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Evaluating Zinc or Selenium Supplementation with Pomegranate Peel Extract in Rabbit Diet: Effects on Carcass Traits, Growth Performance, Antioxidant, and Immunological Responses

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Abstract

THIS study aimed to compare the single and combined effects of dietary supplementation of inorganic zinc (Zn) and selenium (Se) with pomegranate peel extract (PPE) on carcass characteristics, growth rate, antioxidative status, and immune responses of fattening rabbits. The trial lasted for 8 weeks, and the rabbits had an average weight of 587 ± 3.44 g at 5 weeks of age. The treatments included a control group (without supplementation), a PPE group with 200 mg PPE /kg diet, Zn and Se groups with diets fortified with 0.3 mg Se/kg diet and 100 mg Zn, respectively, a PPE+Zn group with both supplements and a PPE+Se group with both substances. The results demonstrate that the supplementation of Zn, Se, and PPE individually or in combination with the diets resulted in improved growth indices. Moreover, the FCR and protein efficiency ratio were enhanced by the supplementation. Slaughter traits and hematological parameters did not display significant alterations among the experimental groups. Serum ALT, AST, urea, total cholesterol, and LDL levels were reduced in the supplemented groups more than in the control group. Moreover, the supplemented groups revealed higher levels of serum immunoglobulin G and immunoglobulin A, along with a tendency towards increased glutathione peroxidase levels and decreased malondialdehyde levels. In conclusion, supplementing the diets of growing rabbits with Zn, Se, and PPE, either individually or in combination, has the potential to enhance growth performance, immune function, and overall health. Notably, no significant differences were observed between the effects of single and combination supplementation.

Keywords: Pomegranate extract, Trace minerals, Supplementation, Feed conversion, blood biochemical.

Introduction

Rabbits are recognized as a valuable livestock species that can serve as an affordable and high-quality source of animal protein for humans. Additionally, the production of rabbits has several advantages over other species, such as poultry, including minimal management costs, efficient utilisation of fibrous diets, and limited competition with people for grains [1]. Accordingly, there has been a notable global interest in developing effective nutritional strategies for rabbit production. These strategies focus on incorporating natural feed supplements that offer growth-enhancing benefits and promote the overall health of rabbits [2-4]. Recently, researchers have explored the use of trace mineral feed additives to enhance the rabbits productive performance [5-7]. Zinc (Zn) plays a crucial role in various biological processes in mammals, including rabbits [8]. It serves as an essential component for numerous enzymes, fulfilling both structural and catalytic functions in metalloenzymes [9]. Zinc, as a redox inactive trace element, holds significance as part of the antioxidant system. It regulates glutathione metabolism and promotes the synthesis of metallothionein, which aids in scavenging free radicals [10, 11]. Moreover, Zn supplementation contributes to a robust immune system, enabling rabbits to combat infections effectively [12]. Studies have indicated that incorporating Zn into the rabbit diet (at a dosage of 100 mg/kg diet) has shown improvements in growth indicators and feed conversion [13, 14].

Selenium (Se) is a critical trace element required for rabbit development and growth. It is essential for several physiological functions, including antioxidant protection, thyroid hormone uptake, and

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immunological function [15-17]. The Se occurrence in the active site of the glutathione peroxidase (GPx) enzyme present in the liver, blood, and other organs has been connected with improved immune responses in rabbits [18]. Supplementing the diet with Se has been shown to improve lipid oxidative stability, thereby contributing to the overall health and meat quality of animals [19]. However, previous studies on the effects of Se supplementation on rabbit performance have contradictory results. Dokoupilová *et al.* [20] described that the addition of Se did not considerably affect the growth rate, feed efficiency, or carcass yield of rabbits. On the contrary, *Zhang et al.* [21] found that Se supplementation enhanced growth, feed efficiency, and antioxidant capacity in growing rabbits. Additionally, Ayyat *et al.* [15] clarified that Se supplementation improved productivity and feed conversion in rabbits.

Pomegranate (*Punica granatum* L.) is a member of the *punicaceae* family widely cultivated in various regions, including Africa, Asia, and Europe [22]. During industrial processing of pomegranates into juice, by-products like the peel constitute a significant portion, accounting for 40 to 60% of the fruit's weight [23]. Pomegranate peel extract (PPE) is rich in polyphenolic compounds, particularly flavonoids, and exhibits notable antioxidant activity [24]. Previous studies have confirmed the presence of great amounts of polyphenolic compounds in PPE [25]. Additionally, the PPE's antimicrobial activities have the potential to combat gastrointestinal infections directly, thus reducing the diarrhea risk [25, 26]. Khan *et al.* [27] have identified the peel of pomegranate fruit as a promising source of natural antioxidants. The peels, along with their extract, serve as a rich source of hydrolysable tannins, including ellagitannins and gallotannins, as well as condensed tannins (proanthocyanidins), and various flavonoids like flavanols, flavonols, and anthocyanins [28-30]. Moreover, dietary supplementation with PPE has been shown to enhance growth, feed conversion, and nutrient digestibility in growing rabbits. It has also demonstrated antioxidant and antimicrobial effects [31].

To the best of our knowledge, there is a lack of prior research examining the collective impact of supplementing either Se or Zn with PPE as a growth promoter for growing rabbits in comparison to individual supplementation. In light of the benefits mentioned above, it was postulated that the combined use of PPE with either Zn or Se may enhance rabbit growth and overall health when compared to singular supplementation. Therefore, the present study aims to explore the potential benefits of supplementing Se or Zn individually or in combination with PPE. The effects on productive performance, antioxidative status, carcass characteristics, immune responses, and blood metabolites in growing male rabbits were investigated.

