

**IMPACT OF SUPPLEMENTAL INORGANIC AND NANO PARTICULATE SOURCES OF SELENIUM ON GROWTH INDICATORS, DIGESTIBILITY, CARCASS TRAITS, AND IMMUNOLOGICAL STATUS OF GROWING RABBITS**

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**ABSTRACT:**

*The aim of the present study was to determine the effect of supplemental two sources of selenium; inorganic and nano (Se-NP), on growth, nutrients digestibility, immunity, and carcass traits of New Zealand White (NZW) rabbits. A total of 45 male rabbits (5 weeks old) at average body weight of 582.46±7.52g were divided into three groups (15 rabbits/group) fed the following diet group: 1) the basal diet as control diet, 2) the basal diet+sodium selenite (0.3 mg Se/kg diet), and 3) the basal diet+ Se-NP (0.3 mg Se/kg diet), during period from 5 to 13 weeks of age.*

*The findings showed that final body weight, relative*

*growth rate, daily weight gain, and daily feed intake were significantly ( $P<0.05$ ) higher in the rabbit fed diets supplemented with organic Se and Se-NP than those of the control group. Se-NP supplementation had improved feed conversion ratio, nutrients digestibility, nutritive values, and hemoglobin concentration in comparison with other groups. Additionally, the highest values of profit margin were observed in the rabbits fed diets supplemented with Se-NP. There were no histological abnormalities in the architecture of liver and kidney tissue in the experimental groups, indicating that the tested doses of Se were safe.*

***In conclusion***, supplementing fattening rabbit diets with Se-NP

*was significantly better than effect on architecture of liver and inorganic selenium in enhancing kidney so it could be used safely. feed utilization, growth performance, nutrients digestibility and economic efficiency without any negative*

**Keywords:** *Selenium sources; Rabbits; Growth Indicators; Digestibility, carcass traits, immunological status.*

## INTRODUCTION

Rabbit production is now facing challenges due to global warming, rising costs of diet ingredients, structural weaknesses, and progressive and consistent decreases in product demand. Numerous favorable characteristics of the rabbit lead it to be hypothetically considered as the ideal meat-producing animal in developing nations. For instance, it has a relatively short life cycle, a rapid gestational period, is exceptionally prolific, and has a feed conversion potential (Lebas *et al.*, 1997). Egypt is the third largest rabbit meat producer worldwide. The production capacity of rabbit meat reached 71,178 thousand tons (FAOSTAT, 2022).

Selenium is a trace mineral that is mandatory for animals and plays a role in a number of vital metabolic processes, including the reduction of oxidative stress, proper thyroid gland activity, maintaining the immune systems, cellular redox status, detoxifying of pollutants, calcium homeostasis in cardiac and skeletal muscle metabolism, and lipid metabolism. The inorganic selenium compound is currently authorized as a nutritional feed additive in the EU and was previously used in livestock nutrition for decades as a source of the essential trace element selenium (EFSA, 2016). Selenium is a vitally significant microelement; according to modern ideas, its biological role is primarily determined by antioxidant, immune-modulating, antiviral properties (Syvyk *et al.*, 2018). Selenium is essential for synthesis of steroids, cellular respiration, coenzyme A production, and oxidation of fatty acids. Its lack in animal rations negatively affects their health and economic indicators (Surai, 2006).

Numerous nutritional approaches have been developed in livestock production for preventing rabbit production reduction (Albonetti *et al.*, 2017). For example, antioxidants including selenium have been investigated for preventing lipid oxidation (Dokoupilová *et al.*, 2007 and Matic *et al.*, 2017). Even though the antioxidant activity of selenium remains unclear (Papadomichelakis *et al.*, 2017) the its impact, either inorganic or nano particles, as a dietary supplement on nutrient digestibility, health status, and carcass quality are inconclusive.

Therefore, the aim of the current investigation was to examine the effect of dietary inorganic or nano selenium on growth indicators, digestibility, carcass traits, and immunological status of growing rabbits

## MATERIALS AND METHODS

### *Animals, treatments and management*

This research was carried out at Zagzig University, specifically in the Faculty of Agriculture's Rabbitry Farm. A total of 45 male New Zealand White (NZW) rabbits, 35-days old, with an average body weight of  $582.46 \pm 7.52$  g were used in this experiment. Rabbits were allocated randomly into three groups (15 rabbits in each). The experimental groups consisted of i) basal diet served as a control group, ii) the basal diet + sodium selenite (0.3 mg Se/kg diet), iii) the basal diet+ Se-NP (0.3 mg Se/kg diet), The study lasted 8 weeks from the 5th to the 13th week of the rabbits' age. Sodium selenite was bought from Chemajet Company (Alexandria, Egypt). Selenium nano particles were produced by NAQAA Foundation for Nanotechnology (Giza, Egypt). The chemical reduction of sodium selenite with ascorbic acid was used to synthesize Se-NP powder according to the modified methods described by Malhotra *et al.* (2014). The x-ray diffraction and scanning electron microscope indicated that most of the Se-NP has a size of 60 nm on average.

Animals were individually housed in a galvanized wire cage ( $35 \times 60 \times 35$  cm) in a well-ventilated building. Rabbits were fed pelleted diets *ad libitum* and freshwater was offered all-time through an automatic nipple. The photoperiod was 11:13 h light-dark cycle with a semi-continuous lighting program. Throughout the trial period, all animals were maintained with the same standards of cleanliness, healthcare, and environment.

The basal diet was prepared and pelleted to cover all nutrient needs reported by De Blas and Mateos (2010) for growing rabbits as shown in Table 1.

