

The Influence of *Panax ginseng* on Growth Performance, Innate Immune Responses, and Microbiome Characteristics of *Penaeus vannamei*

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Received: 25 August 2024; Revised: 22 November 2024; Accepted: 24 November 2024

ABSTRACT

In the field of aquaculture, various feed additives have been investigated. *Panax ginseng* Meyer, a traditional herbal supplement, has shown promising results across multiple farmed aquatic species. However, studies on its application in *Penaeus vannamei*, a major aquaculture shrimp species, are still scarce. This research examined the impacts of *P. ginseng* on *P. vannamei*, focusing on growth performance, innate immune parameters, and the intestinal microbiome. Juvenile shrimp received standard commercial diets supplemented with red ginseng extract at five levels (0.00%, 0.05%, 0.10%, 0.50%, and 1.00%) over a 6-week period. Body weight measurements were taken on days 21 and 42. On day 42, three shrimp from each group were sampled for additional assessments. In the growth trial, the 0.10% group demonstrated markedly better final body weight (FBW), weight gain (WG), specific growth rate (SGR), and feed conversion ratio (FCR) than the 0.00% control group by day 42. Quantitative PCR analysis revealed significantly elevated IGF-BP gene expression in the 0.05%, 0.10%, and 1.00% groups relative to the control. For innate immunity, superoxide dismutase (SOD) activity was notably higher in the 0.05% and 0.50% groups compared to the control. Microbiome profiling indicated that the 0.10% group had increased abundance of Flavobacteriaceae and reduced Vibrionaceae at the family level versus the control. At the genus level, this group showed higher levels of unclassified Flavobacteriaceae and lower *Vibrio* compared to the 0.00% group. Dietary inclusion of *P. ginseng* improved growth performance, immune function, and microbial community structure in *P. vannamei*. Additional investigations into optimal dosage and potential use of red ginseng by-products may further enhance productivity and economic viability in shrimp farming.

Keywords: Aquaculture, Feed additive, Microbiome analysis, Red ginseng, Shrimp

How to Cite This Article: Hariri Y, Nasser H, Zahra F. The Influence of *Panax ginseng* on Growth Performance, Innate Immune Responses, and Microbiome Characteristics of *Penaeus vannamei*. Spec J Pharmacogn Phytochem Biotechnol. 2024;4(2):93-102. <https://doi.org/10.51847/IITukUle8X>

Introduction

Aquaculture represents one of the fastest-growing areas in worldwide food production, surpassing capture fisheries in output [1]. Marine and freshwater products currently account for 17% of global animal protein supply and contribute more than half of animal protein intake in certain developing nations [1]. As consumer demand continues to rise, there is increasing emphasis on improving growth rates and overall production efficiency. Feed additives are commonly employed in aquaculture to promote better growth and deliver additional advantages [2]. Ginseng refers to several medicinal plants belonging to the Araliaceae family. *Panax ginseng* Meyer stands out as one of the most recognized and thoroughly researched varieties, with a long history of traditional medicinal use in East Asia, especially Korea, spanning thousands of years. It is widely valued for its ability to support cognitive performance [3, 4], enhance physical endurance [5], and strengthen immune responses [6]. The primary bioactive constituents responsible for these effects are ginsenosides, a class of steroid glycosides and triterpene saponins. Korean ginseng has been reported to contain 38 distinct ginsenosides [7]. In various animal studies, ginsenosides

have been shown to stimulate macrophage phagocytosis [8, 9], improve learning and memory in rodents [10, 11], promote blood vessel dilation [12, 13], and bolster resilience to external stressors [14].

The value of *P. ginseng* as a dietary supplement has been established in several aquaculture species. For example, its inclusion in the diet of Nile tilapia (*Oreochromis niloticus*) resulted in substantially higher growth rates [15]. African catfish (*Clarias gariepinus*) fed ginseng-supplemented diets exhibited significant improvements in weight gain and feed utilization efficiency [16]. In goldfish (*Carassius auratus*), different levels of ginseng extract in the feed produced a relationship between specific growth rate (SGR) and the logarithm of the extract concentration [17].