Methods

1. *Additives and diet Preparation*

Zinc oxide was produced by El-Nasr Pharmaceutical Chemicals Company (Gesr El Suez, Cairo, Egypt). Selenium selenite was commercially acquired from Chemajet Company (Borg El-Arab, Alexandria, Egypt). We purchased pomegranate fruit (*Punica granatum*) from a local market Egypt. Before being pulverised to pass through a 1 mm screen, the fruits were peeled and rinsed with fresh water. They were then dried in a forced oven at 50 °C for 72 hours. The components that had been dried were kept in sealed plastic containers with lids that fit tightly until they were needed. A litre of 70% ethanol was added to 100 g of pomegranate peel powder, and the mixture was left to soak for 72 hours. After stirring it for two hours, it was strained through Whatman filter paper. The last step was to use a rotary evaporator to evaporate the filtrate at 45°C in a vacuum, resulting in PPE. The tested trace minerals (Zn or Se) and PPE were combined with one kg of feed, homogenized to achieve the supplemental level, gradually mixed with the lasting diet, and pelleted as a total mixed diet. The requirements of growing rabbits were met or exceeded by the formulated experimental diets [32]. The ingredients and nutrient counts for the basic diet are presented in Table 1.

2. *Animals, experimental groups and management*

One hundred twenty New Zealand White (NZW) male rabbits (35 days old; 587 ± 3.44 g, mean ± SEM) were divided into 6 treatment groups contained ten replicate cages/ treatment (two animals/cage). The experimental treatments comprised: 1) Control Group: rabbits fed a diet without any supplementation. 2) PPE Group: rabbits received a 200 mg of PPE per kg diet. 3) Zn Group: rabbits fed a diet enriched with 100 mg of Zn per kg. 4) Se Group: rabbits fed a diet fortified with 0.3 mg of Se per kg. 5) PPE + Zn Group: rabbits a diet of both PPE (200 mg/kg) and Zn (100 mg/kg). 6) PPE + Se Group: rabbits fed a diet of both PPE (200 mg/kg) and Se (0.3 mg/kg). The experiment period was continued for 56 days. The dietary concentrations of the tested additives were based on earlier studies [13, 31, 33]. During the whole 56-day trial period, rabbits in all groups were raised under comparable environmental, hygienic, and management conditions. Rabbits were housed in galvanized wire cages that measured 60 cm in length, 35 cm in width, and 35 cm in height. Each cage had a manual feeder and an automated drinker. The rabbits were raised under natural light and temperature conditions in a well-ventilated building with temperature range of 17-22 °C and 10-hour light/14-hour dark photoperiod throughout the experiment. All rabbits were adapted

to the experimental circumstances for 7 days prior to the start of the trial and fed the basal diet. Throughout the entire trial period, pelleted feed and fresh water via nipple drinkers were available all the time. The animals' health was monitored on a daily basis for indicators of ear mites, diarrhea, scabies, respiratory issues, and mortality.

Growth performance indicators

Individual rabbits were weighed at the start of the study (initial weight) and after 8 weeks (final weight). Feed consumption was recorded daily for each replication. The average daily feed intake (ADFI), average daily gain (ADG), and relative growth rate (RGR) were calculated using the equations mentioned by Alagawany *et al.* [4]. The feed conversion ratio (FCR) was calculated as the ratio of the amount of ADFI (g) to ADG (g). There was no mortality in the rabbits during the experiment.

Carcass characteristics

At the end of the feeding trial, thirty rabbits (5 rabbits/group) that were close to the average weight of each treatment were selected, fasted for 12 h, weighted (preslaughter weight [PSW]), and then slaughtered within 2 h [34]. The rabbits were humanely slaughtered following the halal slaughtering method [35]. After complete bleeding, non-carcass components such as the distal part of the legs, skin, gastrointestinal tract, urinary bladder, and testicles were removed. After that, the internal organs (liver, lungs, kidneys, spleen, and heart) and hot carcass (HC) were immediately weighed. The carcasses were subsequently divided into the following parts: head, forepart, intermediate part, and hind part, as indicated by Blasco and Ouhayoun [34], and the weight of each carcass part was recorded. The prime cut was defined as the sum of the weights of the intermediate and hind sections. The carcass yield is determined as HC weight as a percentage of PSW.

Collection of samples

At the end of the feeding trial, blood samples were obtained from the same sacrificed rabbits, deposited in sterile tubes. The blood collected from each rabbit was separated into two subsamples; the first was collected in a K_3-EDTA tube to assess the hematological features. The second subsample was collected without any anticoagulant, allowed to clot at room temperature, and centrifuged for 20 minutes at 1000 ×g at 4 ºC. Next, the serum samples were separated and kept in sterile tubes at 20 °C until they were analyzed.

Hematological and biochemical estimations

White blood cell (WBC), red blood cell (RBC), hematocrit (Hct), hemoglobin (Hb), and platelet counts were among the hematological markers assessed in the whole blood samples in accordance with the methods described by Jain [36]. Then, the mean corpuscular volume (MCV), mean cell hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were calculated using the previously stated equations [19]. The levels of various lipoproteins in the serum, including total cholesterol (TC), high-density lipoprotein (HDL), low-density lipoprotein (LDL), and very low-density lipoprotein (VLDL), as well as ALT, AST, total protein, triglyceride (TG), creatinine, urea, albumin, were estimated colorimetrically by commercial kits (Diamond Diagnostic, Dokki, Giza, Egypt) in accordance with the guidelines provided by the manufacturer. Globulin levels determined by subtracting albumin from total protein.