### *Growth performance and slaughter traits*

Throughout the study, rabbit weight was measured using a digital balance with a precision of 1g at the beginning of the study and every four weeks. The difference between supplied and un supplied feed for each animal for a period of 24 h was used to compute the daily feed intake (DFI).

**Table 1.** Formulation and composition of the basal diet.

Items	%
<b>Ingredients:</b>	
Alfalfa hay	27.0
Soybean meal (44%)	17.0
Wheat bran	25.0
Yellow corn	21.0
Wheat straw	8.0
Limestone	1.0
Sodium chloride	0.5
DL-methionine	0.1
Vitamin and mineral premix*	0.4
<b>Total</b>	<b>100</b>
<b>Chemical analysis of diets** :</b>	
Crude protein	16.83
Crude fiber	14.06
Ether extract	3.68
Ash	9.45
Digestible energy, kcal/kg ***	2587

\* **Each kilogram of premix contains:** Vit. A, 20000 IU; Vit.D3, 15000 IU; Vit.E, 8.33 g; Vit.K, 0.33 g; Vit.B1, 0.33; Vit.B2, 1.0 g; Vit.B6, 0.33 g; Vit.B5, 8.33 g; Vit. B12, 1.7 mg; Pantothenic acid, 3.33 g; Biotine, 33 mg; Folic acid, 0.83 g; Choline chloride, 200; Manganese 80 g; Zinc 60 g; Iron 30 g; Copper 4 g; Iodine 0.5 g; Selenium 0.1 g; and Cobalt 0.1 g

\*\* Analyzed according to AOAC (1980).

\*\*\* Calculated according to NRC (1977).

The following formulae were used to calculate the growth indicators and feed utilization: Average daily gain (ADG) =  $(FBW - IBW) / \text{trial duration (days)}$ ; Relative growth rate (RGR) =  $[(FBW - IBW) / \frac{1}{2}(IBW + FBW)] \times 100$ ; Feed conversion ratio (FCR) =  $DFI / ADG$ . Where FBW indicates the final body weight and IBW denotes the initial body weight.

At the end of the trial (13 weeks of age), five rabbits/ group were randomly selected, weighed and slaughtered to assess slaughter characteristics. After complete bleeding, carcasses were skinned and the tail viscera were removed. Carcass parts including head, fore part, intermediate part, and hind part were weighed. The carcass weight percentage was determined as the actual carcass weight without giblets.

***Digestibility trial***

At the termination of the study (13 weeks of age), five animals were chosen at random from each treatment for the digestibility assessment. Individual rabbits were housed in metabolic cages (35 × 60 × 35 cm) that facilitated the separation of urine and feces. The experimental diets and water were provided *ad libitum*. The total fecal output and feed intake were recorded every 24 h for five successive days for each rabbit according to Perez et al. (1995). Feces of each animal were dried at 65 °C for 48 h, ground to 1-mm screen and kept until analysis. Feces and Feed samples were analyzed for organic matter (OM), ether extract (EE), dry matter (DM), crude protein (CP), and crude fiber (CF) following the methods of AOAC (2006) and total digestible nutrients (TDN) was calculated following the formula: :

$$\text{TDN} = \% \text{ digestible CF} + \% \text{ digestible CP} + (\% \text{ digestible EE} \times 2.25) + \% \text{ digestible nitrogen free extract.}$$

According to McNitt *et al* (2013), the digestible energy (DE) content was determined to be 4 kilocalories / gram for digested protein and carbs, whereas the digestible energy content of digested fat was found to be 9 kilocalories / gram.

***Blood and histological evaluations***

Blood samples (13 weeks of age), were collected from five rabbits / investigational group from the marginal ear vein. Blood was obtained in EDTA (1 mg/ml) as an anticoagulant for hematological investigations. In order to assess immune response, blood was drawn into anticoagulant-free, sterile tubes. Then blood was left to clot and centrifuged at 1075rpm for 10 min for the serum collection, and then placed in sterilized tubes at -20 °C until analyzed. For hematological assessment, whole blood samples were subjected to measurement of hemoglobin (Hb), red blood cells (RBC), white blood cells (WBC), lymphocytes, hematocrit (Hct), eosinophils, neutrophils, and monocytes according to Grindem (2011) using an automatic hematology analyzer (Hospitex Diagnostics, Italy). Using commercial ELISA assays, the level of complement component 3 (C3), Immunoglobulin M (IgM), and lysozyme activity were determined according the manufacturer's instructions.

Liver and kidney specimens from the same slaughtered rabbit were collected in neutral buffered formalin. The collected tissues were then dehydrated overnight, cleared in xylene, and embedded in paraffin blocks. Serial sections of 5 µm thickness were cut using a microtome (Leica RM 2155, England), stained with hematoxylin and eosin, and mounted on a glass slides to be viewed by the light microscopy for histological examination according to Suvarna *et al.* (2018).

***Statistical analysis***

The collected data were statistically analyzed by SPSS (version 21; Chicago, IL, USA) using one-way ANOVA (analysis of variance) according to Snedecor and Cochran (1982). The differences between the groups were investigated with the application of Duncan's multiple-range test. Differences between means were considered statistically significant at P- value <0.05.using Duncan's range test (Duncan, 1955).

The data of growth performance, blood parameters, and digestibility were statistically analyzed according to model1:

$$Y_{ij} = \mu + S_i + e_{ij},$$

Where  $Y_{ij}$  = An observation,  $\mu$  = Overall means,  $S_i$  = Fixed effect of  $j^{\text{th}}$  selenium source ( $i = 1 \dots 3$ ), and  $e_{ij}$  = Random error.

