Effects of *Padina australis* (Hauck) polysaccharide extract on growth, antioxidant and nonspecific immune parameters of the western white leg shrimp, *Litopenaeus vannamei* (Boone)

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Abstract

This study aimed at determining impacts of various amounts of water-soluble poly-saccharide extracts from brown algae, *Padina australis* (Hauck) (WPP) on actions of anti-oxidant enzymes, the growth, and non-specific immune response of shrimp, *Litopenaeus vannamei* (Boone). Triplicate groups of shrimp (1 ± 0.1g) have been given feed 4 iso-nitrogenous and iso-lipidic diets composing of 4 levels; that is, 0 (control), 0.5, 1.0, and 1.5 g kg^{-1} of WPP extract for eight weeks. Analyses showed that significantly positive effects of diets with 1.0 g kg^{-1} of WPP extract on the growth function of the studied groups. Additionally, improvements of the anti-oxidant enzyme activities, such as catalase, malondialdehyde, super-oxide dismutase, and glutathione peroxidase via feeding 0.5 and 1 g kg^{-1} of WPP extracts. Furthermore, activity of phenoloxidase and lysozyme in the treatment fed by 0.5 and 1 g kg^{-1} of WPP extract have been considerably greater in comparison to the controls and 1.5 g kg^{-1} treatments. Findings demonstrated that usage of 1.0 g kg^{-1} of water soluble poly-saccharides extract of the *P. australis* are useful on growth function, antio-xidant and nonospecific immune variables of the *L. vannamei*.

Keywords: Antioxidant enzyme and Immune response, *Padina australis*, *Litopenaeus vannamei*, Water-soluble polysaccharides

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Introduction

The shrimp farming industry has been great progress during the past decades and presently the Pacific white leg shrimp, *Litopenaeus vannamei*, is one main cultural specie in the world. However, according to the FAO, the global production of this shrimp is over 3 million tons annually; its commercial farming has been suffering from various degrees of infectious diseases and immune suppression (FAO 2016).

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Many of the bacterial, fungal and protozoan-caused diseases are now managed by chemotherapy but unfortunately, the frequent use of commercial antibiotics and veterinary drugs promote the development of antibiotic resistance bacteria (Yeh, Li, Tsui, Lin & Chen 2010). Therefore, at present there is a vital need for exploration and evolution of efficient natural factors with more desirable potentials, lower consequences, acceptable bio-availability, and minimum poisonousness to improve growth performance and health condition of cultivable shrimp species (Thitamadee, Prachumwat, Srisala, Jaroenlak, Salachan, Sritunyalucksana & Itsathitphaisarn 2016). In this regard, attention to the use of plant sources has been increased and the biological effects of number of terrestrial and aquatic plant have been tested as an alternative to chemical treatments (Vatsos & Rebours 2015).

Recently, many studies have focused on the use of seaweeds extract as therapeutic agents to control diseases in aquaculture species (Venkatesan, Lowe, Anil, Manivasagan, Kheraif, Kang & Kim 2015; Wells, Potin, Craigie, Raven, Merchant, Helliwell & Brawley 2017; Wijesekara, Pangestuti & Kim 2011). Many biological properties have been reported from seaweeds metabolites. Such compositions showed anti-viral, anti-fungal, and anti-bacterial characteristics. The walls of the cells of seaweed vary with the walls of terrestrial plants because their polysaccharides composition and structure (degree of branching, substituents, sulphation and type of linkages) are quite different from terrestrial plants (Yangthong, Hutadilok- Towatana, Thawonsuwan & Phromkunthong 2016; Yuguchi, Bui, Takebe, Suzuki, Nakajima, Kitamura & Thah 2016).

