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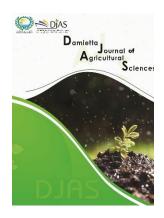
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The Combined Effects of Selenium Nanoparticles and *Lactobacillus plantarum* on the Performance of Whiteleg Shrimp (*Penaeus vannamei*)

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Abstract:

probiotics and nanoparticles are strongly recommended in aquaculture; little is known about their synergistic effects. Thus, this study assessed the potential synergistic effects of Se nanoparticles (SeNps) and L. plantarum (Lpm) on growth performance, feed conversion ratio, serum biochemistry, and antioxidant activity in whiteleg shrimp (P. vannamei). For P. vannamei, three test diets and a control diet were supplemented with SeNps, Lpm, or both. In comparison to the control, the results showed that shrimp fed SeNps and/or Lpm had greater ultimate weight, weight gain, and specific rate of growth, with the highest values observed in shrimp fed both SeNps and Lpm. Fish fed SeNps or Lpm had a poorer feed conversion ratio, with shrimp fed each of SeNps and Lpm having the lowest ratio. The biochemical variables showed non-significant variations between the groups and normal values. Dietary SeNps and/or Lpm had a substantial impact on the AST and ALT levels compared to the control. TAC, SOD, and CAT activities were significantly (P < 0.05 higher in the group that consumed diets supplemented with Lpm, either alone or in combination with SeNps. MDA levels were higher in P. vannamei control diet than in other groups. Dietary supplementation with a blend of SeNps and Lpm substantially (P < 0.05) increased the levels of HSP70, IGF-1, and IL-1\beta in the P. vannamei treated group.

Key words: Se Nanoparticles, L. plantarum, growth performance, and Antioxidants, Whiteleg shrimp

INTRODUCTION

There are a number of antibiotic alternatives for fish aquaculture (Lozano et al., 2018). Examples of these alternatives include environmentally friendly food production, food conversion, microbial balance, immunity, growth, health status, and nutrition (Das et al., 2017). Microorganisms, when administered in sufficient quantities, confer health benefits to the host (FAO/WHO, 2006). **Probiotic** supplementation may help manage a variety of bacterial infections in rainbow (Sharifuzzaman et al., 2017), other fish species and shrimp (Carvalho et al., 2022).

The ability to produce bioactive compounds like ethanol, lactic acid, acetate, formic acid, enzymes, free fatty acids, antimicrobial peptides, and volatile substances allows some strains of lactic acid bacteria to have probiotic properties and act as broad-spectrum antibacterials against a range of pathogens (Olofsson *et al.*, 2016).

On the other hand, mammals require the element selenium (Se). Se is necessary for metabolic functions related to reproduction,

growth, development, and health. As a dietary supplement, it is necessary for cultured fish (Naderi et al., (2017); Takahashi et al., 2020). Additionally, seleno-proteins—need selenium as a cofactor help increase the elimination of reactive oxygen species (ROS)-and prevents oxidative stress (Baeverfjord et al., 2019). Rathore et al. (2021) claim that elemental Se nanoparticles (SeNps) can be employed to stimulate the immune system, promote development, and act as an antioxidant in aquacultured species. Several research studies have also documented the advantages of feeding aquatic animals SeNPs to improve their physiological and health conditions as well as their growth performance (Deilamy et al., (2021); Ibrahim et al., (2021), and Karamzadeh et al., 2021). SeNPs have a low toxicity and a high level of usefulness. Sarkar et al. (2015). Significantly, adding SeNPs as a dietary supplement has been shown to improve growth performance and production in aquatic animals more effectively than other forms of Se (Mechlaoui et al., 2019). It has been demonstrated that a meal supplemented with Se-enriched Lpm protects against Cadmium (Cd) toxicity, lowering oxidative stress in fish *Luciobarbus capito* (Shang *et al.*, 2022) and having immunomodulatory and anti-inflammatory effects in mice (Khattab *et al.*, 2022). This study sought to determine the impact of selenium nanoparticles (SeNps) and/or probiotic (Lpm) on growth performance, feed consumption, antioxidant status, and liver Enzyme activity of *P. vannamei* considering the advantages that both can offer the whiteleg shrimp farming.