The carcass characteristic data were statistically examined based on model 2:

$$Y_{ijk} = \mu + S_i + b(X-x) + e_{ijk},$$

where  $Y_{ij}$ ,  $\mu$ ,  $S_i$ , and  $e_{ij}$  were as described in the Equation 1,  $b$  = Partial linear regression coefficients of  $Y_{ij}$  on pre-slaughter weight,  $X$  = Value of pre-slaughter weight and  $x$  = Overall average of pre-slaughter weight.

**RESULTS AND DISCUSSION*****Growth performance and feed efficiency***

The absence of significant variance in initial body weight between the experimental groups showed the groups were homogeneous at the start of the experiment. Regarding the impact of selenium form on the growth indicators of male rabbits, it was found that the supplementation of selenium positively significantly ( $P < 0.01$  or  $0.05$ ) affected the final live body weight, daily gain weight, relative growth, feed conversion, and feed intake (Table 2) .

Rabbits fed diets supplemented with nano-selenium had the highest body weight, daily body gain and relative growth rate compared to the other groups (Figure 1). Rabbits fed diets supplemented with selenium (inorganic or nano) recorded higher daily feed intake when compared to the control group. On the other hand, rabbits fed nano-selenium form recorded the best feed conversion. Final body weight and daily weight again (5-13 weeks of age) were increased in rabbits fed diet supplemented with nano-selenium by about 16.18 and 22.18 %, respectively compared to the control group; while the same trend was found in rabbits fed diet supplemented with inorganic selenium (9.64 and 13.01 %, respectively ,Figure 1). Also feed conversion improved by 7.14% in rabbits fed

**Table 2.** Performance parameters of growing rabbit as affected by dietary inorganic (Sodium selenite) and nano-selenium supplementation from 5 to 13 weeks of age.

Parameters	Experimental groups			Sig.
	Control	Inorganic Se	Nano Se	
<b>Body weight, g</b>				
5 weeks (Initial)	582.40±14.28	584.70±11.13	580.30±15.75	NS
8 weeks	1383.80 <sup>b</sup> ±39.23	1487.70 <sup>ab</sup> ±39.81	1593.00 <sup>a</sup> ±35.62	**
13 weeks	2188.00 <sup>c</sup> ±26.59	2399.00 <sup>b</sup> ±59.66	2542.20 <sup>a</sup> ±50.69	**
<b>Daily weight gain, g</b>				
5-8 weeks	28.62 <sup>c</sup> ±1.16	32.25 <sup>b</sup> ±1.17	36.17 <sup>a</sup> ±1.10	**
8-13 weeks	28.72 <sup>c</sup> ±0.99	32.55 <sup>b</sup> ±0.97	33.90 <sup>a</sup> ±0.69	**
5-13 weeks	28.67 <sup>c</sup> ±0.39	32.40 <sup>b</sup> ±0.95	35.03 <sup>a</sup> ±0.82	**
<b>Relative growth rate, g</b>				
5-8 weeks	81.37 <sup>c</sup> ±1.91	86.97 <sup>b</sup> ±1.44	93.17 <sup>a</sup> ±1.99	**
8-13 weeks	45.26±1.97	46.91±0.98	45.95±0.62	NS
5-13 weeks	115.97 <sup>c</sup> ±1.37	121.47 <sup>b</sup> ±1.26	125.64 <sup>a</sup> ±1.53	**
<b>Daily feed intake, g</b>				
5-8 weeks	84.20 <sup>b</sup> ±1.29	89.00 <sup>a</sup> ±1.50	86.00 <sup>ab</sup> ±1.11	*
8-13 weeks	148.30 <sup>b</sup> ±2.38	177.00 <sup>a</sup> ±2.26	176.70 <sup>a</sup> ±1.96	*
5-13 weeks	116.50 <sup>b</sup> ±1.38	133.20 <sup>a</sup> ±1.76	131.60 <sup>a</sup> ±1.00	**
<b>Feed conversion, g feed/g gain</b>				
5-8 weeks	2.97 <sup>a</sup> ±0.11	2.79 <sup>a</sup> ±0.09	2.40 <sup>b</sup> ±0.09	**
8-13 weeks	5.23±0.24	5.48±0.15	5.24±0.16	NS
5-13 weeks	4.06 <sup>a</sup> ±0.05	4.13 <sup>a</sup> ±0.10	3.77 <sup>b</sup> ±0.11	*

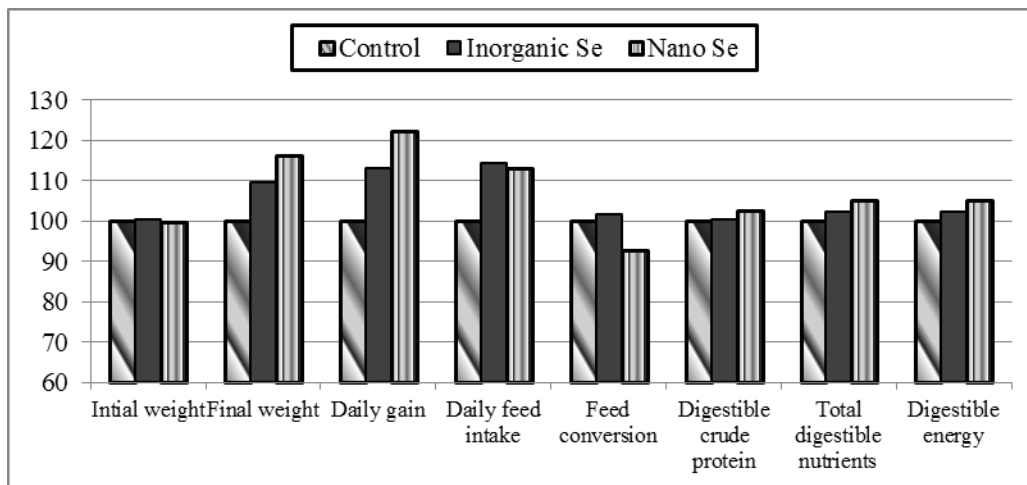
Means in a row different superscripts differ significantly (  $P \leq 0.05$  ).