Serum immune and antioxidant biomarkers

The serum samples were analyzed to evaluate the level of immunoglobulin G (IgG), immunoglobulin A (IgA), and lysozyme activity utilizing commercial ELISA kits following the manufacturer's directions. The activities of superoxide dismutase (SOD), glutathione peroxidase (GPx), and malondialdehyde (MDA) were evaluated as a biomarker of antioxidant status by commercial kits (Bio Diagnostic Company, Giza, Egypt) in line with the manufacturer's directions.

3. Data analysis

Data analysis was done using GLM procedure of SPSS statistics software (version 21; Chicago, IL, USA), with dietary trace minerals and PPE as fixed factors. Model (1) was used; $Y_{ij} = \mu + S_i + \varepsilon_{ij}$, where Y_{ijk} = the dependent variable, μ= the overall mean, Si = the fixed effect of supplementation, and ε_{ij} = the overall error term. The data of carcass characteristics were statistically analyzed considering PSW as a covariate variable based on Model (2) as follows: $Y_{ij} = \mu + S_i + b_{(X-x)} + \varepsilon_{ij}$, where Si, and ε_{ij} are defined in Model (1), and b(X−x) is the covariate variable. For evaluating the individual comparisons between the means, the post hoc Tukey's test was applied if the means were significantly different at $p < 0.05$. A statistical tendency was considered when the p-values were within the range of 0.05 to 0.10.

Results

Growth indices and slaughter traits

The results in Table 2 showed that, during the whole experiment, adding Zn, Se, and PPE individually or in combinations to the fattening NZW male rabbit diets increased ($P < 0.01$) the final body weight, ADG, and RGR. Similarly, ADFI and protein intake increased in all supplemented groups except for the PPE group, which exhibited no significant differences compared to the control group. In addition, FCR was enhanced ($P = 0.004$) by the individual or combined supplementations in rabbits' diet during the entire study period. Rabbits groups supplemented with Zn, PPE+ Zn, and PPE+ Se had

higher protein efficiency ratio values than the control group. Overall, the combination of Zn or Se with PPE exhibited a tendency towards increased growth indicators compared to individual supplementation; however, this increase was not statistically significant (Table 2).

Slaughter trait parameters are shown in Table 3. The covariance analysis showed no significant changes between the experimental groups concerning the weights of internal organs, such as the liver, kidneys, heart, lungs, spleen, and kidney fat. Additionally, there were no significant alterations detected in carcass yield or the weights of carcass parts (head, hind part, intermediate part, and fore part) among the experimental groups (Table 3).

Hematological and biochemical parameters

As shown in Table 4, all hematological parameters, including WBC, RBC, Hct, Hb, platelets, MCV, MCHC), and MCH, were not significantly different between the experimental groups. The ALT level in the serum of NZW rabbits was reduced ($P = 0.025$) in all supplemented groups compared to the control group. Additionally, the AST level in the serum of NZW rabbits in the PPE, Se, and PPE+ Se groups was significantly lower ($P = 0.012$) than that in the control group. However, serum concentrations of creatinine and protein profile did not display significant differences between the experimental groups. Serum urea concentrations were lower ($P = 0.019$) in rabbits treated with PPE, PPE+ Zn, and PPE+ Se compared to the control group. There was a tendency for an increase in the serum levels of total protein ($P = 0.064$) and albumin ($P = 0.10$) as Zn, Se, PPE, PPE+ Zn, and PPE+ Se were added to the rabbit diets (Table 4). Serum contents of total cholesterol and LDL were decreased ($P < 0.001$) by the individual or combined supplementation of Zn, Se, or PPE than the non-supplemented group (Fig. 1 and 2).

Immune and antioxidant biomarkers

As indicated in Figure 3, the IgG and IgA levels in the serum of NZW rabbits increased ($P = 0.001$ and 0.013, respectively) in all supplemented groups than the control group, while lysozyme activity remained unchanged ($P = 0.275$). Additionally, there was a trend towards an increase in GPx levels ($P = 0.065$) and a decrease in MDA levels ($P = 0.10$) in the serum of NZW rabbits in the supplemented groups than the control group (Figure 4). While the combination of Zn or Se with PPE did not exhibit significant differences in most assessed blood parameters compared to individual supplementation, there was a tendency for IgA to show a greater increase than with individual supplementation.

Discussion

Supplementing rabbits' diets with Zn or Se with PPE improved their growth performance, according to the current study's findings. This could be related to the increased ADFI and protein intake recorded in all supplemented groups except for the PPE group. Besides, FCR was significantly enhanced in rabbit's fed-supplemented diets. One possible explanation for this enhancement could be the crucial function of Se and Zn in the metabolism of carbohydrates, lipids, proteins, and energy [37, 38]. In addition, Zn is essential for the production of growth hormones, enzymes, and structural proteins [39, 40]. Furthermore, the documented role of Zn and Se in synthesizing growth hormone could be another possible reason [41, 42]. For PPE, growth-promoting activity could occur via different mechanisms as it contains great amounts of polyphenols comprising flavonoids (ellagitannins, gallotannins, anthocyanins, gallagyl esters, hydroxycinnamic acids, and dihydroflavonol, hydroxybenzoic acids) and hydrolyzable tannins (gallic acid and ellagic acid) [43]. The bioactives mentioned above can protect the intestinal mucosa from oxidative damage and pathogens while also limiting peristaltic activity in digestive diseases, hence avoiding diarrhoea [44]. In this regard, PPE increased beneficial bacteria counts in the ileal and cecal contents of broiler chickens while decreasing the abundance of pathogenic *Escherichia coli* in the aforementioned sections [45]. In a recent study, Imbabi *et al.* [46] found that supplementing heat-stressed V-line rabbits' diets with whole pomegranate extract (1000 – 1500 mg/kg) enhanced growth performance and FCR. Moreover, Nassrallah *et al*. [47] reported that PPE dietary supplementation improved FCR in growing rabbits because of enhancements in the nutrient digestibility and the antioxidant activity of PPE phenolic compounds.