The Pacific white shrimp, *Penaeus vannamei*, is among the most economically important species in global aquaculture. Nevertheless, information on the influence of *P. ginseng* on this species remains limited. Therefore, the objective of the present study was to evaluate the potential positive effects of *P. ginseng* on the growth, innate immunity, and gut microbiota of *P. vannamei*. Assessments included growth indices such as SGR, feed conversion ratio (FCR), and protein efficiency ratio (PER), quantification of insulin-like growth factor binding protein (IGF-BP) expression, measurement of innate immune markers including phenoloxidase (PO) and superoxide dismutase (SOD) activities, and analysis of the microbial community. This work highlights the practical use of the natural additive *P. ginseng* to support shrimp health and growth, thereby contributing to sustainable and high-quality seafood production in the aquaculture sector.

Materials and Methods

Ethics statement

The experimental procedures were reviewed and approved by the Institutional Animal Care and Use Committee (IACUC) of Kyungpook National University (approval number 2023-0220). All measures were taken to ensure animal welfare and reduce any potential distress.

Ginseng extract and experimental shrimp feed

Red ginseng extract (40% moisture content) was supplied by the Korean Society of Ginseng for use as a feed additive. The extract contained 11 ginsenosides, including the six major ones (Rb1, Rb2, Rc, Rd, Re, and Rg1). Microbiological testing confirmed the absence of bacteria and fungi. The extract was diluted with distilled water to prepare five concentrations: 0.00%, 0.05%, 0.10%, 0.50%, and 1.00%. Each dilution was combined in a 1:1 weight ratio with commercial shrimp feed. The resulting mixtures were dried completely and kept refrigerated at 4°C until needed.

Feeding trial

A total of 225 juvenile *P. vannamei* shrimp (initial average body weight 0.32 g) were randomly assigned to five treatment groups corresponding to the red ginseng extract supplementation levels (0.00% control, 0.05%, 0.10%, 0.50%, and 1.00%). Each group consisted of three replicate tanks with 15 shrimp per tank. The 22 L tanks contained 25 ppt seawater with continuous aeration and a constant temperature of 28°C. Water was exchanged at 50% every four days. Shrimp were fed three times daily at a rate of 5% of body weight (initially calculated from starting weight and expected growth). Feed amounts were adjusted on day 21 following body weight re-measurement. Shrimp health was monitored daily, and any mortalities were removed promptly. The trial lasted 42 days.

Growth parameters were evaluated on days 21 and 42. Final body weight (FBW), weight gain (WG), specific growth rate (SGR), feed conversion ratio (FCR), and protein efficiency ratio (PER) were calculated using the formulas:

$$\text{Weight gain (\%)} = [(\text{final mean body weight} - \text{initial mean body weight}) / \text{initial mean body weight}] \times 100 \quad (1)$$

$$\text{Specific growth rate (\%)} = 100 \times [\ln(\text{final body weight}) - \ln(\text{initial body weight})] / \text{days [18]}. \quad (2)$$

$$\text{Feed conversion ratio} = \text{dry feed fed} / \text{wet weight gain (g) [19]}. \quad (3)$$

$$\text{Protein efficiency ratio} = \text{wet weight gain (g)} / \text{total protein fed [20]}. \quad (4)$$

At the end of the 6-week trial, three shrimp per tank (nine per group, total N = 45) were randomly selected and humanely euthanized. Samples of hepatopancreas, hemolymph, and intestine were collected for subsequent analyses.