3. MATERIALS AND METHODS: Feeding Experimental study and *P. vannamei* rearing:

This study was carried out in a private farm in Shatta, Damietta City, Damietta Governorate, Egypt. In this investigation, 240 healthy juvenile of P. vannamei weighing 2.44±0.004g were employed. Following their transfer to the experimental enclosures (hapa) on the same farm, the juveniles were randomly divided into four treatments (3 hapa/ treatment), at random, and the juvenile were not fed in order not to increase stress during transportation. Hapa (1 x 2 x 1.25 m) *P. vannamei* (20 juvenile /hapa). A feed that had been modified to 5% of the P. vannamei biomass by weight was given to them. The tested diets were manually administered to the P. vannamei twice a day, at 9:00 am and 3:00 pm, and the quantity of feed they ingested each day was recorded. Every two weeks, P. vannamei were collected from each hapa, weighed, and the amount of feed was modified to consider variations in body weight over the trial. Selenium nanoparticles (SeNps) with an average size (TEM) of 65±10 nm and a concentration of 300 ppm (Nano Gate, Cairo, Egypt) and Lpm/kg diet; Lpm, 2×10^{11} CFU/g, Free Trade Egypt Company, Egypt). were used in four isocaloric and isonitrogenous diets as a safe feed supplement. As the control group, the first group was fed a commercial meal devoid of feed additives (T1). Kg diet supplemented with 0.004 ml of SeNps (T2), 400 mg of Lpm (T3), and a combination of 0.004 ml Se Nano + 400 mg Lpm (T4) per kg of diet comprised the other treatment groups. Throughout the 14-weeks feeding trial, the rearing water temperature was constant and was ideal for *P. vannamei* production. A natural photoperiod of 12 hours of light and 12 hours of darkness was kept for the shrimp groups. Throughout the 14-weeks feeding trial, the rearing water temperature was constant and was ideal for *P. vannamei* production. A natural photoperiod of 12 hours of light and 12 hours of darkness was kept for the shrimp groups. In the meantime, daily measurements were made of the rearing water temperature (25.33 \pm 0.69 °C), salinity (22.93 ppt), dissolved oxygen levels (4.8 \pm 0.9 mg/L), and pH (7.27 \pm 0.09). 0.033 \pm 0.002 mg/L for nitrite (NO₂-), 0.037 \pm 0.003 mg/L for nitrate (NO₃-), and 0.038 \pm 0.001 mg/L for ammonia (NH₃+) were the mean values for the other predicted water quality criterion biweekly. According to Adiwidjaya *et al.* (2003), these values fall within the permissible ranges needed for *P. vannamei* growth to be at its best.

Formulating and Analyzing Tested Diets:

The experimental diets were prepared by thoroughly mixing the dry ingredients and adding 200 milliliters of water for each kilogram of diet. Once the mixture reached a paste consistency, it was pelleted using a laboratory pellet machine with a 1 mm diameter. mix 30 ml of Nutri-B Gel (binding agent) with 4 ml of water and the feed additive 0.004ml SeNPs and/or 0.400 mg L. plantrum. Then, this mixture was evenly blend with 1kg of shrimp diet. The pellets were stored in plastic bags at 4 °C until they were needed after drying at room temperature. The developed diets were designed to meet P. vannamei shrimp nutritional requirements as listed in Table 1 and suggested by Elkin et al. (2007) The chemical compositions of formulated feed, including the concentrations of moisture, dry matter, crude protein, crude fat, and ash, have been analyzed using the AOAC (2000) standard applicable techniques.

Growth performance and indicators of feed utilization:

Growth indices such as weight gain (WG), average daily gain (ADG), specific growth rate (SGR), and feed utilization measurements such as feed conversion ratio (FCR), protein efficiency ratio (PER), and survival rate percentage per hapa were measured for shrimp in each treatment that were routinely weighed every two weeks during the *P. vannamei* trial. Before obtaining weight samples using the following formulas, they were deprived for a whole day:

- ightharpoonup Weight gain (WG, g/ fish) = FW (g)-IW (g).
- ➤ Daily weight gain (DWG, g/ fish/day) = WG (g) /P (days).
- > Specific growth rate (SGR, % day 1) = (Ln FW-Ln IW) /P) × 100.
- ightharpoonup Feed conversion ratio (FCR, g / g) = TFI (g) /WG (g).

- ightharpoonup Relative growth rate (RGR, %) = (WG/IW) imes 100.
- ightharpoonup Protein efficiency ratio (PER, g/g) = WG (g)/PI (g).
- ightharpoonup Mortality rate percentage (%) per hapa = (number of stocked fish per hapa number of fish harvested per hapa) /number of stocked fish per hapa $\times 100$

Hemolymph Sampling Method:

Following a 14-week experimental investigation, all *P. vannamei* were fasted for 24 hours prior to sampling. Six shrimp were randomly selected from each treatment group for this purpose. According to Naiel *et al.* (2022), each *P. vannamei* sample was anesthetized 40 mg/L olive oil as described by Feldman *et al.* (2000). Following a 10-minute centrifugation at 800 g at 4°C, hemolymph was extracted and kept for subsequent analysis at -75°C.