Hematological and biochemical parameters can yield information on the safety of dietary supplementation in rabbits [48]. In the existing study, it was evident that all hematological parameters, including WBC, RBC, Hct, Hb, platelets, MCV, MCHC, and MCH were not significantly differed with dietary supplementation of Zn or Se with PPE. Similarly, non-significant changes in blood pictures were recorded following Se supplementation in rabbits [15]. Also, hematologic parameters were not affected by PPE addition to broilers diets [49]. Besides, dietary supplementation of Zn or Se with PPE decreased serum ALT levels and PPE, Se, and PPE+ Se groups showed reduced AST levels, suggesting the hepato-protective effect of supplementation. Moreover, rabbit-fed PPE+Zn and PPE+ Se supplemented diets showed reduced serum urea concentrations, reflecting the enhanced renal function. AST and ALT are intracellular enzymes whose serum levels are commonly used as a hepatocellular damage screening test whereas urea and creatinine reflect renal function in rabbits [6]. Similarly, the levels of AST and ALT in the control rabbit serum were much higher ($P < 0.05$) than in the

group fed basal diet +1250 or 1500 mg/kg of whole pomegranate extract [46]. The hepatorenal protective effects of Se and Zn could be mainly related to their antioxidant activity [15, 50]. The anti-inflammatory and antioxidant activities of different solvent extracts of PPE have been earlier recognized in the studies of Ismail *et al*. [43] and Imbabi *et al*. [46].

In this study, a significant reduction in the serum total cholesterol and LDL were recorded in rabbits fed diets fortified with Zn, Se, and/or PPE. One possible mechanism by which the PPE improved plasma lipid profiles was by lowering cholesterol absorption and increasing faecal excretion and by inhibiting the activities of two important cholesterol metabolism enzymes, 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase and sterol O-acyltransferase [51]. An explanation for selenium's anti-cholesteremic effects may lie in its ability to reduce free fatty acid release from adipose tissue and inhibit lipolysis in this tissue [52]. The hypothesized molecular mechanisms that contribute to the decrease in serum lipid levels after zinc supplementation include an increase in insulin secretion and an inhibition of adipose tissue lipolysis, a decrease in the release of free fatty acids into the bloodstream and their availability to the liver, and an increase in the synthesis of excessive lipoproteins [53, 54]. Furthermore, zinc has a direct effect on lipid metabolism [55].

There was a trend towards an increase in the levels of GPx and a decrease in MDA in the serum of NZW rabbits in the Zn, Se, and PPE-supplemented groups. Research has shown that PPE and its solvent extracts have antioxidant properties [43]. Se can be found in rabbits and other animals as a cofactor of various enzyme complexes, including SOD, GPx [56], and thioredoxin reductase, an important enzyme in thyroid hormone metabolism [57], and taking Se supplements can protect cells from radical damage [58]. An increase in the total concentration of sulfhydryl groups in the body and a subsequent decrease in lipid peroxidation may be caused by Zn inducing the synthesis of proteins rich in sulfhydryl groups, such as glutathione (the principal intracellular non-protein thiol) and metallothionein (cysteine-rich protein) [11]. And because of its direct binding to the sulfhydryl, Zn can prevent oxidation of protein sulfhydryl groups and decrease their reactivity [59]. In our investigation, we observed greater GPx activity, which might be explained by the fact that Zn is directly engaged in glutathione de novo production by controlling the expression of glutamate-cysteine ligase [11].

The immune status is an important indicator of the overall health of rabbits [48]. Based on the current study findings, the IgG and IgA levels in the serum of NZW rabbits increased in all supplemented groups compared to the control group. Additionally, there was a tendency for an increase in the serum levels of total protein and albumin as Zn, Se, PPE, PPE+ Zn, and PPE+ Se were added to the rabbit diets reflecting enhanced immune function. Among the many cellular and molecular roles played by Se and Zn are the following: modulation of cell signalling molecules (nuclear factor kappa B), suppression of immunosuppressive glucocorticoids, reduction of intramastitis rates and duration, and regulation of neutrophil, lymphocyte, and natural killer cell function through activation of interleukin-2, ultimately leading to an enhancement of cell-mediated and humoral immune responses [12]. Similar immune-enhancing activity has been previously documented with PPE addition to broiler diets in the study of saleh [49]. There is evidence that PPE and its solvent extracts have antiviral, antibacterial, and immune-boosting properties [43]. By modulating cytokine production, enhancing intestinal microbial flora, and removing free radicals produced during metabolic processes, polyphenols from PPE may enhance the immunological response of broiler chickens [60, 61]. Consumption of feeds and foods high in polyphenols causes a buildup of polyphenol metabolites in the colon, which exhibit prebiotic characteristics [60].

Conclusion

In conclusion, the study demonstrates that supplementing with Zn or Se individually, as well as in combination with PPE, improves growth indicators, feed conversion, protein efficiency, liver and kidney functions, blood cholesterol levels, and immunity. The slight advantages observed with the combined supplementation compared to individual supplementation suggest that a single supplementation approach is recommended under normal conditions. Future research should explore the combined effects of Zn or Se with plant extracts under stress conditions, focusing on overall health, reproductive performance, and meat quality in rabbits.

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Declaration of Conflict of Interest

The authors declare that there is no conflict of interest.

Ethical Approval

The experimental protocols applied in this study were performed in accordance with the guidelines for animal welfare of the Animal Production Department of XXXX University, Egypt, and were approved by Ethics Committee of the Local Experimental Animals Care with the assigned approval number ZU-IACU/2/F/67/2019.