Quantitative analysis of growth gene expression

Hepatopancreas samples were used to measure growth-related gene expression via quantitative real-time PCR (qPCR), targeting the IGF-BP gene. Total RNA was isolated from approximately 50 mg of tissue using the RNeasy Micro kit (cat. no. 74004; Qiagen, Valencia, CA, USA). First-strand cDNA was synthesized with AccuPower® RocketScript™ Cycle RT PreMix (cat. no. K-2201; Bioneer). qPCR was conducted on the StepOnePlus real-time PCR system (ThermoFisher Scientific, Waltham, MA, USA) using β -actin as the internal reference gene. Primer sequences are listed in **Table 1**.

Table 1. The primers used in the growth gene analysis and microbiome profiling.

Name	Primer sequence (5'–3')	Description	Reference
β-actin F	GAGCAACACGGAGTTCGTTGT	Housekeeping gene	[21]
β-actin R	CATCACCAACTGGGACGACATGGA		
IGF-BP F	GTGGGCAGGGACCAAATC	IGF-BP expression	[22]
IGF-BP R	TCAGTTACCACCAGCGATT		
16s rRNA_515F	GTGCCAGCMGCCGCGGTAA	16S rRNA V4 region	[23]
16s rRNA_806R	GGACTACHVGGGTWTCTAAT		

Biochemical analysis of innate immunity

Evaluation of innate immune responses involved determining phenoloxidase (PO) activity following the procedure reported by Hernandez *et al.* [24]. In brief, hemolymph samples were collected from the shrimp via a 27-gauge 1 mL syringe that contained Alsever's solution to prevent clotting. The mixture was centrifuged at 800×g for 20 minutes. The pellet obtained was washed with the anticoagulant and then resuspended in PBS. Fifty microliters of this cell suspension were combined with 50 μ L of trypsin solution (0.1 mg/mL prepared in cacodylate buffer at pH 7.0) and allowed to incubate for 10 minutes at 25°C. Subsequently, 50 μ L of L-3,4-dihydroxyphenylalanine (L-DOPA) at 3 mg/mL in the same cacodylate buffer was introduced, followed by another 10-minute incubation at 25°C. The resulting PO activity was quantified by recording absorbance readings at 425 nm with a spectrophotometer.

Antioxidant defense was assessed by measuring superoxide dismutase (SOD) activity present in the hemolymph samples. This was carried out using the EZ-SOD assay kit (cat. no. DG-SOD400; DoGenBio, Seoul, South Korea) in accordance with the provided manufacturer guidelines.

Microbiome profiling

The structure of the gut microbial community in the shrimp was investigated. Total DNA was obtained from pooled intestinal samples collected from three shrimp per replicate tank employing the DNeasy Blood & Tissue kit (cat. no. 69504; Qiagen, Valencia, CA, USA). Amplification via PCR focused on the V4 region of the bacterial 16S rRNA gene, with the specific primers detailed in **Table 1**. Library preparation and paired-end sequencing of the amplicons were performed on the Illumina MiSeq sequencer (Illumina, San Diego, CA, USA). Bioinformatics processing of the raw sequencing data, including quality filtering and analysis, utilized the QIIME 2 software suite [25]. Amplicon sequence variants (ASVs) were identified from the processed reads. Alpha diversity indices, reflecting species richness and distribution within individual samples, were computed using the Shannon and Chao1 metrics. Beta diversity, indicating compositional differences across samples, was evaluated based on Bray-Curtis dissimilarity distances. These patterns were visualized using both Principal Coordinates Analysis (PCoA, also termed metric multidimensional scaling) and non-metric multidimensional scaling (NMDS). Taxonomic assignment of the ASVs was achieved through comparison with established reference databases.

Statistical analysis

Experimental results are reported as means $\pm$ standard deviation (SD). Statistical comparisons were conducted with repeated-measures analysis of variance (ANOVA). When the overall ANOVA yielded a significant F-value ($P < 0.05$), pairwise differences between group means were examined using Tukey's multiple comparison test.