NS = Not significant, \*  $P < 0.05$ , \*\*  $P < 0.01$ , Sig: Significance

diet supplemented with nano-selenium than the control group from 5-13 week. Jianhua *et al.* (2000) found that selenium is associated with thyroid function and more specifically, with the secretion of thyroid seleno enzymes and deiodinases, which promote the metabolism of T3 from T4. This explains the beneficial impact of selenium on growth.

In the present study, nano-selenium fortification enhanced the growth efficacy of growing NZW rabbits more than inorganic selenium supplementation. Mohapatra *et al.* (2014) observed that nanoparticles can be used as an additional source of trace minerals in diets due to their unique characteristics distinct from those of other selenium forms. In this concern, Abd Allah *et al.* (2020) showed

that rabbits fed basal diet supplemented with nano-selenium had significantly higher final live body weight, total body gain and better feed conversion ratio than those fed the control basal diet and those fed organic selenium diet. According to Emara *et al.* (2019), nano-selenium administration significantly increased ( $P < 0.01$ ) daily gain and final body weight in comparison with the remaining groups, with average values of 24.40 g/day and 2.09 kg, respectively. In comparison with the control group, inorganic selenium did not have a significant effect on the tested traits.



**Figure 1.** Growth performance and feed efficiency index of growing rabbits as affected by selenium form, when considering the values of the control group (fed diet without selenium supplementation) as 100%.

The higher effectiveness of nano particles has been attributed to their larger surface area, smaller size of particles, and better mucosal permeability, enhance gastrointestinal absorption, and tissue depositions. Furthermore, the reaction that occurs between selenium nano particles and the carboxyl group of proteins results in good absorptive ability and strong biological activity (Zhang *et al.*, 2001).

### ***Digestibility of nutrients***

Table (3) demonstrates the effect of various selenium sources on digestibility coefficients. All digestibility coefficients of various nutrients and nutritive values were significantly ( $P < 0.05$  or  $0.01$ ) affected by dietary selenium supplementation, except for CF. The Se-NP-supplemented rabbit diets had higher digestibility of



**Table 3.** Nutrients digestibility and nutritive values of growing rabbit as affected by dietary inorganic (Sodium selenite) and nano-selenium supplementation.

Parameter	Experimental groups			Sign.
	Control	Inorganic Se	Nano Se	
<i>Nutrient digestibility, %</i>				
Dry matter	61.52 <sup>b</sup> ±0.98	63.56 <sup>ab</sup> ±0.29	65.19 <sup>a</sup> ±0.44	**
Ether extract	81.62 <sup>a</sup> ±0.97	76.79 <sup>b</sup> ±0.96	79.90 <sup>ab</sup> ±1.79	*
Crude protein	73.80 <sup>b</sup> ±0.33	74.11 <sup>b</sup> ±0.46	75.79 <sup>a</sup> ±0.44	*
Crude fiber	20.88±0.65	20.20±0.46	20.92±0.66	NS
Nitrogen free extract	68.77 <sup>b</sup> ±1.12	72.42 <sup>a</sup> ±0.39	74.24 <sup>a</sup> ±0.63	**
Organic matter	62.94 <sup>b</sup> ±0.85	64.85 <sup>ab</sup> ±0.29	66.54 <sup>a</sup> ±0.55	**
<i>Nutritive values, %</i>				
Digestible crude protein	12.42 <sup>b</sup> ±0.06	12.47 <sup>b</sup> ±0.08	12.76 <sup>a</sup> ±0.07	*
Total digestible nutrients	61.76 <sup>b</sup> ±0.79	63.21 <sup>ab</sup> ±0.27	64.93 <sup>a</sup> ±0.57	*
Digestible energy,	2470 <sup>b</sup> ±31.50	2528 <sup>ab</sup> ±10.73	2597 <sup>a</sup> ±22.79	*

Means in a row different superscripts differ significantly ( $P \leq 0.05$ ).

NS = Not significant, \*  $P < 0.05$ , \*\*  $P < 0.01$ , Sig: Significance

DM, CP, and OM, as well as higher nutritive values than the rest studied groups. Digestible CP, TDN and DE increased by 2.70, 5.13 and 5.14%, respectively in rabbits fed diet supplemented with nano-selenium compared to the control group; whereas the corresponding values for rabbits fed diets supplemented with inorganic selenium were 0.42, 2.35, and 2.35%, respectively (Figure 2).

The positive impacts of supplemental Se-NP on animal performance and digestibility of nutrients may be attributable to its function in enhancing feed utilization by stimulating digestive enzyme activity (Shi *et al.* 2011). The findings of Xu *et al.* (2003) which demonstrated that broiler chicks absorbed Se-NP more readily than inorganic forms. Similarly, Xun *et al.* (2012) showed that Se-NP fortification in the diet increases the digestibility of DM, OM, CP, EE, neutral detergent fiber, and acid detergent fiber in the whole gastrointestinal tract.