Author`s contribution

Conceptualization, M.S.A., U.M.A., and A.A.A.; Data curation, M.S.A., D.M.F., A.A.A., and S.A.E.; Formal analysis, D.M.F. and M.F.A.E.; Investigation, M.S.A., U.M.A., D.M.F., S.A.E. and A.A.A.,; Methodology, D.M.F., S.A.E., and M.F.A.E.; Resources, D.M.F., U.M.A., and M.S.A.; Software, M.S.A. and S.A.E.; Supervision, M.S.A., U.M.A., and A.A.A.; Validation, M.S.A. and U.M.A.; Writing – original draft, M.S.A., A.A.A., and D.M.F. All authors have read and agreed to the published version of the manuscript.

TABLE 1. Ingredients and composition of the basal diet.

Components	[%]	Nutrient levels	[%]
Soybean meal	17	**Chemical analysis**[2]	
Lucerne meal	27	Crude protein (N × 6.25)	16.83
Wheat bran	25	Ether extract	3.68
Yellow corn	21	Ash	9.45
Wheat straw	8	Neutral detergent fiber	39.08
Limestone	1	Acid detergent fiber	19.81
Sodium chloride	0.5	**Calculated values**[3]	
DL-methionine	0.1	Digestible crude protein	12.38
Premix [1]	0.4	Digestible fiber	18.68
Total	100	Lysine	0.84
		Methionine + cystine	0.63
		Digestible energy, MJ/kg	10.80

[1] Premix provided the following vitamins and minerals per kilogram of diet: vitamin A, 8000 IU; vitamin D_3, 600 IU; vitamin E, 34 mg; vitamin K_3, 1.32 mg; vitamin B_1, 1.32; vitamin B_2, 4.0 mg; vitamin B_6, 1.32 mg; vitamin B_{12}, 0.01 mg; pantothenic acid, 13.32 mg; biotin, 0.13 mg; folic acid, 3.32 mg; choline chloride, 800 mg; manganese 32 mg; zinc 60 mg; iron 120 mg; copper 16 mg; iodine 2 mg; selenium 0.4 mg; and cobalt 0.4 mg.

[2] According to AOAC [62].

[3] Calculated according to tables of ingredients of Maertens *et al*. [63].

TABLE 2. Effects of dietary supplementation on growth performance parameters of growing rabbit.

Parameter	Experimental groups[1]						P value
	Control	Zn	Se	PPE	PPE+ Zn	PPE+ Se	
Initial weight, g	596.7±5.77	580±10.22	596±5.21	578±11.33	589±6.57	584±9.21	0.504
Final weight, g	1878.9 ^{c} ±31.50	2137.9 ^{ab} ±42.39	2062.5 ^{ab} ±24.71	2020.6 ^{b} ±45.83	2123.5 ^{ab} ±55.96	2157.9 ^{a} ±47.54	<0.001
Daily weight gain, g	22.9 ^{b} ±0.58	27.82 ^{a} ±0.74	26.19 ^{a} ±0.49	25.76 ^{a} 0.95	27.4 ^{a} ±1.09	28.11 ^{a} ±0.89	0.001
Relative growth rate, %	103.48 ^{b} ±1.46	114.52 ^{a} ±1.62	110.25 ^{a} ±1.19	110.76 ^{a} ±2.55	112.77 ^{a} ±2.31	114.6 ^{a} ±1.93	0.002
Daily feed intake (g)	91.07 ^{c} ±1.84	99.71 ^{a} ±1.76	97.14 ^{ab} ±1.26	94.57 ^{bc} ±2.10	96.2 ^{ab} ±1.31	96.39 ^{ab} ±0.98	0.014
Protein intake, g/day	15.33 ^{c} ±0.31	16.78 ^{a} ±0.30	16.35 ^{ab} ±0.21	15.95 ^{bc} ±0.35	16.19 ^{ab} ±0.22	16.22 ^{ab} ±0.17	0.014
Feed conversion ratio	4.00 ^{a} ±0.14	3.60 ^{b} ±0.07	3.71 ^{b} ±0.05	3.69 ^{b} ±0.08	3.55 ^{b} ±0.11	3.46 ^{b} ±0.09	0.004
Protein efficiency ratio	1.50 ^{b} ±0.06	1.66 ^{a} ±0.03	1.60 ^{ab} ±0.02	1.62 ^{ab} ±0.04	1.69 ^{a} ±0.06	1.73 ^{a} ±0.05	0.010

Means in the same row with different superscripts (a, b, c) are significantly different ($P < 0.05$).

[1] The experimental groups consisted of a control group with no supplementation, Zn: 50 mg Zn /kg diet, Se: 0.3 mg Se/kg diet, PPE: 200mg PPE/ kg diet, PPE+Zn: 200mg PPE+50 mg Zn/ kg diet, and PPE+Se: 200 mg PPE + 0.3 mg Se/kg diet.

Protein intake = feed intake × dietary crude protein%.

Protein efficiency ratio = weight gain /protein intake.

TABLE 3. Effects of dietary supplementation on slaughter traits of fattening rabbit.