Documents of bio-activity of algal polysaccharides significantly obtained by in vitro experiments and fewer data are available in animal trials. In fish, it has been shown that polysaccharides can induce the augmented macrophage phagocyte activities in carp, *Cyprinus carpio* (Linnaeus) (Fujiki, Matsuyama & Yano 1993), or the enhancedd segment of neutrophils, amount of phagocytosis, oxidative burst and interleukins expression in the rainbow trout, *Oncorhynchus mykiss* (Walbaum) (Peddie, Zou & Secombes 2002). In shrimp, polysaccharide component from *Sargassum siliquosum* J.Agardh effectively augment innate immune variables and expressing the immune-associated genes in *L. vannamei* (Yudiati, Isnansetyo & Handayani 2016). Also, *Fenneropenaeus indicus* (H. Milne-Edwards) shrimps submerged in hot water extracts at 300 and 500 mg *Sargassum glaucescens* J.Agardh /L and 500 *mg Padina boergesenii* enhanced the phagocyte activities and increased clearance effectiveness to *Vibrio harvey* (Baumann *et al*.) (Ghaednia, Merhrabi, Mirbakhsh, Yeganeh, Hoseinkhezri, Garibi & Ghaffar Jabbari 2010, 2011).

Padina australis is a brown algae found in Persian Gulf, in south of Iran. It has been documented that specimens of the brown seaweed have a lot of cell wall polysaccharides. Several compounds of this alga have shown potent biological activity (Jaswir, Noviendri, Salleh, Taher & Miyashita 2011; Kantachumpoo & Chirapart 2010; Yuguchi et

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al. 2016). Yuguchi et al.'s (2016) current research demonstrated that polysaccharides component of *P. australis* is able for stimulating immunological activities of intestine via Peyer's patch cells. Although, there are many studies about polysaccharide potential of *P. australis*, its effect on aquatic animals have been less exploited. Therefore, the present research has been done to determine impacts of various amounts of water soluble poly-saccharide extracts of brown algae, *P. australis* on the growth, activity of anti-oxidant enzymes, and non-specific immune reactions of white leg shrimp, *L. vannamei.*

Materials and Methods

Algal extracts and experimental diets

The brown seaweed, *P. australis*, has been collected from Chabahar coastal region of the Oman Sea in southeastern of Iran on the summer of 2016, then they have been completely washed by the distilled water, and air-dried at 60 ˚C. The crude poly-saccharide has been drawn out according Tabarsa and colleagues (2012). The dried raw materials have been crushed in a blender, screened (dimension of the pore was less than 0.5 mm), and kept at –20 ˚C prior to extracting polysaccharide. 85% ethanol (200 mL) was used to treat algae powder (20 g) at room temperature overnight while it has been stirred constantly by mechanical device for removing lipophilic materials, including carotenoid and chlorophyll-associated pigment, and proteins with little molecular weights. Then, it has been centrifuged at 18,500 gram for 10 minutes at 10 ˚C. Afterwards, precipitates have been gathered, washed with acetone, and dried at room temperature. The dried bio-mass (20 g) has been drawn out with distilled water (400mL) at 65 ˚C via mechanical shaking for two hours. Then, it has been centrifuged at 18,500 grams for ten min at 10 ˚C. Supernatant has been achieved and concentrated to 100mL via evaporating under lower levels of pressure at 60 ˚C. Ethanol (99%) (933mL) has been added to the supernatant for obtaining a concentration of 70% for ethanol, and solution has been maintained at 4 ˚C all night. The crude poly-saccharide has been achieved via filteration of the solution and dried at 30 ˚C in an oven all night (Tabarsa, Han, Kim & You 2012). Afterwards, WPP extract has been included in the basal control diet (Table 1), as suitable concentrations, for obtaining 4 various experimental groups at inclusion level of 0, 0.5, 1.0, and 1.5 g kg^{-1} of WPP extract. The diets process has been conducted by mixing the procured diets (Table 1) within a homogeneous mix. 1 mm pellets have been generated through a man-made improved grinder (National, Japan), so that the air dried has been let for keeping humidity nearly at 10% (Table 1) and made cold at 4-8 ˚C to be used later.

Shrimp and farming condition

The shrimps *L. vannamei* in the post-larvae stage have been obtained from a privately-owned hatchery (Chabahar, Iran). The shrimps have been adapted to the lab conditions for two weeks before initiating the feed trials in the Offshore Fisheries Research Center, situated on Chabahar in south of Iran. Post-larvae shrimp

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(mean 1 ± 0.1 gram) have accidentally been assigned into twelve plastic tanks containing 50 shrimp in each tank for the feed trial when the overal weight was determined. 3 replicate groups of shrimps have been fed by hand, so that it clearly satisfy 3 times a day (0800, 1300, and 1700 for six days/week) for eight weeks. Water has been kept at 29 ± 2 °C, dissolved oxygen 4.0 ± 0.5 mg L^{-1}, pH 7.5 ± 0.5, salinity 33.2 ± 0.35 g L^{-1}. Shrimp has been given diets with 0, 0.5, 1.0 and 1.5 g kg^{-1} WPP as 4 groups. After eight weeks of the experiment, thirty shrimp among all of the dietary treatments have been contaminated by bacteria *P. damselae* within ten days for evaluating disease resistance of the contaminated shrimps.