Biochemical Measurements:

A colorimetric approach was used to quantify the serum protein constituents, such as total protein (TP) and albumin (Alb). The ALB value was subtracted from the TP to determine the globulin (Glob) level. The manufacturer's procedure was followed when evaluating the aspartate transaminase (AST), alanine aminotransferase (ALT), and glucose using kits (Biodiagnostic, Giza, Egypt).

Antioxidant Activity measurements:

Serum levels of catalase, superoxide dismutase (SOD), and total antioxidant capacity (TAC) were measured using the procedures outlined by Marklund and Marklund (1974). The Draper and Hadley (1990) method was used to quantify malondialdehyde (MDA).

Gene Expression:

Following the manufacturer's instructions. Trizol reagents (iNtRON Biotechnology) were used to isolate and extract total RNA from the liver samples. Two microliters of RNase were combined with twenty microliters of DNA mixed in Tris-buffer solution (pH = 8.0) and incubated for three to four hours at 37°C to avoid RNA contamination. Nanodrop (Quawell, USA) was then used to measure the RNA concentration. For a few chosen genes, such as insulin-like growth factor I (IGF-I), heat shock protein (HSP70), and interleukin-1 β (IL-1 β), realtime PCRs were accomplished. Table 2 provides an illustration of the primers used in this investigation. Following the Pereira-Gomez et al. (2020) protocol, real-time PCR amplifications were carried out using the Sensi Fast SYBR Lo-Rox kit (Bioline) in 20 µl reaction mixtures that contained 2 μ l of cDNA, the gene-specific primers (0.5 μ M each), and SYBR 10 μ l. Initial denaturation at 95°C for 10 minutes, 40 cycles at 95°C for 15 seconds, and 60°C for 1 minute were the parameters for the thermal cycling. Three separate estimates of the genes were made. The 2– CT formula was used to estimate the fold change Livak and Schmittgen (2001).

Statistical analysis:

One-way analysis of variance (ANOVA) was used to examine the data, and Duncan's test was used to exhibit mean differences at ($P \le 0.05$). Data analysis results are presented as mean \pm standard error (SE). All statistical analyses were performed via (SAS, 2012).

RESULTS:

The growth results of P. vannamei fed T1 (control diet) and diets supplemented with SeNps and/or Lpm for 14 weeks are shown in Table 3. One-way ANOVA revealed that all growth parameters, including final weight (FW), weight gain (WG), specific growth rate (SGR), and relative growth rate (RGR), significantly affected by dietary SeNp and Lpm supplementation and their interaction (**Table**, 3). Additionally, the diet supplemented with high amounts of both SeNps and Lpm produced the greatest FBW, WG, SGR, and RGR values of any group ($P \le 0.05$). Furthermore, when compared to the CTR group, SR (%) increased significantly $(P \le 0.05)$ in all experimental groups, and P. vannamei administered the combination supplementation of SeNps and Lpm had the highest SR%.

Feed utilization:

Dietary SeNps or Lpm supplementation and the control group, as well as their interaction, had a significant impact on all feed efficiency, according to the one-way ANOVA analysis (**Table, 4**). Additionally, the best TFI, FCR, FE, and PER values were obtained by combining of both SeNps and Lpm supplementation in the diet from all groups (P < 0.05).

Serum Biochemistry:

Compared to the other experimental groups, a dietary SeNps in combination with Lpm did not substantially increase serum TP (**Table**, **5**). Additionally, the presence of all feed supplements and their interaction did not significantly enhance the concentrations of ALB and GLOB. In the meantime, the *P. vannamei* group that received SeNps and combination with Lpm showed a considerable drop in ALT levels. Lastly, in comparison to the other groups, the *P. vannamei* group fed SeNps and/or Lpm did not exhibit a significant change in AST levels.

Antioxidant and immunological responses:

In terms of the antioxidant activity results (**Table**, **6**), it was found that, in comparison to other *P. vannamei* groups, the group that received meals supplemented with Lpm either alone or in conjunction with SeNps had considerably ($P \le 0.05$) higher TAC, SOD, and CAT activities. However, MDA levels in P. *vannamei* diets supplemented with fortified feed did not differ significantly from those in the control group (T1).

Gene transcription:

The effects of feed additives on the transcription of P. vannamei target genes were demonstrated by the data in Figure 1. The transcription of HSP70, $IL-1\beta$, and IGF-1 in P. vannamei has been discovered to be considerably regulated by adding SeNps and/or Lpm to their feed diets. In particular, compared to the other treatment groups, the P. vannamei shrimp group fed diets supplemented with a combination of SeNps and Lpm showed a highly substantial (P < 0.05) elevation of HSP70, IGF-1, and $IL-1\beta$ (Fig 1).