Results and Discussion

Feeding trial and growth metrics calculation

The shrimp maintained good health conditions over the entire 42-day experimental period, displaying typical feeding activity and normal swimming behavior. No notable variations in daily mortality rates were observed among the five dietary treatment groups. Body weight assessments occurred at the end of week 3 (day 21) and week 6 (day 42). On day 21, the groups receiving 0.10% and 1.00% supplementation exhibited significantly superior ($P < 0.05$) values for final body weight (FBW), weight gain (WG), specific growth rate (SGR), feed conversion ratio (FCR), and protein efficiency ratio (PER) in comparison with the unsupplemented 0.00% control. By day 42, the 0.10% group continued to demonstrate significantly better ($P < 0.05$) performance in FBW, WG, SGR, and FCR relative to the control group, although PER did not show statistically meaningful differences across treatments. Complete data on these growth parameters along with survival percentages at both time points are compiled in **Tables 2A, 2B**.

Table 2. The FBW, WG, SGR, FCR, PER values, and the survival rate of *P. vannamei* after (A) 3 weeks and (B) 6 weeks of the experiment. 2A.2B.

A						
Group	FBW	WG	SGR	FCR	PER	Survival (%)
0.00 %	0.77 $\pm$ 0.05 ^{bc}	141 $\pm$ 14.5 ^{bc}	4.40 $\pm$ 0.30 ^{bc}	1.22 $\pm$ 0.12 ^{ab}	2.07 $\pm$ 0.22 ^{bc}	95.6 $\pm$ 3.85
0.05 %	0.85 $\pm$ 0.00 ^{ab}	165 $\pm$ 1.38 ^{ab}	4.88 $\pm$ 0.03 ^{ab}	1.03 $\pm$ 0.01 ^{bc}	2.42 $\pm$ 0.03 ^{ab}	91.1 $\pm$ 10.2
0.10 %	0.90 $\pm$ 0.04 ^a	180 $\pm$ 11.5 ^a	5.15 $\pm$ 0.21 ^a	0.95 $\pm$ 0.07 ^c	2.64 $\pm$ 0.19 ^a	91.1 $\pm$ 3.85
0.50 %	0.75 $\pm$ 0.05 ^c	135 $\pm$ 15.5 ^c	4.26 $\pm$ 0.33 ^c	1.28 $\pm$ 0.15 ^a	1.95 $\pm$ 0.24 ^c	95.6 $\pm$ 7.70
1.00 %	0.87 $\pm$ 0.06 ^a	171 $\pm$ 19.2 ^a	4.98 $\pm$ 0.36 ^a	1.00 $\pm$ 0.13 ^c	2.52 $\pm$ 0.30 ^a	91.1 $\pm$ 3.85
B						
Group	FBW	WG	SGR	FCR	PER	Survival (%)
0.00 %	1.75 $\pm$ 0.19 ^b	448 $\pm$ 58.6 ^b	4.14 $\pm$ 0.26 ^b	1.11 $\pm$ 0.13 ^{ab}	2.27 $\pm$ 0.28	82.2 $\pm$ 10.2
0.05 %	1.83 $\pm$ 0.04 ^b	471 $\pm$ 12.6 ^{ab}	4.25 $\pm$ 0.05 ^{ab}	1.10 $\pm$ 0.08 ^{ab}	2.28 $\pm$ 0.17	73.3 $\pm$ 6.67
0.10 %	2.02 $\pm$ 0.10 ^a	529 $\pm$ 32.7 ^a	4.48 $\pm$ 0.13 ^a	1.06 $\pm$ 0.12 ^c	2.37 $\pm$ 0.26	75.6 $\pm$ 3.85
0.50 %	1.64 $\pm$ 0.02 ^b	413 $\pm$ 5.67 ^b	3.99 $\pm$ 0.03 ^b	1.27 $\pm$ 0.13 ^a	1.96 $\pm$ 0.21	86.4 $\pm$ 10.2
1.00 %	1.76 $\pm$ 0.06 ^b	449 $\pm$ 22.1 ^b	4.15 $\pm$ 0.10 ^b	1.17 $\pm$ 0.05 ^{ab}	2.13 $\pm$ 0.10	82.2 $\pm$ 7.70

- 1) All data represent the average of triplicate groups and are shown as mean $\pm$ standard deviation (SD).
- 2) Within each column, values bearing different superscript letters differ significantly ($P < 0.05$).
- 3) The absence of superscript letters signifies that no statistically significant differences were detected among the treatment groups.