Table 1. Formulating and proximating chemical analyses of the basal control diets (% as fed basis)

Constituents	**(%)**
Fish meal	30.0
Soybean meal	8.0
Wheat meal	7.0
Squid meal	35.0
Shrimp meal	10.0
Yeast	2.0
Fish oil	1.0
Lecithin	4.0
Vitamins and minerals [a]	2.0
Proximate compositions	**(%)**
Protein	46.7
Lipid	16.7
Moisture	9.3
Ash	10.5
Fiber	3.9
Nitrogen free extract	12.7

[a] Vitamins and minerals provided per Kg. Vitamins: vitamin D, 2500 U; vitamin A, 2500 U; vitamin E, 2000 U. Minerals: 501 mg CuSO4; 0.01 MnSO4 500 CoSO4 ; 1500 mg ZnSO4; 500 KI; 35 Na2SeO3.

Computations of the growth function

Growth function has been calculated by the early and last weights of the whole shrimp in all groups and iterations.

The procedure has been conducted in this way (Wahli, Verlhac, Griling, Gabaudan & Aebischer 2003)

WG (Weight gain) = [(final body weight - initial body weight)/ initial body weigh] × 100

SGR (Specific growth rate) = [(ln final body weight - ln initial body weight) /days] × 100

FCR (Feed conversion ratio) = Wet weight gain × 100/feed intake

Activities of the anti-oxidant enzyme

5 shrimps within each treatment of groups have been randomly sampled in terms of antio-xidant enzymes activities in the end of eight weeks of experiment. Shrimps have been homogenized in 10 (w/v) phosphate buffer solution (0.1 mol L^{-1}, pH 7.2) on ice. Centrifugation of homogenate has been done (6000 rpm, 10 minutes) at 4 °C, and supernatant has been employed for determining malondialdehyde (MDA), superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) activities. SOD activity unit has been regarded as the amounts of the specimen, which catalyzed decomposition of 1 mol of O_2- into H_2O_2 and O_2 in each minute. Absorbance has been recorded at 550 nm. GPx has been conducted with commercially available chemical colorimetric assay kits (ZellBio GmbH, Ulm, Germany). When H is oxidized to GSSG, GPx decreases cumene hydro-peroxide. The produced GSSG is reduced to GSH by consuming NADPH through GR. Declining NADPH (that is readily measured at 340 nm) will be proportionate to the activities of GPx.

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Measurement of CAT action has been done by a calorimetrically enzymatic assay kit at 405 nm. CAT performance unit in such an assay has been viewed as the amounts of the specimen catalyzing decomposition of 1 µmole of H_2O_2 to water and O_2 in one minute. CAT containing 0.5 U mL^{-1} sensitiveness may be determined by this technique. It is argued that intraassay and interassay coefficients of variations respectively have been 6.3% and 7.9%. MDA levels has been measured via a commercially available chemical colorimetrical assay kit based on company instruction (MDA assay kit; ZellBio GmbH, Ulm, Germany). It applies MDA-TBA adduct generation via reacting MDA and thiobarbituric acid (TBA) at high temperatures. Measuring MDA has been done in acidic medium and thermal conditions (90 to 100 °C) colorimetrically at 535 nm. MDA of 0.1 µM sensitiveness may be determined by this procedure. It was found that intraassay and interassay coefficients of variations respectively have been 5.8% and 7.6%. Total soluble proteins have been measured via Bradford method (1976) through BSA as a criterion. The activities of enzyme have been stated as certain activities (U mg^{-1} protein). Each enzymatic assay has been performed in triplicate (Bradford, 1976).