Parameter	Experimental groups[1]						P value[2]
	Control	Zn	Se	PPE	PPE+ Zn	PPE+ Se	
Organs weight, g							
Liver	55.90±3.69	78.73±10.97	62.70±4.43	62.45±1.66	62.09±2.05	73.89±3.62	0.188
Kidneys	11.89±0.13	15.26±0.49	14.07±0.49	12.85±0.84	14.84±0.96	14.16±0.23	0.093
Heart	5.65±0.57	5.90±0.27	6.68±0.45	6.68±0.45	7.73±1.19	7.63±0.76	0.192
Lungs	11.74±0.40	15.23±1.39	12.97±0.79	13.28±1.02	13.19±1.09	16.02±1.42	0.241
Spleen	0.79±0.10	1.20±0.17	1.39±0.39	0.99±0.12	1.05±0.13	1.45±0.34	0.469
Carcass parts, g							
Head	119.5±1.32	116.5±4.94	113.0±2.35	120.0±1.41	121.8±3.92	118.0±3.24	0.138
Fore part	426.5±12.72	467.0±27.73	428.0±16.11	448.5±14.98	472.0±26.25	457.0±22.09	0.832
Intermediate part	205.8±4.55	241.0±9.28	241.8±16.39	200.8±5.36	228.3±15.76	218.5±9.22	0.082
Hind part	418.8±8.61	450.8±9.39	414.8±13.58	438.8±24.33	465.0±19.54	454.0±14.51	0.383
Carcass yield, %	56.84±0.42	56.82±0.49	57.10±1.66	57.76±1.05	58.39±0.67	57.05±0.65	0.782
Kidney fat, g	20.22±5.87	23.33±1.86	23.46±4.32	23.78±4.27	26.71±5.84	22.31±5.43	0.992

[1] The experimental groups consisted of a control group with no supplementation, Zn: 50 mg Zn /kg diet, Se: 0.3 mg Se/kg diet, PPE: 200mg PPE/ kg diet, PPE+Zn: 200mg PPE+50 mg Zn/ kg diet, and PPE+Se: 200 mg PPE + 0.3 mg Se/kg diet.
[2] To exclude the pre-slaughter weight effect, the data of organs weight and carcass parts were statistically analyzed by covariance analysis. Hence the measurements could be compared between groups on carcasses of the same weight

TABLE 4. Effects of dietary supplementation on hematological and biochemical parameters of growing rabbit.

Parameter	Experimental groups[1]						P value
	Control	Zn	Se	PPE	PPE+ Zn	PPE+ Se	
RBCs ($\times10^6$/ml)	4.7 ±0.49	5.54±0.39	5.65±0.36	5.62±0.31	5.36±0.19	5.59±0.33	0.427
Hemoglobin (g/dl)	14.42±0.71	16.88±0.85	16.93±0.88	17.6 ±0.97	16.71±0.63	16.36±0.74	0.159
Hematocrit (%)	38.78±1.33	40.84±1.83	38.87±2.13	40.77±2.12	38.7±1.22	39.53±1.91	0.904
MCV(fL)	84.44±6.65	79.07±6.02	78.97±8.38	81.92±6.78	80.03±2.47	74.75±3.25	0.908
MCH (pg/dl)	31.13 ±1.56	30.3 ±1.01	30.85±0.95	32.61±0.57	31.24±0.38	30.92±1.07	0.697
MCHC (g/dl)	37.1±1.00	38.68±1.57	39.91±2.66	40.39±2.34	39.17±1.51	41.42±0.69	0.620
Platelets ($\times10^3$/ml)	136.8±8.90	154.5±4.21	148.8±8.58	144.3±25.06	153.0±9.91	163.3±3.79	0.739
WBCs ($\times10^3$/ml)	8.41±0.65	8.53±0.20	9.02±0.27	9.97±0.73	9.05±0.12	9.1±0.59	0.320
ALT (u/l)	43.33 ^{a} ±4.09	34.1 ^{b} ±1.02	29.1 ^{b} ±3.20	31.58 ^{b} ±3.00	28.08 ^{b} ±1.08	30.42 ^{b} ±3.17	0.025
AST (u/l)	30.83^{a} ±1.75	27.21 ^{ab} ±3.23	22.82 ^{bc} ±0.18	21.48 ^{bc} ±2.00	27.14 ^{ab} ±1.03	21.17 ^{c} ±0.55	0.012
Creatinine (mg/dl)	0.73±0.10	0.63±0.11	0.40±0.01	0.49±0.10	0.51±0.05	0.48±0.08	0.145
Urea (mg/dl)	19.35 ^{a} ±1.49	16.92 ^{ab} ±1.25	16.79 ^{ab} ±1.18	12.67 ^{b} ±0.79	13.07 ^{b} ±2.05	12.98 ^{b} ±0.96	0.019
TP (mg/dl)	5.93±0.49	7.57±0.05	7.62±0.36	6.35±0.55	6.98±0.23	7.21±0.50	0.064
Albumin (mg/dl)	3.53±0.33	4.34±0.12	4.35±0.11	3.88±0.31	3.99±0.06	4.12±0.10	0.103
Globulin (g/d)	2.4±0.17	3.23±0.07	3.27±0.25	2.47±0.24	2.99±0.22	3.1±0.43	0.115

Means in the same row with different superscripts (a, b, c) are significantly different ($P < 0.05$). RBC, red blood cells; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; WBC, white blood cell; ALT, alanine aminotransferase; AST, aspartate aminotransferase; TP, total protein.
The experimental groups consisted of a control group with no supplementation, Zn: 50 mg Zn /kg diet, Se: 0.3 mg Se/kg diet, PPE: 200mg PPE/ kg diet, PPE+Zn: 200mg PPE+50 mg Zn/ kg diet, and PPE+Se: 200 mg PPE + 0.3 mg Se/kg diet.mg PPE + 0.3 mg Se/kg diet.