Quantitative analysis of growth gene expression

Samples of hepatopancreas tissue were harvested from nine individuals in every treatment group, amounting to a total of 45 shrimp across all five dietary regimes. After isolating total RNA from these hepatopancreas samples, reverse transcription was performed to produce cDNA, which was subsequently analyzed using real-time quantitative PCR (qPCR) to measure expression of the IGF-BP gene. By the end of the sixth week, IGF-BP transcript levels were significantly upregulated ($P < 0.05$) in the 0.05%, 0.10%, and 1.00% supplementation groups relative to the 0.00% control. Normalized relative expression values for the IGF-BP gene are displayed in **Figure 1**.

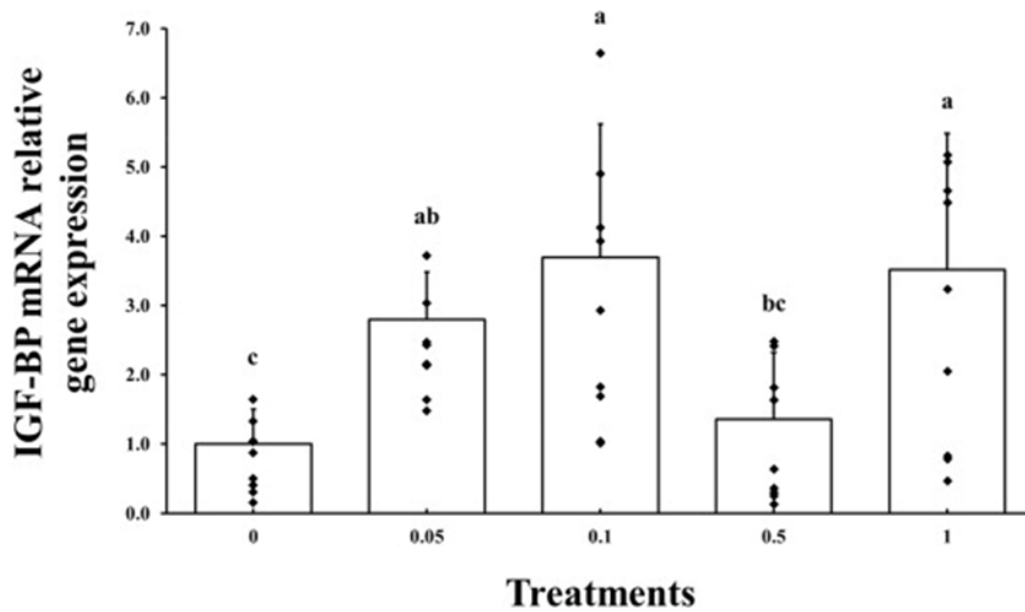


Figure 1. Relative mRNA expression levels of insulin-like growth factor binding protein (IGF-BP) in the hepatopancreas following 6 weeks of dietary supplementation. Columns labeled with different letters indicate statistically significant differences ($P < 0.05$).

Biochemical analysis of innate immunity

Hemolymph samples collected from the shrimp were used to evaluate key innate immune parameters, focusing on phenoloxidase (PO) and superoxide dismutase (SOD) activities. At the end of week 6, PO activity levels remained statistically similar across all treatment groups. In contrast, SOD activity was significantly enhanced ($P < 0.05$) in the 0.05% and 0.50% supplementation groups compared with the 0.00% control group. Detailed PO and SOD activity results for *P. vannamei* after the 6-week feeding trial are summarized in **Table 3**.