### **Carcass traits**

Pre-slaughter fasting weight was significantly ( $P < 0.05$ ) affected by the dietary treatments, while actual carcass weight and dressing percentages insignificantly affected by selenium supplementation in rabbit diets (Table 4). Furthermore, all treatments had no significant impact on the adjusted carcass weight and its cuts. Payne and Southern (2005) and Downs *et al.* (2000) did not

**Table 4.** Internal organs weight and carcass parts of growing rabbit as affected by dietary inorganic (Sodium selenite) and nano-selenium supplementation.

Parameters	Experimental groups			Sig.	LW-Sig.
	Control	Inorganic Se	Nano Se		
Pre-slaughter weight (g)	2212.3 <sup>b</sup> ±81.46	2522.7 <sup>ab</sup> ±95.79	2627.7 <sup>a</sup> ±110.89	*	–
Actual carcass weight (g)	1263.3±48.11	1458.7±73.77	1467.3±51.75	NS	–
Adjusted carcass weight (g)	1406.8±21.01	1418.06±15.30	1364.4±18.22	NS	NS
Carcass weight (%)	57.10±0.29	57.77±0.72	55.88±0.44	NS	***
<b>Carcass parts weight (g)</b>					
Head	133.84±2.94	136.59±2.14	127.89±2.55	NS	**
Fore part	481.89±8.17	464.43±5.95	441.67±7.08	NS	***
Intermediate part	311.73±13.56	323.17±9.88	302.09±11.76	NS	*
Hind part	424.79±15.68	468.97±11.43	460.57±13.60	NS	*

Means in the same row within each classification with different letters, differ significantly (P<0.05).

NS = Not significant, \* P<0.05, \*\* P<0.01, \*\*\* P<0.001, Sig: Significance, LW-sig: Live body weight significance.

find any differences in the yields of carcass, breast, and thigh muscles in broilers administered with selenium-enriched diets. In addition, Cai *et al.* (2012) indicated that Se-NP had no significant impact on the carcass parts of broilers.

#### ***Hematological and immunological parameters***

All studied blood hematological parameters (within normal rang) were insignificantly affected with the treatments, except blood hemoglobin was significantly (P<0.05) affected (Table 5). Hemoglobin level was significantly increased (P< 0.05) by selenium (either inorganic or nano form) supplementation in rabbit diets. Rabbits fed diet supplemented with nano-selenium recorded the higher hemoglobin concentration in comparison with other experimental groups. On the other hand, the concentration of each RBCs, Hct, MCV, MCH, MCHC and lymphocyte concentrations were insignificantly improved in rabbit blood fed diets supplemented with selenium when compared to the control. Besides, complement C3 was affected significantly (P<0.01) with selenium supplementation in rabbit diets, while IgM and lysozyme activity were significantly affected (Table 5). Lysozyme activity, IgM, and C3 levels increased by 72.04, 24.11 and 47.19%, respectively in rabbits fed inorganic selenium supplemented diet when compared with the control. Also, C3 and IgM levels increased by 81.19% and 164.54%, respectively for the rabbits fed diet

**Table 5.** Hematological parameters of growing rabbit as affected by dietary inorganic (Sodium selenite) and nano-selenium supplementation.

Parameters	Experimental groups			Sig.
	Control	Inorganic Se	Nano Se	
<b>Erythrogram</b>				
RBCs, $10^6 \times \mu\text{L}$	5.20 $\pm$ 0.05	5.44 $\pm$ 0.06	5.44 $\pm$ 0.18	NS
Hb, g/dL	11.07 <sup>b</sup> $\pm$ 0.18	11.77 <sup>ab</sup> $\pm$ 0.15	12.07 <sup>a</sup> $\pm$ 0.33	*
Hct, %	32.20 $\pm$ 1.18	33.43 $\pm$ 0.48	36.27 $\pm$ 1.57	NS
MCV, fL	61.98 $\pm$ 2.82	61.49 $\pm$ 0.23	66.69 $\pm$ 1.31	NS
MCH, pg/dL	21.29 $\pm$ 0.47	21.65 $\pm$ 0.14	22.20 $\pm$ 0.34	NS
MCHC, g/dL	34.43 $\pm$ 0.84	35.19 $\pm$ 0.31	33.35 $\pm$ 1.18	NS
<b>Leukogram</b>				
WBCs, $10^3/\mu\text{L}$	6.93 $\pm$ 2.32	6.63 $\pm$ 0.91	6.57 $\pm$ 2.05	NS
Lymphocyte, %	53.00 $\pm$ 10.69	51.67 $\pm$ 5.36	64.67 $\pm$ 7.42	NS
Neutrophils, %	33.00 $\pm$ 7.00	33.33 $\pm$ 6.01	26.33 $\pm$ 6.84	NS
Monocytes, %	10.33 $\pm$ 3.76	9.67 $\pm$ 0.33	4.67 $\pm$ 0.67	NS
Band neutrophils, %	2.00 $\pm$ 0.00	3.00 $\pm$ 0.58	3.00 $\pm$ 1.00	NS
Eosinophils, %	1.67 $\pm$ 0.33	2.33 $\pm$ 0.88	1.33 $\pm$ 0.33	NS

Means in the same row within each classification with different letters differ significantly (P<0.05).