Lysozyme assay

The turbidimetric assay for lysozyme has been administered (Parry Jr, Chandan & Shahani 1965) that showed insignificant modifications (Ellis, 1990). Briefly, 0.03% lyophilized cells of *micrococcus lysodeikticus* (Sigma, ATCC No. 4698) in 0.05 mM sodium phosphate buffer (pH 6.2) has been used by a substrate for assaying lysozyme. 25 microliters of total shrimp homogenate have been poured into 175 µl bacterial suspension duplicate wells of a microtitre plate. Incubation of the mix has been done under room temperatures and measurement of an absorbance at 600 nm has been done after 15 sec by the ELISA plate reader (Argus, PerkinElmer, France). Each unit of lysozyme activities has been described as a plasma decrease number of lysozymes in absorbance of 0.001 mL^{-1} min^{-1}.

Phenoloxidase assay

Centrifugation of the total shrimp homogenate has been done at 700 × g at 4 °C for twenty minutes for measuring phenoloxidase action. Afterwards, the supernatant fluids have been eliminated and pellet has been washed, resuspended slowly in cacodylate citrate buffer (0.45 M sodium chloride, 0.01 M sodium cacodylate, pH 7.0, 0.10 M trisodium citrate) and centrifuged once more. Next, resuspension of the pellet has been done with 200 µl cacodylate buffer (0.26 M magnesium chloride, 0.45 M sodium chloride, 0.01 M calcium chloride, pH 7.0, 0.01 M sodium cacodylate), and incubation of a 100 µl aliquot has been done by 50 µl trypsin (1 mg ml^{-1}), serving as an activator, for ten minutes at 25 to 26 °C. Next, 50 µl of DOPA has been included, followed by 800 µl of cacodylate buffer five minutes later. Measurement of optical density at 490 nm has been done by a spectro-photometer.

Statistical analyses

Before comparing, data checking has been done for normalization (Kolmogorov-Smirnov test)

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and homogeneity of variance. Data analysis has been done via one-way ANOVA through SPSS 22 (Armonk, NY, USA). Analysis of statistical variations between mean values with independent variables has been done by one-way ANOVA, which performs mean comparison with Duncan's test at a reliability of 0.05.

Results

Growth performance

Table 2 reports the growth function and feed usage of shrimp *L.vannamei*, which has been given feed by 4 experimental diets for eight weeks. No mortalities have been seen within the experimental interval. Over the eight weeks of experiment, dietary WPP, significantly enhanced shrimp growth and feed use in *L. vannamei* in comparison to the non-WPP complemented groups ($P < 0.05$). Incorporating WPP at 1.0 g augmented WG up to 425 % and SGR up to 2.86%. The shrimp diets contain WPP 0.5 and WPP1.0 with nearly FCR 1.85 and 1.65 respectively, which has been higher than the control WPP 0 (2.06) and WPP 1.5 (2.3) ($P < 0.05$).

Table 2. Growth function of *L. vannamei* given feed the experimental diet with distinct amounts of water-soluble poly-saccharides extracts of brown algae *P. australis* (WPP) for eight weeks

Parameters	Experimental diets WPP (g kg^{-1})			
	0	**0.5**	**1.0**	**1.5**
Initial body weight (g $fish^{-1}$)	1.06 ± 0.12	1.07 ± 0.10	1.09 ± 0.09	1.06 ± 0.07
Weight gain (%)	425.0 ± 16.8 ^{b}	423.0 ± 14.72^{b}	476.2 ± 8.17^{a}	424.17 ± 15.30^{b}
Specific growth rate (%)	1.61 ± 0.31^{b}	1.60 ± 0.40^{b}	1.69 ± 0.55^{a}	1.58 ± 0.28^{b}
Feed conversion ratio (%)	2.06 ± 0.06^{a}	1.85 ± 0.01^{b}	1.65 ± 0.02^{c}	2.03 ± 0.03^{a}

Values (means ± SE, n = 3) with various superscripts in the same row are significantly different ($P < 0.05$).