Table 3. The PO and SOD activity of *P. vannamei* measured spectrophotometrically after 6 weeks of the experiment.

Group	PO	SOD
0.00 %	0.122 ± 0.01	80.7 ± 8.08 ^b
0.05 %	0.134 ± 0.01	92.6 ± 4.71 ^a
0.10 %	0.130 ± 0.01	87.5 ± 3.21 ^{ab}
0.50 %	0.131 ± 0.00	92.4 ± 3.16 ^a
1.00 %	0.124 ± 0.01	87.0 ± 5.98 ^{ab}

- 1) All data represent the average of triplicate groups and are shown as mean ± standard deviation (SD).
- 2) Within each column, values bearing different superscript letters differ significantly ($P < 0.05$).
- 3) The absence of superscript letters signifies that no statistically significant differences were detected among the treatment groups.

Microbiome analysis

Examination of the gut bacterial community composition at the family taxonomic rank showed that shrimp fed the 0.10% red ginseng extract diet had a substantially elevated proportion of Flavobacteriaceae together with a notably diminished presence of Vibrionaceae compared to the unsupplemented control group. At the genus level, the 0.10% treatment likewise resulted in a significantly increased abundance of unclassified Flavobacteriaceae and a clear reduction in *Vibrio* relative to the control. Abundances of all remaining bacterial groups remained statistically comparable across the various dietary treatments at both family and genus classifications. Community profiles at these taxonomic levels are displayed in **Figures 2a, 2b**.

Analysis of alpha diversity metrics pointed to small, non-significant variations in microbial richness and evenness at the genus level among the different feeding regimes (data not shown). Beta diversity assessment similarly found

no statistically meaningful separation in bacterial genus-level community structure between the treatment groups (data not shown).

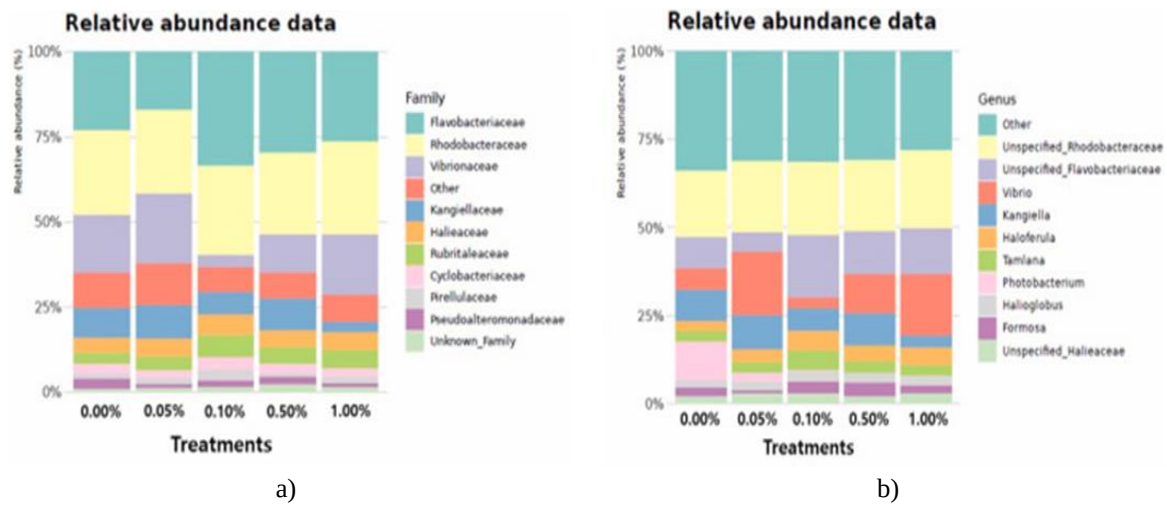


Figure 2. The bacterial community structure of the (a) family and (b) genus level.