NS = Not significant, \* P<0.05, Sig: Significance

RBCs, Red blood cells. Hct, The hematocrit. Hb, Hemoglobin. MCV, Mean corpuscular volume. MCHC, Mean corpuscular hemoglobin concentration. WBCs, White blood cells.

supplemented with Se-NP when compared with the control group, while lysozyme activity decreased (Figure 2).

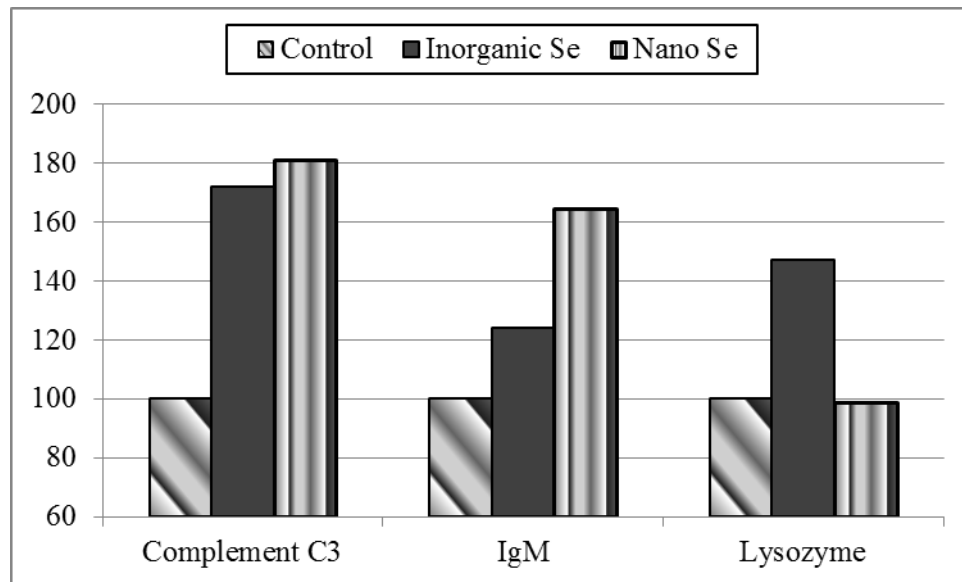
Hematological examination provides a useful tool for diagnosing metabolic problems. It is also beneficial for diagnosing diseases of certain tissues and organs (Roland *et al.*, 2014). Leukocytes or white blood cells (WBC) play an essential function in the immune system's functioning and include neutrophils, eosinophil, granulocytes, monocytes, and lymphocytes. Mohriet *al.* (2011) reported that the addition of selenium had no effect on hematological parameters, which is in agreement with our findings. In addition, Shinde *et al.* (2009) noted that fortification with 0.3 ppm selenium in the form of sodium selenite had no significant impact on the concentrations of RBC, WBC, Hct, and Hb.

**Table 6.** Immunological indices of growing rabbit as affected by dietary inorganic (Sodium selenite) and nano-selenium supplementation.

Parameters	Experimental groups			Sig.
	Control	Inorganic Se	Nano Se	
Complement C3 (mg/dL)	36.94 <sup>b</sup> ±2.06	63.55 <sup>a</sup> ±1.86	66.93 <sup>a</sup> ±3.69	**
Immunoglobulin M (mg/ml)	5.64±0.516	7.00±0.57	9.28±1.58	NS
Lysozyme(U/mL)	29.67±6.44	43.67±8.37	29.33±4.67	NS

Means in the same row within each classification with different letters differ significantly ( $P < 0.05$ ).

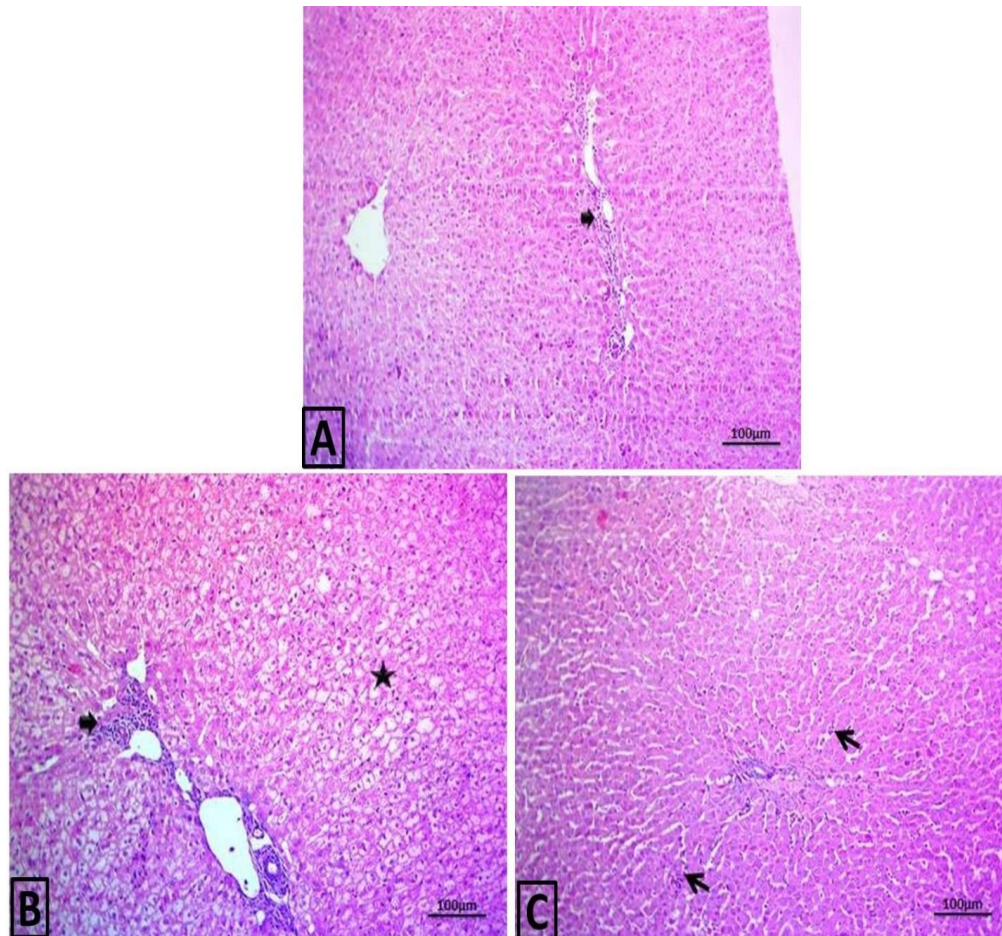
NS = Not significant , \*\*  $P < 0.01$ , Sig: Significance