Anti-oxidant enzyme activities

Table 3 presents the activities of anti-oxidant enzymes of total tissues of shrimps such as glutathione peroxidase (GPx), catalase (CAT) value, malondialdehyde (MDA) contents, and superoxide dismutase (SOD). The actions of GPx, SOD, MDA, and CAT of the shrimps have been influenced by diets complemented by WPP ($P<0.05$). *L.vannamei* receiving WPP1.0 demonstrated greater GPx (334.8 ± 7.5 U mg^{-1} protein) compare to WPP 0.5 (321.3 ± 11.3 U mg^{-1} protein) and WPP1.5 (311.6 ± 10.1 U mg^{-1} proteins). CAT activities of the shrimps fed diets with WPP1.5 (4.01 ± 0.2 U mg^{-1} proteins) have been similar to WPP 0 (3.94 ± 0.11 U mg^{-1} protein) and lower than WPP 0.5 (4.5 ± 0.32 U mg^{-1} protein) and WPP 1.0 (4.53 ± 0.4 U mg^{-1} protein) ($P<0.05$). MDA contents of *L.vannamei* fed diets composing of WPP 0.5 (6.83 ± 0.4 U mg^{-1} protein) and WPP 1.0 (6.68 ± 0.3 U mg^{-1} protein) have been less than WPP 0 to WPP 1. 5 ($P < 0.05$).

Table 3. Antioxidant activities of *L.vannamei* fed the experimental diets containing different levels of water-soluble polysaccharides extract of algae *P. australis* (WPP) for 8 weeks

Antioxidant enzyme (U mg protein)	Experimental diets WPP (g kg^{-1})			
	0	**0.5**	**1.0**	**1.5**
SOD	25.20 ± 0.42 ^{c}	46.86 ± 1.5 ^{b}	49 ± 0.50 ^{a}	25.23 ± 0.25 ^{c}
GPX	305.22 ± 9.4 ^{c}	321.31 ± 11.3 ^{b}	334.80 ± 7.5 ^{a}	311.67 ± 10.1^{bc}
CAT	3.94 ± 0.11 ^{b}	4.50 ± 0.32^{a}	4.53 ± 0.40^{a}	4.01 ± 0.23^{b}
MDA	7.15 ± 0.33^{a}	6.83 ± 0.44^{b}	6.68 ± 0.31^{b}	7.12 ± 0.14^{a}

Values (means ± SE, n =3) with different superscripts in the same row are significantly different ($P < 0.05$).

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Activity of lysozyme

The present paper has demonstrated impacts of diets with WPP on lysozyme activities of the shrimps. Table 4 reports the analyzed information about activities of lysozyme of shrimps in various treatments and controls. Lysozyme activities increased directly with concentration of WPP up to 1.0 g kg $^{-1}$. Shrimp receiving WPP at 0.5 and 1.0 respectively were 19.4 ± 0.5 and 20.6 ±1.1μg mL^{-1} that have been considerably greater than in the shrimps that received WPP 0 and WPP 1.5 with 18.3 ± 0.02 and 18.2 ± 0.15 (μg mL^{-1}) (P<0.05).

Table 4. Immune parameters of *L.vannamei* fed the experimental diets containing different levels of water-soluble polysaccharides extract of algae *P. australis* (WPP) for 8 weeks

Immune parameters (μg mL^{-1})	Experimental diets			
	WPP (g kg^{-1})			
	0	0.5	1.0	1.5
PO	22.7 ± 0.13^{c}	30.3 ± 0.17 ^{b}	35.1± 0.24^{a}	22.7 ± 0.21^{c}
Lysozyme	18.3 ± 0.02^{c}	19.4 ± 0.52 ^{b}	20.6 ±1.10^{a}	18.2 ± 0.15 ^{c}

Values (means ± SE, n =3) with different superscripts in the same row are significantly different (P <0.05). ns values are not significant (P >0.05).

Phenoloxidase (PO) activities

Phenoloxidase activities exhibited considerable differences (P < 0.05) among each experimental group after eight weeks of culture (Table 4). Levels of phenoloxidase activities of the shrimps slowly enhanced from 22.7-35.1 (μg mL^{-1}) when the level of WPP increased from 0 to 1.0 g kg^{-1} of the shrimps fed.