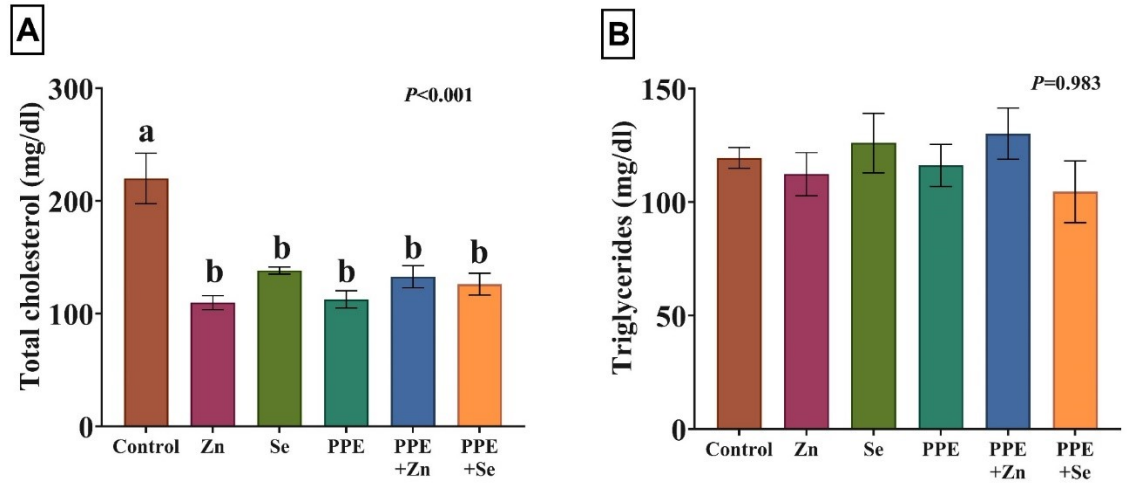

Fig. 1. Effects of dietary supplementation on serum concentrations of total cholesterol (A) and triglycerides (B). Different letters above the error bar indicate significant differences at P < 0.05. Values are expressed as means ± standard error. The experimental groups consisted of a control group with no supplementation, Zn: 50 mg Zn /kg diet, Se: 0.3 mg Se/kg diet, PPE: 200mg PPE/ kg diet, PPE+Zn: 200mg PPE+50 mg Zn/ kg diet, and PPE+Se: 200 mg PPE + 0.3 mg Se/kg diet.

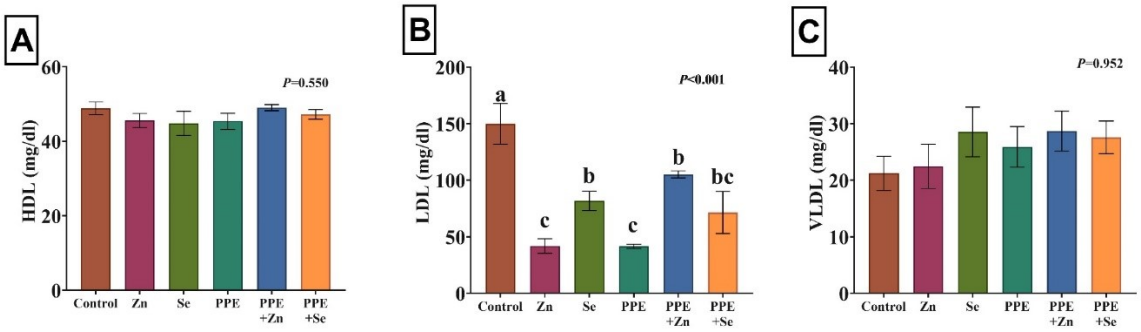

Fig. 2. Effects of dietary supplementation on serum concentrations of high density lipoprotein (HDL; A), low density lipoprotein (LDL; B), and very low-density lipoprotein (VLDL; C). Different letters above the error bar indicate significant differences at P < 0.05. Values are expressed as means ± standard error. The experimental groups consisted of a control group with no supplementation, Zn: 50 mg Zn /kg diet, Se: 0.3 mg Se/kg diet, PPE: 200mg PPE/ kg diet, PPE+Zn: 200mg PPE+50 mg Zn/ kg diet, and PPE+Se: 200 mg PPE + 0.3 mg Se/kg diet.

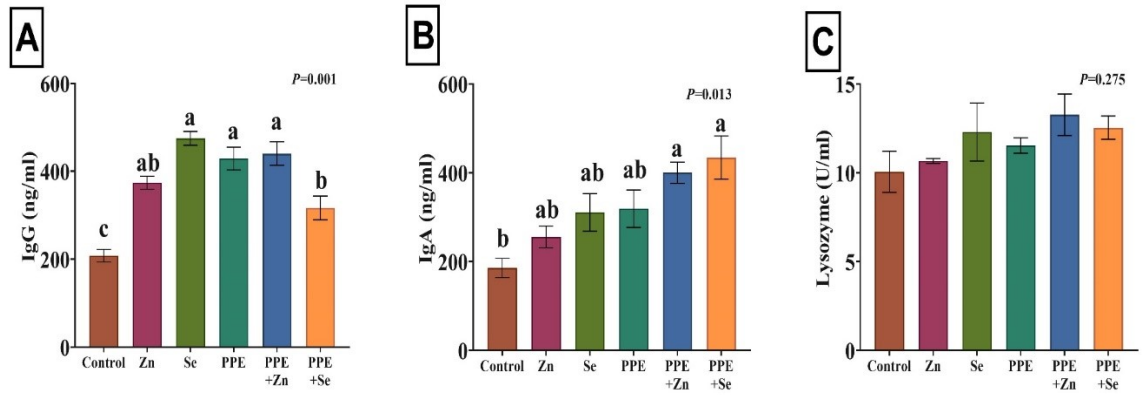

Fig. 3. Effects of dietary supplementation on serum concentrations of immunoglobulin G (IgG; A), immunoglobulin A (IgA; B), and lysozyme (C). Different letters above the error bar indicate significant differences at P < 0.05. Values are expressed as means ± standard error. The experimental groups consisted of a control group with no supplementation, Zn: 50 mg Zn /kg diet, Se: 0.3 mg Se/kg diet, PPE: 200mg PPE/ kg diet, PPE+Zn: 200mg PPE+50 mg Zn/ kg diet, and PPE+Se: 200 mg PPE + 0.3 mg Se/kg diet.