Growth performance receives considerable attention in aquaculture because it directly determines overall production yield and economic outcomes. Researchers typically assess growth using multiple standardized indices. One of the most commonly applied is the specific growth rate (SGR), which quantifies the relative increase in body weight per unit of time [18] and is routinely cited in scientific publications [26-28]. Additional widely used metrics include the feed conversion ratio (FCR), which indicates the efficiency with which feed is transformed into body tissue [19], and the protein efficiency ratio (PER), which evaluates how effectively dietary protein is converted into weight gain [20]. Expression of the insulin-like growth factor binding protein (IGF-BP) gene is also frequently utilized as a molecular marker in growth-related studies [29, 30]. IGF-BP modulates insulin-like growth factor (IGF) activity, a critical regulator involved in numerous physiological pathways [31, 32]. In crustaceans, extensive research has documented the varied roles of IGF-BP and its homologs, including control of androgenic hormone signaling [33, 34], regulation of ovarian development [35, 36], modulation of immune responses [37, 38], and influence on somatic growth [39]. Consequently, the current investigation primarily sought to determine the effectiveness of *P. ginseng* as a dietary additive for *Penaeus vannamei* through the application of these established evaluation methods.

Feed additives are actively investigated in aquaculture as tools for promoting enhanced growth. These additives encompass a diverse range of substances, such as probiotics, prebiotics, synbiotics, organic acids, and medicinal herbs [40]. Among herbal options, *P. ginseng* has exhibited beneficial effects in several other farmed aquatic species, including Nile tilapia (*Oreochromis niloticus*) [15], African catfish (*Clarias gariepinus*) [16], and goldfish (*Carassius auratus*) [17]. In the present shrimp feeding trial, incorporation of red ginseng extract into the diet resulted in significant improvements in overall growth indices for both the 0.10% and 1.00% groups at week 3 (including elevated PER), and continued advantages for the 0.10% group at week 6, although no significant PER differences were observed at the later stage. These findings indicate that an optimal inclusion level of ginseng in the feed can effectively support better shrimp growth. Furthermore, ginseng supplementation appeared to enhance PER during the initial feeding period; although this effect may lessen over extended durations, it does not appear to compromise the nutritional quality of protein in the diet. In addition, the IGF-BP gene expression results corresponded closely with the growth performance data, as the groups that achieved significantly higher SGR (0.10% and 1.00%) also displayed significantly elevated IGF-BP expression levels compared to the 0.00% control. The 0.50% group showed no statistically significant difference from the negative control in IGF-BP expression. Antioxidant enzymes are commonly employed as indicators of innate immune function [41]. Superoxide dismutase (SOD) contributes by catalyzing the conversion of reactive oxygen species (ROS) into hydrogen peroxide [41], allowing subsequent membrane transit. Although ROS possess strong microbicidal activity against external pathogens, they can also induce oxidative damage to the host's own biomolecules. SOD therefore plays an important role in regulating innate immunity while supporting the enzymatic antioxidant system that protects tissues from free radical-induced harm. Previous studies in *P. vannamei* have shown that external stimuli can

elevate SOD levels [42], which is interpreted as a sign of innate immune system activation. In this experiment, following administration of red ginseng extract, the 0.05% and 0.50% groups demonstrated significantly higher SOD activity than the 0.00% control group, indicating an immunostimulatory benefit. Phenoloxidase (PO) activity has similarly been used as a key parameter in invertebrate immune research [43]. Upon activation, PO is released from hemocytes and participates in melanin synthesis, which facilitates pathogen melanization [44]. Interestingly, the current study found no significant differences in PO activity between any of the experimental groups and the negative control. This outcome suggests that the shrimp immune system did not recognize the ginseng additive as a foreign threat. It also rules out the possibility that the observed increase in SOD activity resulted from non-specific exogenous stimulation caused by the ginseng itself.