DISCUSSION

Using probiotics should enhance fish nutrition and/or health, and they shouldn't have antibiotic-resistant genes encoded in their plasmids. According to Denev *et al.* (2009), probiotics have been suggested as a substitute for antibiotics and chemotherapeutants in order to lessen the harmful effects of stress, stop disease outbreaks, and boost

fish immune systems and antioxidant capacity. Furthermore, the gut microbiome is supported by probiotic dietary supplements. According to Hai (2015), probiotics are being utilized to reduce infections because illness is a major cause of losses in intensive fish farming. Feeding diets with SeNps and/or Lpm to P. vannamei resulted in a considerable improvement in growth performance and feed utilization (lower FCR and PER), according to this study. T4, T3, and T2 were the best doses for feed consumption and growth performance, according to the data. The results primarily support those of Du et al. (2022), who discovered that P. vannamei that received an oral dose of Lpm for four weeks shown considerable gains in growth performance and feed efficiency. Increased feed utilization, growth promotion, immune response enhancement, and improved stress tolerance are just a few of the benefits of using Lpm as a probiotic in fish production according to (Dawood et al., 2019; Valipour et al., 2019), (Zhai et al., 2017), (Silarudee et al., 2019), and (Dawood et al., 2019). Selenium (Se), the main component of the 50-deiodinase enzyme, which catalyzes the bioconversion of thyroxin to triiodothyronine, has also been demonstrated to be able to control the production and release of growth hormones and considerably improve the growth performance of animals (Ibrahim et al., 2011).

Table 1. Examination of the experimental diets' ingredients and chemical composition

	T1 (Control)	T2 (C-N)	T2 (I)	T4
Items	T1 (Control)	T2 (SeNps)	T3 (Lpm)	(SeNps+Lpm)
Fish meal (62% CP)	27	27	27	27
Meal of Corn gluten (60% CP)	6	6	6	6
Meal of Soybean (48% CP)	42	42	42	42
Starch	6	5.996	5.6	5.596
Fish oil	9.6	9.6	9.6	9.6
Corn oil	6.4	6.4	6.4	6.4
Mineral and Vitamin premix ^a	3	3	3	3
Nano Selenium 300 ppm/ml	0	0.004	0	0.004
L. plantarum (Lpm) mg/kg	0	0	0.4	0.4
Sum	100	100	100	100
Moisture	6.5	6.45	6.52	6.48
Ash	14.42	14.43	14.42	14.41
Crude protein	37.12	37.11	37.09	37.06
Crude lipid	7.83	7.57	7.4	7.88
Fiber	6.7	6.65	6.72	6.68
NFE ^b	33.93	34.24	34.37	33.97
Gross energy, MJ/kg ^c	423.1738	421.93	421.93	423.47
	1 (2022)	1.1 0.1	17. T 50	

Vitamins and minerals premix detailed by Fath El-Bab et al (2022) a Giving, per kilogram of the mixture: Vitamin E, 5.8 g; vitamin K3, 3.3 g; thiamin, 3.3 g; riboflavin, 6.6 g; pyridoxine (as pyridoxine hydrochloride), 3.3 g; niacin, 16.6 g; folic acid, 3.3 g; vitamin B12 (cyanocobalamin), 0.01 g; D-biotin, 0.1 g; vitamin c (ascorbic acid), 33.3 g; calcium pantothenate, 13.3 g; copper sulfate, 3 g; I, 0.4 g; Co, 0.3 g; Mn, 10 g; zinc oxide, 30 g; sodium selenite, 0.08 g; calcium, 0.8 g. b GE estimates were calculated by multiplying the energy content of the preceding by the protein, fat, and carbohydrate content of feed raw materials: GE is equal to 21.62 kJ/g of protein, 39.52 kJ/g of fat, and 17.2 kJ/g of carbohydrates. c NFE = 100- (CP + EE + CF + Ash).

Table 2. The accession number and particular primer sequences utilized for RT-qPCR analysis.

Tubic 2. The decession name of and particular primer sequences damped for the question many sis.					
Target gene	Forward	Target gene	Forward		
β-actin	GTCATCACCACGACGGACAGG	TTTGCGGTGGACGAGAAGCA	AF384096.1		
IGF1	TGCTGTATCAGTGCGATGCCA	CAGCTTTGGAAGCAGCTCAG	EF563837.1		
HSP70	CGCATCATCAAAATGTTCTGC	TTGTCCAATCCCCAACCTTTA	EU805481.1		
IL-1β	ACAGCACTCTCGGGCTGAACA	GCTCCACCCTCCATTAACACT	115592467		

Table 3. Performance measurements and survival rate for *P. vannamei* when fed diets supplemented with Lpm and/or SeNps.