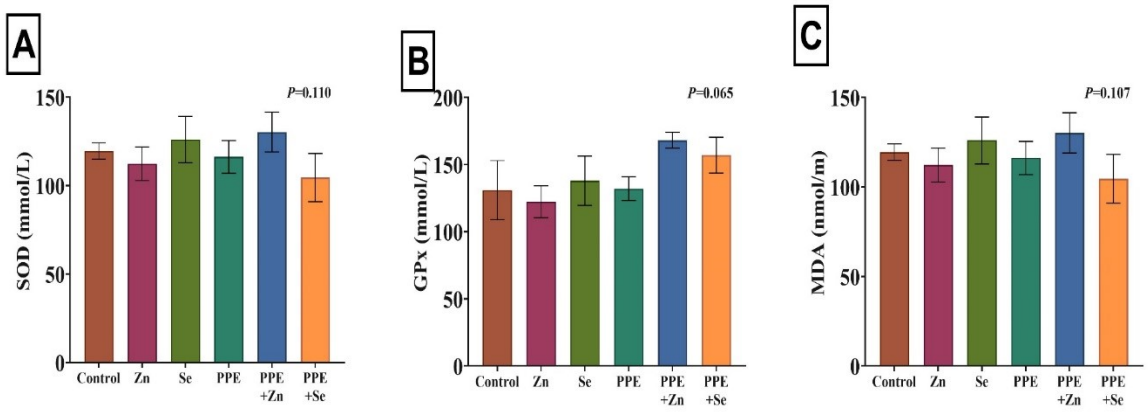

Fig. 4. Effects of dietary supplementation on serum concentrations of superoxide dismutase (SOD; A), Glutathione peroxidase (GPx; B), and Malondialdehyde (MDA; C). Different letters above the error bar indicate significant differences at P < 0.05. Values are expressed as means ± standard error. The experimental groups consisted of a control group with no supplementation, Zn: 50 mg Zn /kg diet, Se: 0.3 mg Se/kg diet, PPE: 200mg PPE/ kg diet, PPE+Zn: 200mg PPE+50 mg Zn/ kg diet, and PPE+Se: 200 mg PPE + 0.3 mg Se/kg diet.

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تقييم إضافة الزنك أو السيلينيوم مع مستخلص قشر الرمان في علائق الأرانب: تأثيرات على صفات الذبيحة وأداء النمو والاستجابات المضادة للأكسدة والمناعية

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الملخص

إجريت هذه الدراسة بهدف مقارنة تأثير إضافة الزنك غير العضوي والسيلينيوم الى علائق التسمين بشكل فردى أو ممزوج مع مستخلص قشر الرمان على خصائص الذبيحة ومعدل النمو والحالة المضادة للأكسدة والاستجابات المناعية. استمرت التجربة لمدة 8 أسابيع، وكان متوسط وزن الأرانب فى بداية التجربة 587 ± 3.44 جم وعمر 5 أسابيع. شملت المعاملات مجموعة كنترول (بدون إضافات)، ومجموعة بإضافة مستخلص قشر الرمان بمعدل 200 ملغم/ كجم علف، مجموعة السلينيوم مدعمة بـ 0.3 ملغم سلينيوم/ كجم علف ومجموعة الزنك مدعمة بـ 100 ملغم الزنك/ كجم علف، ومجموعة مخلوط مستخلص قشر الرمان + الزنك. ومجموعة مستخلص قشر الرمان + السلينيوم التي تحتوي على نفس الجرعات من كلتا المادتين.أظهرت النتائج أن إضافة الزنك والسيلينيوم ومستخلص قشر الرمان الى العلائق بشكل فردي أو خليطة أدت إلى تحسن مؤشرات النمو. وعلاوة على ذلك، تم تحسن معامل تحويل العلف وكفاءة الاستفادة من البروتين مع كل الاضافات الغذائية. لم تظهر صفات الذبيحة و قياسات صورة الدم أيه إختلافات معنوية بين المجموعات التجريبية. إنخفضت مستويات انزيم الاسبرتيت أمينو ترانزفيريز، الألانين أمينو ترانزفيريز واليوريا والكوليسترول الكلي ومستويات الكوليسترول منخفض الكثافة مع كل الاضافات الغذائية بالمقارنة مع مجموعة الكنترول. علاوة على ذلك، أظهرت المجموعات المكملة زيادة معنوية فى الجلوبيولين المناعي G والجلوبيولين المناعي A، إلى جانب الميل نحو زيادة مستويات بيروكسيداز الجلوتاثيون وانخفاض مستويات المالونديالدهيد. وإجمالياً، فإن إضافة الزنك والسيلينيوم ومستخلص قشر الرمان إلى علائق الأرانب النامية ، سواء بشكل فردي أو مخلوطة، أدى إلى تعزيز أداء النمو ووظيفة المناعة والصحة العامة. والجدير بالذكر أنه لم يلاحظ أي فروق ذات دلالة إحصائية بين الإضافات الفردية والمخلوطة.

الكلمات الدالة: مستخلص الرمان؛ المعادن النادرة؛ المكملات؛ تحويل العلف؛ النمو؛ كيمياء الدم.