Discussion

Distribution of the specimens of the marine brown algal genus *Padina* has been done completely all over of tropical regions, so that they can be easily recognized in the farm. Numerous bioactive compounds from genus *Padina* are already reported and among their compounds polysaccharide gain much attraction because they possess various practical features, including anti-oxidants, anti-cancers, anti-coagulations and immunomodulation, which can be applied in drug sectors (Fayad, Nehmé, Tannoury, Lesellier, Pichon & Morin 2017; Vatsos & Rebours 2015; Yuguchi et al. 2016). One of the curent studies illustrated that sulfated polysaccharide from *P. australis*, significantly stimulate intestinal immunological activity via Peyer's patch cell (Yuguchi et al. 2016). However, there are several reports on the immunomodulatory potential of *P. australis* under in vitro condition, but limited study has exploited its potential under in vivo condition for aquatic animals (Chiao-Wei, Siew-Ling & Ching-Lee 2011; Jaswir et al. 2011; Santoso, Yoshie-Stark & Suzuki 2004). Therefore, this paper has been developed to find impacts of poly-saccharide extraction of algae *P. australis* on development and immune response on the white leg shrimp *L. vannamei*.

In the current study, water-soluble polysaccharides extract from *P. australis* which is one of the majority algae species along the south coast of Iran was used in shrimp *L. vannamei* diet. Increased weight gains and

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specific growth rates have been observed in *L. vannamei* fed diets containing 1.0 of WPP. Historically, many species of seaweed have been evaluated as feed ingredients or additives for aquatic organisms with positive results on growth performance and disease resistance. For example, in a study by Peñaflorida & Golez (1996) more favorable weight gains in *Penaeus monodon* (Fabricius) was observed when they have been given diets containing 5 % *Kappaphycus alvarezii* (Doty ex Silva) (Peñaflorida & Golez 1996). Totally, in the majority of studies the inclusion of seaweed had significant main effect on immune status rather than the weight gains and specific growth rates of aquatic animal. For example, using aqueous extract from *Sargassum* sp. improved the immune reactions in Asian sea bass, *Lates calcarifer* (Bloch), but exhibited no significant influence on the weight gains and feed conversion ratio of this fish (Yangthong et al. 2016). Researchers proposed that the existence of antinutritional phytochemicals in the crude seaweed extract may have effect on growth performance. The significant enhancement in weight gains of *L. vannamei* in this paper demonstrated that complemented diets containing a more purified seaweed extract with just polysaccharides of *P. australis* are useful as growth promoter. Findings also illustrated that concentration of 1.0 g kg^{-1} WPP diets remarkedly increased WG and enhanced FCR as compared to higher concentrations of 1.5 g kg^{-1}. This indicates that 1.0 g kg^{-1} WPP supplementation is the adequate concentration that promotes growth in *L. vannamei*. The hypothesis is that polysaccharides of seaweeds trigger the growth of the useful bacteria, and improve the health of intestine and stimulate growth function (Venkatesan et al. 2015).

To increase disease resistance in cultured shrimp, the use of water-soluble polysaccharides extract from *P. australis* was practiced and the results revealed that different concentration of WPP has significant influence on lysozyme activities of *L. vannamei*. Lysozyme activity in *L. vannamei* increased by increasing WPP level up to WPP1.0. Lysozyme is an important antibacterial protein produced by shrimp in response to a pathogen challenge. Lysozyme destroys bacteria via hydrolysis of b-1, 4-glycosidic links between N-acetyl-glucosamine and N-acetylmuramic acid of the peptidoglycan layers in the bacterial cell walls. Furthermore, recent work by Chen et al. 2015 indicated that *Gracilaria tenuistipitata* var extract improved lysozyme activity in *L. vannamei* (Chen, Chen, Lin, Yeh & Huang 2015). The fucoidan has also been reported to improve the growth and resistance against *Vibriosis* in *Penaeus monodon* and *Penaeus japonicas* (Spence Bate) larvae (Traifalgar, Koshio, Ishikawa, Serrano & Corre 2012; Traifalgar, Serrano, Corre, Kira, Tung, Michael & Ishikawa 2009). PO activity also had a difference in each treatment. This suggest the most favorable outcomes in the shrimp fed with WPP 1.0 diet. Similarly, enhancement of PO activities have been found in *L. vannamei* by using diet with hot water extracts of *Gelidium amansii* (Fu, Hou, Yeh, Li & Chen 2007). Reports indicated that administrating sodium alginate drawn out from brown algae *Macrocystis pyrifera* (L.) and *L. nigrescens*

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(Bory de Saint-Vincent) increased *L. vannamie* resistance versus *Vibrio alginolyticus* (Miyamoto *et al*.) (Cheng, Liu, Yeh & Chen 2004; Cheng, Liu, Kuo & Chen 2005).