	Treatments				P-Value
Items	T1 (Control)	T2 (SeNps)	T3 (Lpm)	T4 (SeNps+Lpm)	1 varae
IW (g)	2.44±0.01	2.54±0.009	2.40±0.011	2.43±0.006	0.423
IL (cm)	5.10 ± 0.027	5.09 ± 0.03	5.08 ± 0.036	5.11 ± 0.03	0.928
FW (g)	21.88 ± 0.06^d	26.13 ± 0.14^{c}	28.67 ± 0.20^{b}	29.58 ± 0.13^a	0.0001
FL (cm)	14.50 ± 0.074^{c}	$15.14\pm.06^{b}$	15.52 ± 0.14^{a}	15.71 ± 0.075^a	0.0001
WG(g)	19.44 ± 0.06^{d}	23.58 ± 0.14^{c}	26.28 ± 0.20^{b}	27.15 ± 0.13^a	0.0001
ADG g/d	0.20 ± 0.001^{d}	$0.24 \pm .001^{c}$	0.27 ± 0.002^{b}	0.28 ± 0.001^a	0.0001
SGR $(\%/d)^2$	2.24 ± 0.003^{d}	2.38 ± 0.006^{c}	2.53 ± 0.007^a	2.54 ± 0.01^{a}	0.0001
RGR (g/g)	795.31 ± 0.025^{d}	928.33 ± 0.058^{c}	1094.22 ± 0.086^{b}	1115.60 ± 0.06^{a}	0.0001
SR%	87.5 ± 0.71^{d}	90.00 ± 0.86^{c}	92.5±0.41 ^b	97.50 ± 0.66^{a}	0.023

T1: CTR, P. vannamei group fed basal diet;

IW=Initial weight (g); FW= Final weight (g); WG= Weight gain (g); DG= daily gain; SGR= Specific growth rate (%); RGR = Relative growth rate (g/g); SR%= Survival rate.

Table 4. Feed utilization of *P. vannamei* fed diets supplemented with selenium nanoparticles and/or *Lpm*.

	Ireatments					
Items	T1 (Control)	T2 (SeNps)	T3 (Lpm)	T4 (SeNps+Lpm)	P-Value	
FI	28.42 ± 0.22^{c}	30.27 ± 0.48^{b}	35.54 ± 0.21^{a}	35.012 ± 0.23^{a}	0.0001	
FCR	1.46 ± 0.012^{a}	1.36 ± 0.015^{b}	1.29±0.021°	1.28 ± 0.01^{c}	0.0001	
FE	0.69 ± 0.006^{c}	0.79 ± 0.008^a	0.78 ± 0.015^a	0.74 ± 0.006^{b}	0.047	
PER	1.63±0.014°	1.88±0.019a	1.85±0.036 ^a	1.76±0.018 ^b	0.017	

T1: CTR, P. vannamei group fed basal diet;

a,b,c,d: Values within the same column having different superscripts are significantly different (P < 0.05). Data were presented as the mean \pm mean pool standard error (PSE).

TF=Total consumed feed (g); FCR= Feed conversion ratio (g/g); FE= Feed efficiency; PER= Protein efficiency ratio.

Table 5. Biochemical parameters of *P. vannamei* fed diets supplemented with selenium nanoparticles and/or *Lpm*.

	Treatments				
Items	T1 (Control)	T2 (SeNps)	T3 (Lpm)	T4 (SeNps+Lpm)	P-Value
Glucose mg/dL	59.62 ± 2.22^{b}	69.5±0.89 ^b	77.02 ± 4.12^{a}	70.07 ± 2.49^{a}	0.012
TP (g/dL)	6.65 ± 0.27	6.93 ± 0.5	7.6 ± 0.61	8.32±1.12	0.39
ALB (g/dL)	3.7 ± 0.17	4.19 ± 0.35	4.56 ± 0.33	4.47±0.57	0.436
GLO (g/dL)	2.99 ± 0.10	2.80 ± 0.14	3.04 ± 0.34	3.54 ± 0.59	0.534
AST (UL)	44.41 ± 1.23^a	39.36 ± 0.75^{b}	37.57 ± 0.91^{b}	38.95±0.35 ^b	0.0216
ALT (UL)	78.95 ± 1.349^a	65.89 ± 2.12^{b}	63.82 ± 2.62^{b}	55.53±6.13°	0.001

T1: CTR, *P. vannamei* group fed basal diet; T2: SeNps, *P. vannamei* group fed diet supplemented with .4 ml Selenium nanoparticles per kg; T3: Lpm, *P. vannamei* group fed diet supplemented with 400mg Lpm per kg; T4: SeNps+Lpm, *P. vannamei* group fed diet supplemented with 0.4 ml SeNps= 400 mg Lpm per kg; a,b,c,d: Values within the same column having different superscripts are significantly different (P < 0.05). Data were presented as the mean ± mean pool standard error (PSE). TP= total protein; ALB= albumin; GLOB= globulin; ALT= alanine transaminase; AST=aspartate aminotransferase.

