee or coffee components on gut microbiome and short-chain fatty acids in a mouse model of metabolic syndrome. *Sci. Rep.*, 8: 16173. DOI:10.1038/s41598-018-34571-9.
- Ojo O A, Adetoyi SA 2017.** Effect of *Moringa oleifera* leaf extract on the haematological and serum biochemistry of rabbits reared in a semi-humid environment. *Afr. J. Biotechnol.*, 16: 1386-1390.
- Polamuri D, Valentina C G, Suresh R, Islam A 2020.** In-vitro anticancer and antioxidant activity of green coffee beans extract. *Asian Food Sci. J.*, 17(2): 24-35.
- Ríos-Hoyo A, Gutiérrez-Salmeán G 2016.** New dietary supplements for obesity: What we currently know. *Curr. Obes. Rep.*, 5: 262-270.
- Santos A, Mauro MS, Diaz DM 2006.** Prebiotics and their long-term influence on the microbial populations of the mouse bowel. *Food Microbiol.*, 23:498-503.
- SAS 2004.** SAS Statistics Users Guide Statistical Analysis System. 8th Edn., 8.2 - Version SAS Institute Inc, Cary, NC.
- Scanes C G 2015.** Eds. *Sturkie's Avian Physiology*, 6th ed.: Academic Press, London, UK, 9 p.
- Scheuermann G N, Cunha Junior A, Cypriano L, Gabbi A M 2009.** Phytogetic additive as an alternative to growth promoters in broiler chickens. *Ciênc. Rural.*, 39: 522–527.
- Sies H 2015.** Oxidative stress: a concept in redox biology and medicine. *Redox. Biol.* 4: 180-183.
- Sturkie P D 1986.** Body Fluids: Blood. In Sturkie P. D. [ed.], *Avian Physiology*. (4th ed). Springer-Verlag, Berlin, pp. 102-120.
- Sunmonu T O, Oloyed O B 2007.** Biochemical assessment of the effects of

Green Coffee Extract, Antioxidant, Growth, Blood, Microbial Ecology, Rabbits.

- crude oil contaminated catfish (*Clarias gariepinus*) on the hepatocytes and performance of rat. *Afr. J. Biochem. Res.*, 1: 83-89.
- Szylite O J, Dabard M D, Christiane D, Bensaada M, Raibaud P 1988.** Production of volatile acids as result of bacterial interactions in the cecum of gnotobiotic rats and chickens fed alactose-containing diets. *Reprod. Nutr. Develop.* 28 (6A):1455-1465.
- Ukers W H 2010.** All about coffee. (2nd ed). Gala Research.725p.
- Van Dam R M, Pasman W J, Verhoef P 2004.** Effect of coffee consumption on fasting blood glucose and insulin concentrations. *Diabetes Care.*, 27(12):2990-2992.
- Yang C J, Yang I Y, Oh D H, Bae I H, Cho S G, Kong I G, Uganbayar D, Nou I S, Choi K S 2003.** Effect of green tea by-product on performance and body composition in broiler chicks. *Asian-Australas. J. Anim. Sci.*, 16: 867-872.
- Zain M Z M, Bab AS, Shori A B 2018.** Effect of polyphenols enriched from green coffee bean on antioxidant active and sensory evaluation of bread. *J King Saud Univ Sci.*, 30: 278-28.

أداء النمو والاستجابات الفسيولوجية للأرانب النامية المعاملة بمستخلص البن الأخضر

عنايات أبو العزائم حسن، مرفت نبيل غزال، صفاء عطايا بركات وهناء عوض بسيوني
معهد بحوث الإنتاج الحيواني - مركز البحوث الزراعية-الدقى -جيزة-مصر

تم تقسيم إجمالي عدد ٤٥ من ذكور الأرانب النيوزيلندية المفطومة عند عمر ٦ أسابيع بمتوسط وزن 600.6 ± 40 جم بشكل عشوائي إلى ٣ مجاميع متساوية، ١٥ أرنب / لكل معاملة لمدة ٨ أسابيع. أجريت الدراسة لتقييم تأثير مكملات مستخلص البن الأخضر (GCE) في الغذاء على أداء النمو والاستجابة الفسيولوجية للأرانب. غذيت الأرانب في المجموعة الأولى علي العليقة الأساسية كمجموعة مقارنة. بينما غذيت المجموعة الثانية والثالثة علي العليقة الأساسية مضاف إليها ٢ و ٤ مل من GCE /كجم علي التوالي. أظهرت النتائج تحسنا في المجموعات المعاملة التي تم تغذيتها علي عليقة تحتوى ٢ و ٤ مل من GCE /كجم زيادة معنوية ($P < 0.05$) في وزن الجسم وتحسن معدل التحويل الغذائي أثناء فترة النمو عن المجموعة الكنترول. سجلت المجموعة التي تغذت علي ٤ مل GCE / كجم عليقة أعلى وزن للذبيحة و خصائص الذبيحة % مقارنة بالكنترول. وأقل نسبة من دهون الكتف والبطن. أدت إضافة مستويات مختلفة من GCE إلى عليقة الأرانب إلى زيادة كبيرة في البكتيريا النافعة (total bacterial count and lactobacilli counts) وأيضاً انخفاض البكتيريا الضارة (*E. Coli*) (*clostridium spp* and *ureolytic bacteria*). كان كل من الهيموجلوبين، وكريات الدم الحمراء والبيضاء والهيماتوكريت أعلى معنويًا في المجاميع المعاملة بـ GCE عن الكنترول. لا يوجد تأثير معنوي في كلا من البروتين الكلى والألبومين، الجلوبيولين، %الألبومين إلى الجلوبيولين، وظائف كل من الكبد و الكلى ونسبة الدهون في البلازما بين جميع المجاميع. كان نشاط أنزيم الكتاليز أعلى معنويًا في الدم في المجاميع المضاف إليها GCE إلى العليقة، بينما سجلت جميع المعاملات انخفاضاً معنويًا في قيمة (H_2O_2) عن المجموعة الكنترول. نستنتج أن إضافة ٤ مل من GCE /كجم إلى عليقة الأرانب النامية إلى تحسين أداء النمو، البيئة الميكروبية، خصائص الدم والكفاءة الاقتصادية دون وجود آثار ضارة على وظائف الكبد والكلى.