The term “microbiome” denotes the entire community of microorganisms and their genetic components within a specific environment [44]. Strong associations between microbial communities and their animal hosts have been well documented in numerous investigations [45-47]. In the present study, dietary supplementation with red ginseng extract led to a significantly lower *Vibrio* abundance at both family and genus levels in the 0.10% group, reflecting a beneficial modification of the gut microbiota. Reducing *Vibrio* populations has remained a major goal in shrimp aquaculture due to their association with severe disease outbreaks. Acute hepatopancreatic necrosis disease, a particularly destructive form of vibriosis triggered by specific toxin-producing *Vibrio* strains, is characterized by its sudden onset and high lethality [48]. Thus, the reduction in *Vibrio* load observed in the microbiome represents a favorable outcome. However, whether this shift is a direct result of the ginseng additive, a secondary effect of enhanced immunity, or other unidentified mechanisms requires further clarification. Additionally, Flavobacteriaceae species are commonly present throughout shrimp development, constitute part of the core intestinal microbiome in *P. vannamei*, and are generally regarded as having minimal pathogenicity toward the host [49]. Despite some uncertainty surrounding the observed microbiome changes, they do not appear to present any obvious risks.

A key challenge concerning the practical application of red ginseng extract in commercial shrimp farming is its cost. One promising approach to overcome this limitation involves the use of red ginseng residues generated during the extraction process. These residues retain valuable nutritional components, including carbohydrates, essential amino acids, dietary fibers, micronutrients, and substantial levels of ginsenosides [50, 51]. Korea produces more than 1,000 tons of red ginseng residues each year, yet the majority are currently discarded through burning or landfilling [52]. Utilizing these byproducts could contribute significantly to resource recycling and the development of high-value materials. Moreover, the high carbohydrate content (approximately 70%) in the residues may offer additional advantages. Biofloc technology, which enhances water quality through the proliferation of heterotrophic bacteria, is readily stimulated by carbohydrate supplementation [53]. Carbohydrates serve as an energy source that enables these microbes to immobilize nitrogenous waste. Given the carbohydrate richness of red ginseng residues, they are expected to support biofloc formation [53], while also providing the previously mentioned benefits associated with ginsenosides.

Furthermore, because ginseng is a natural herbal product, it is not anticipated to exhibit toxic effects, as confirmed in this study. Survival rates showed no significant differences between the 0.00% control group and any of the supplemented groups, demonstrating that ginseng does not cause notable toxicity in shrimp. This result is consistent with earlier findings that ginseng supplementation can exert multiple biological effects in *P. vannamei*, including immune enhancement, without detectable adverse side effects [54].

Collectively, the present study illustrates the potential of *P. ginseng* to improve growth, innate immunity, and gut microbiome composition in *P. vannamei*. Specifically, after six weeks of feeding, the 0.10% group displayed significant growth enhancement relative to the negative control. The 0.50% group exhibited improved SOD activity, and the 0.10% group showed positive microbiome alterations compared to the control. These benefits did not follow a clear logarithmic relationship with increasing concentrations of red ginseng extract. Similar observations have been reported in previous studies on varying ginseng dosages in other aquaculture species, where efficacy did not consistently increase with higher concentrations [16, 55]. In one study involving olive flounder (*Paralichthys olivaceus*), growth parameters such as SGR and feeding efficiency declined at elevated ginseng levels after eight weeks of supplementation [55], while lysozyme activity reached its highest value at the lowest concentration tested [55]. These findings highlight the critical need to identify optimal supplementation levels. Therefore, additional research is warranted to refine the dilution ratios and determine the most effective concentration of red ginseng extract that simultaneously optimizes all measured outcomes. As feed additives are easily administered through oral routes and have demonstrated clear efficacy, their successful commercialization

could substantially increase productivity in aquaculture while capitalizing on the valuable traditional Korean resource, *P. ginseng*.

Acknowledgments: None

Conflict of Interest: None

Financial Support: None

Ethics Statement: None

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