T2: SeNps, P. vannamei group fed diet supplemented with .4 ml Selenium nanoparticles per kg;

T3: Lpm, P. vannamei group fed diet supplemented with 400mg Lpm per kg;

T4: SeNps+Lpm, P. vannamei group fed diet supplemented with 0.4 ml SeNps= 400 mg Lpm per kg;

a,b,c,d: Values within the same column having different superscripts are significantly different (P < 0.05). Data were presented as the mean \pm mean pool standard error (PSE).

T2: SeNps, P. vannamei group fed diet supplemented with .4 ml Selenium nanoparticles per kg;

T3: Lpm, P. vannamei group fed diet supplemented with 400mg Lpm per kg;

T4: SeNps+Lpm, P. vannamei group fed diet supplemented with 0.4 ml SeNps= 400 mg Lpm per kg;

Table 6. Antioxidant activities and immune responses of *P. vannamei* fed diets supplemented with selenium nanoparticles and/or *Lpm*.

		Treatments			
Items	T1 (Control)	T2 (SeNps)	T3 (Lpm)	T4 (SeNps+Lpm)	P-Value
MDA (IU/L)	3.77 ± 0.60	3.55±0.53	3.21±0.61	2.82±0.54	0.681
TAC (IU/L)	19.94 ± 0.67^{ab}	17.44 ± 0.68^{b}	23.26 ± 0.99^a	20.86 ± 1.52^{ab}	0.024
SOD (IU/L)	22.07 ± 1.73^{b}	28.41 ± 0.93^{ab}	26.65 ± 3.29^{ab}	30.66 ± 1.30^{a}	0.081
CAT (IU/L)	3.00 ± 0.15^{b}	3.85 ± 0.13^{ab}	4.39 ± 0.28^{a}	3.71 ± 0.64^{ab}	0.138

T1: CTR, P. vannamei group fed basal diet;

IgG= Immunoglobulin G; IgA= Immunoglobulin A; MDA=malonaldehyde; TAC= Total antioxidant capacity; SOD=super oxide dismutase, CAT=catalas

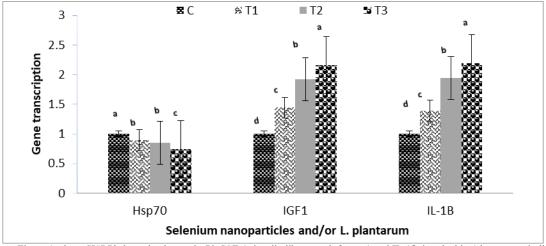


Figure 1: shownHSP70: heat shock protein 70; IGF-1: insulin like growth factor-1 and IL-1β: interleukin-1 beta genes in livers of *P. vannamei* fed diets supplemented with SeNps and/or Lpm for at 14 weeks.

Furthermore, Se, an essential component of GPx, may increase antioxidant activity by shielding cell membranes from ROS. This could enhance the immunocompetence, stress tolerance. development of aquatic species (Lin and Shiau, 2005; Tian et al., 2014; Fan et al., 2022). When different forms of selenium were added to their diets, a number of farmed crustacean species, including giant river prawns, Chinese mitten crabs, Chinese white shrimp cherry shrimp, and oriental river prawns, according to (Chiu et al., 2010; Tian et al., 2014; Qiang et al., 2020; Wang et al., 1994; Wang et al., 2009; Kong et al., 2017), respectively, demonstrated improved growth performance. Many people believe that feed additives like probiotics and nanoparticles are the main source of stimulation for the blood biochemical variables of aquatic animals (Femi-Oloye et al., 2020). The best dosage for raising blood protein in this case was found to be 650 mg Lac/kg of diet, indicating the advantageous function of probiotics in preserving shrimp immunity (Dawood et al., 2019). When P. vannamei are exposed to aquatic stress, both ALT and AST levels are notably raised, which usually leads to hepatopancreatic damage (Naiel et al., 2019; Naiel et al., 2024). In the current study, all groups supplemented with SeNps and/or Lpm showed a considerable suppression of AST and ALT levels. Furthermore, when comparing the ALT and AST levels of Nile tilapia to the control group, Soltan and El-Laithy (2008). found that probioticsupplemented meals significantly reduced the levels of these enzymes and clearly showed that increasing the amount of probiotic in diets, especially at a concentration of 600 mg/kg diet, decreased the activities of ALT and AST. Since TG stores extra calories and provides the fish body with energy. Fath El-Bab et al. (2022), reported that the significant rise in TG seen in this study in response to the dietary addition of Lpm is compelling evidence of the beneficial effects of probiotic supplementation on lipid metabolism. The antioxidant defense system, which is maintained by antioxidant state, is intimately related to immunological condition and physiological activities in fish tissue (Hoseinifar et