**Figure 2.** Immunological activity index of growing rabbits as affected by selenium form, when considering the values of the control group (fed diet without selenium supplementation) as 100%.

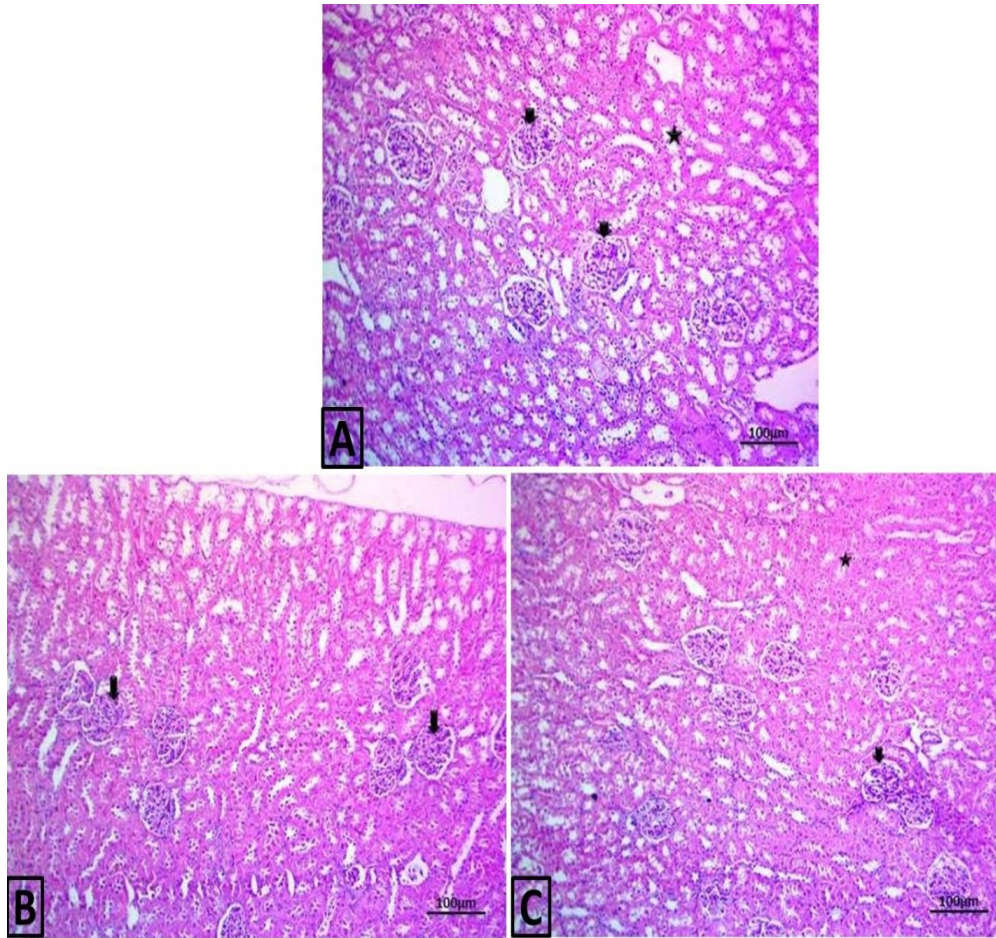
### *Histology of liver and kidney*

The liver tissue from the control group of rabbits showed hyperplasia of kupffer cells and a seemingly normal center vein and a small number of lymphocytes in the portal venues (Figure 4A). Rabbits provided inorganic Se showed low hepatocyte degeneration and periportal inflammatory infiltrations



**Figure 4.** Representative photomicrographs of the rabbit liver of (A) control group : showing apparently normal center vein, minute lymphocytes in portal tracts (arrow). H&E X 100. (B) Inorganic selenium: showing moderate periportal inflammatory infiltrations (arrow) and mild degenerated hepatocytes (star). H&E X 100. (C) Se-NP group: showing apparently normal hepatic architectures with marked disseminated a few numerous inflammatory cells in the sinusoids (arrows). H&E X 100.





**Figure 5.** Representative photomicrographs of the rabbit Kidney of (A) control group: showing apparently normal glomeruli (arrows), followed by apparently normal renal tubules (star). H&E X 100. (B) Inorganic selenium group: showing apparently normal glomeruli (arrow) and proximal tubules. H&E X 100. (C) Se-NP group: showing nearly normal renal tubules (star) and marked increase glomerular cellularity (arrow). H&E X 100.

(Figure 4B). Sacrificed rabbits in the Se-NP group had seemingly normal hepatic structures, but histological examination revealed a few inflammatory cells dispersed throughout the sinusoids (Figure 4C). The kidney specimens of the

control group displayed normal glomeruli, proximal and distal renal tubule histomorphology (Figure 5A). Kidney tissues from rabbits that were fed inorganic Se revealed seemingly normal glomeruli and proximal tubules with or without mildly lowered glomeruli and few exfoliated renal epitheliums (Fig.5B). Whereas kidney tissues of rabbits administered Se-NP revealed renal tubules that were virtually normal and an increase in glomerular cellularity (Figure 5C).

### ***Economic feed efficiency***

Economic feed efficiency was higher in rabbits fed diets supplemented with selenium when compared to those fed control diet as shown in Tables 2 and 7. Total gain and return from gain increased by 12.95 and 27.7%, respectively in rabbits fed diet supplemented with nano-selenium when compared to the control group; while the same trend for rabbits fed diet supplemented with inorganic selenium being 14.33 and 9.2 %, respectively. Rabbits fed diet supplemented with nano-selenium recorded higher return from total body gain and return from gain when compared with other experimental groups.

**Table 7.** Economic feed efficiency of growing rabbit as affected by dietary inorganic (Sodium selenite) and nano-selenium supplementation.

Parameters	Experimental groups		
	Control	Inorganic Se	Nano Se
Total body gain (kg)	1605.60	1814.30	1961.90
Total feed intake (Kg)	6.524	7.459	7.369
Return from gain (LE)*	80.28	90.72	98.10
Price of daily feed intake (LE)	6.89	6.99	7.19
Feed cost, LE/rabbit	44.95	52.14	52.98
Return from gain, LE/rabbit**	35.33	38.58	45.12
Relative return%	100	109.2	127.7

\*Price was calculated according to the local market price which was 50 L.E/Kg rabbit live weight

\*\*Return from gain, LE/rabbit= Return from gain (LE) - Feed cost

### **Conclusion**

The current study suggested that dietary supplementation with Se-NP was utilized efficient than inorganic selenium which reflect on final body weight, relative growth rate, daily weight gain, daily feed intake, feed conversion ratio,

nutrient digestibility, and nutrient values. In addition, the highest economic feed efficiency was found in rabbits fed diets containing Se-NP. Nonetheless, both nano and inorganic forms of selenium enhanced rabbit growth performance and immunity. Therefore, dietary Se-NP appears to be more suitable for growing rabbits in order to increase the efficiency of production.

**Ethical Approval:** All the procedures for this study were conducted in accordance with a protocol approved by the institutional animal care and use Committee of Zagazig University (approval number: ZU-IACUC/2/F/144/2023).

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### تأثير إضافة مصادر غير عضوية ونانوية للسيلينيوم على مؤشرات النمو، الهضم، صفات الذبيحة والحالة المناعية للأرانب النامية

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تؤثر المصادر المختلفة من العناصر المعدنية النادرة بشكل متباين على أداء الأرانب بناءً على درجة امتصاصها في الكائن الحي. تهدف هذه الدراسة إلى تقييم تأثير مصدرين للسيلينيوم (غير العضوي والنانو) كإضافة غذائية على النمو، وهضم العناصر الغذائية، والمناعة، وصفات الذبيحة في الأرانب النيوزيلندية البيضاء. تم تقسيم عدد ٤٥ من الأرانب الذكور (بعمر ٥ أسابيع) إلى ثلاث مجموعات (١٥ أرانب / مجموعة) وتم تغذيتهم على العلائق التالية: (١) عليقة الكنترول، (٢) عليقة الكنترول + سيلينيات الصوديوم (٠.٣ مجم

سلينيوم/ كجم عليقة)، أو ٣) عليقة الكنترول + نانو سلينيوم (٠.٣ مجم سلينيوم/ كجم عليقة). استمرت التجربة لمدة ٥٦ يومًا.

**أظهرت النتائج** زيادة معنوية في وزن الجسم النهائي، ومعدل النمو النسبي، ومعدل الوزن اليومي، وكمية العلف المأكول اليومية في الأرانب المغذاه على علائق مدعمة بكلا من السلينيوم في صورتية غير العضوى والنانوسلينيوم بالمقارنة بمجموعة الكنترول في حين حققت المجموعة المدعمة بالنانو سلينيوم أفضل النتائج. أدت إضافة النانو سلينيوم إلى تحسن نسبة التحويل الغذائى، وهضم العناصر الغذائية، والقيم الغذائية، وتركيز الهيموجلوبين بالمقارنة بباقي المجموعات. بالإضافة إلى ذلك، حققت الأرانب المغذاه على علائق مدعمة بالنانو سلينيوم أعلى هامش ربح نهائى بالمقارنة بباقي المجموعات. لم تظهر أى تشوهات نسيجية في أنسجة الكبد والكلى في كل المجموعات التجريبية، مما يشير إلى أن الجرعات المختبرة من السلينيوم كانت آمنة.

**التوصية:** نستخلص من ذلك أن إضافة النانو سلينيوم في علائق الأرانب النامية أفضل بشكل معنوى من السلينيوم غير العضوي في تعزيز الإستفادة من الغذاء وأداء النمو وهضم العناصر الغذائية والكفاءة الإقتصادية. بالإضافة إلى ذلك، لم يكن لمستوى النانو سلينيوم الذي تم اختباره أى تأثير سلبى على أنسجة الكبد والكلى .