T2: SeNps, P. vannamei group fed diet supplemented with .4 ml Selenium nanoparticles per kg;

T3: Lpm, P. vannamei group fed diet supplemented with 400mg Lpm per kg;

T4: SeNps+Lpm, P. vannamei group fed diet supplemented with 0.4 ml SeNps= 400 mg Lpm per kg;

a,b,c,d: Values within the same column having different superscripts are significantly different (P < 0.05). Data were presented as the mean \pm mean pool standard error (PSE).

al. 2020). In this investigation, the addition of SeNps and/or Lpm decreased MDA evels while increasing total antioxidant capacity. Furthermore, the activity of SOD and CAT increased and peaked at T4. These results were consistent with those of Yanez-Lemus et al. (2022), who found that the MDA content was lowest and the SOD and CAT levels were highest in Rainbow Trout diets supplemented with Lpm, a Selenium Nanoparticle-Enriched and Potential Probiotic, compared to the control group. Additionally, Yang et al. (2010), demonstrated a significant increase in the SOD and CAT activities of shrimp fed diets treated with Rhodosporidium paludigenum yeast. Regardless of the kind of dietary selenium, it changes into selenocysteine (Sec) to be incorporated into selenoproteins with antioxidant To incorporate Se as Sec into a properties. selenoprotein, a specific mechanism called the Sec insertion sequence must decode the UGA codon in the 3'-untranslated section mRNA (Zoidis et al., 2018). Dietary trace elements have demonstrated to either increase or decrease the activity of antioxidant enzymes in decapod species. Additionally, decapod crustaceans employ a variety of antioxidant defense strategies (Frías-Espericueta et al., 2022). Adding different forms of selenium to the diet has been shown to increase the activities of antioxidant enzymes (e.g., CAT, SOD, and GPx) in a variety of crustacean species, such as P. vannamei (0.81 mg/kg, sodium selenite, Yu et al., 2021), Se-N (Qin et al., 2016), Se-bio-fortified corn (Yuan et al., 2018), and Se-methionine (0.4 mg/kg, Yu et al., 2022) in Chinese Mitten Crab; sodium selenite (1 mg/kg, Chiu et al., 2010); and organic selenium (0.82 mg/kg, Qiang et al., 2020) in giant freshwater prawn and sodium selenite (0.45 mg/kg, Wang et al., 2009) in cherry shrimp. In a different study, Yu et al. (2022) found that feeding P. vannamei shrimp a diet containing 0.4 mg/kg Se-N and organic selenium rather than sodium selenate enhanced their GPx and SOD activity. Ibrahim et al. (2021) assert that cytokines, such as $IL-1\beta$, are crucial for regulating the immune response. Numerous studies have shown that their expression can be utilized to predict variations in immune response (Dawood et al., 2017 and Dawood et al., 2020b). Both SeNps and Lpm have immunomodulatory effects that result from the interaction of beneficial microbial cells with intestinal epithelial cells. This interaction may enhance mucosal immunity in the gut by producing strengthening epithelial junctions, mucosal immunoglobulins, producing antimicrobial peptides, controlling inflammatory responses, and ultimately increasing the immune response (Daming et al., 2003). In this study, a dietary combination of

SeNps and Lpm significantly improved IL-1\beta regulation compared to other treatment groups. It has been demonstrated that probiotics enhance proinflammatory cytokine transcription (Hasan et al., 2019). Additionally, SeNps upregulates zebrafish IL-1β (Soltani et al., 2019). Meanwhile, gene analysis revealed that probiotic-fed O. niloticus exhibited increased levels of interleukin-1 beta (IL-1β) (Omar et al., 2024). Elevated immunity is linked to higher *IL-Iβ* transcription in sea bream grown in specific conditions. This overexpression in response to diets supplemented with SeNps further highlights the ability of selenium nanoparticles to reduce inflammation by attracting and activating neutrophils in affected areas, thereby enhancing the immune response and overall health of aquatic animals (Enferadi et al., 2018). Heat shock protein 70 (HSP70) is often expressed by aquatic animal cells in response to environmental stimuli (Ming et al., 2010). Elevated serum levels of cortisol and glucose have been linked to unfavorable rearing conditions. This may cause muscle and liver tissues to undergo glycolysis, producing glucose as an energy source for stressed fish (Dawood et al., 2020a). It was shown that dietary Se deficit (0.05 mg/kg) markedly increased HSP70 in order to shield the Chinese mitten crab from oxidative stress This implies that Se may impact the regulation of HSP70 expression (Qiang et al., 2020). Another study found no effect of dietary Se on seabream HSP70 gene expression, suggesting that HSP70 expression may differ by species and be impacted by culture conditions. Similarly, Penglase et al. (2010) discovered that the HSP70 expression of cod (Gadus morhua L.) larvae were unaffected by Se-enriched rotifers. HSP70 transcription was also shown to be lower in P. vannamei shrimp fed diets containing a combination of SeNPs and Lpm. This further supports the idea that both Lpm and SeNPs work in concert to control the overall shrimp response to ecological stressors This implies that Se may impact the regulation of HSP70 expression (Qiang et al., 2020).

CONCLUSION

Overall, our findings demonstrate the potential application of SeNp, either alone or in combination with *L. plantarum*, for the rearing of *P. vannamei* shrimp. Dietary supplements containing SeNps and/or *L. plantarum* have been shown to enhance growth performance and alter several relevant hemolymph biochemical parameters and resistance to diseases. Thus, this SeNps may be utilized as a feed supplement in *P. vannamei* aquaculture to improve growth performance and lower illness during the rearing period, either alone or in combination with *L. plantarum*.

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The authors declare no conflict of interest associated with the paper. The authors alone are responsible for the content and writing of this article.

AUTHORS CONTRIBUTION

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الملخص العربى

التأثيرات المشتركة لجسيمات النانو سيلينيوم وبكتيريا اللاكتوباسيلوس بلانتاروم على أداء نمو الجمبري أبيض الساق ايمان علاء الجندى - إبراهيم عطا أبو النصر و أحمد فاروق فتح الباب قسم الانتاج الحيواني و الداجني و السمكي - كلية الزراعة - جامعة دمياط

يُنصح بشدة باستخدام البروبيوتيك والجسيمات النانوية في تربية الأحياء المائية؛ ولا يُعرف الكثير عن تأثيراتها التآزرية. لذا، الجريت هذه الدراسة لتقييم التأثيرات التآزرية المحتملة لجسيمات النانو سيلينيوم وبكتيريا اللاكتوباسيلوس بلانتاروم على أداء النمو، ونسبة تحويل العلف، والكيمياء الحيوية في المصل، ونشاط مضادات الأكسدة في الروبيان الفائمي أبيض الساق. تم إعداد ثلاث علائق تجريبية مضاف اليها جسيمات النانوسيلينيوم وبكتيريا اللاكتوباسيلوس بلانتاروم أو كليهما بالإضافة الى العليقة ضابطة. وقد أظهرت النتائج أن الروبيان الذي تغذى على علائق تحتوى على الإضافة الغذائية كانت ذات اعلى معدل وزن جسم نهائي والزيادة في الوزن ومعدل النمو النوعي مقارنة بالعليقة الضابطة، مع ملاحظة ان اقل قيم لمعدل تحويل الغذائي للروبيان الذي تغذى على عليقة تحتوى على كل من جسيمات النانوسيلينيوم وبكتيريا اللاكتوباسيلوس بلانتاروم الى العلائق مقارنة بالمعاملات. كما تأثرت انزيمات الكبي المحتوعة الضابطة. كما عليه المحموعة المحموعة الضابطة وجبات أنشطة السعة الكلية لمضادات الأكسدة و سوبر أكسيد ديسميوتاز وانزيم الكتاليز أعلى بشكل ملحوظ في المجموعة التي تناولت وجبات أشطة السعة الكلية لمضادات الأخرى. كما ارتفعت قيم جينات بروتين الصدمة الحرارية، شبيه الانسولين وانتركولين 1 بيتا بشكل ملحوظ في مجموعة الجبري النوسيلينيوم. ومع ذلك، ارتفعت مستويات المالونديالدهيد في المجموعة في مجموعة الحمبري التي تغذت على وجبات مُكملة بمزيج من بكتريا اللاكتوباسيلاس بالانتيرم وجسيمات النانوسيلينيوم..