Removing free radicals is of high importance for anti-oxidant defense in the cell or food systems, because they invade a majority of biological molecules via adjusting free radical chain responses, which damage considerably organisms. In this research, administration of different concentration of WPP has resulted in different antioxidant enzyme activities in shrimps. The level of SOD activity in experimental shrimp fed diets of 1.0 g kg^{-1} of WPP has been considerably greater than other groups. Furthermore, results obtained in shrimp GPx and CAT activity were affected by dietary treatment. The higher GPx and CAT activity were observed in shrimp received 0.5 and 1.0 g kg^{-1} of WPP. The MDA content were also positively affected by WPP treatment. The MDA level in shrimps given feed 0.5 and 1.0 g kg^{-1} doses of WPP have been considerably lower than controls and WPP 1.5 dose. Many researchers have reported various types of antioxidants in different kinds of seaweeds (Bourguiba, Zahlila, Bouaïcha, Amri & Mezghani 2017; Chandini, Ganesan & Bhaskar 2008; Godard & colleagues, 2009). Their results suggest that the polysaccharides may be an indication of potential antioxidant and several evidence showed that biological activity of poly-saccharides is dependent on their structural properties, including degree of sulfation and dispersion patterns of sulfate, kind of glycosidic bonds, branch structure, molecular weights, and mono-saccharide composition sulfate groups exhibit high nucleophilic features they might chelate with metal ions that defend specimens versus oxidative injuries (Sun, Wang, Li & Liu 2014). Therefore, poly-saccharides from seaweed are of special importance in bio-chemical and medical fields.

This study results show that the brown alga, *P. australis* which is a prominent and popular alga occupies the south coast of Iran have considerable potential as an anti-bacterial agent and immune stimulant in *L. vannamei* culture.

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Conflict of interests

The authors declare that there is no conflict of interest.

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اثر عصاره پلی ساکارید جلبک پادینا (*Padina australis*) بر رشد، شاخص‌های ایمنی غیر اختصاصی و آنتی اکسیدانی میگوی پا سفید غربی (*Litopenaus vannamei*)

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چکیده

این مطالعه با هدف بررسی اثر سطوح مختلف عصاره پلی ساکارید محلول در آب جلبک قهوه پادینا (*Padina australis*) بر رشد، فعالیت آنزیم‌های آنتی اکسیدانی و پاسخ های ایمنی غیر اختصاصی میگوی پاسفید غربی (*Litopenaus vannamei*) انجام شد. تیمارهای میگو (با سه تکرار) با وزن ۰/۱±۱ گرم با جیره حاوی چربی و نیتروژن یکسان و حاوی ۴ سطح ۰، ۰/۵، ۱و ۱/۵ گرم عصاره پلی ساکارید محلول در آب جلبک پادینا بر کیلوگرم غذا به مدت ۸ هفته تغذیه شدند. نتایج نشان داد که رژیم غذایی محتوی ۱ گرم عصاره پلی ساکارید محلول در آب جلبک پادینا بر کیلوگرم غذا اثر مثبتی بر عملکرد رشد تیمار مطالعه شده داشت. به علاوه، فعالیت آنزیم‌های آنتی اکسیدان شامل سوپر اکسید دیسموتاز، گلوتاتیون پراکسیداز، کاتالاز و میزان مالون دی آلدئید در تغذیه با ۰/۵ و ۱ گرم عصاره محلول در آب پلی ساکارید جلبک پادینا بر کیلوگرم غذا بهبود یافت. هم‌چنین فعالیت فنل اکسیداز و لیزوزیم در تیمار حاوی ۰/۵ و ۱ گرم محلول در آب پلی ساکارید جلبک پادینا بر کیلوگرم غذا به صورت معنی‌دار بیشتر از تیمار کنترل و ۱/۵ گرم محلول در آب پلی ساکارید جلبک پادینا بر کیلوگرم غذا بود. این نتایج توصیه کرد که استفاده از ۱ گرم عصاره محلول در آب پلی ساکارید بر کیلوگرم غذا می‌تواند منجر به بهبود عملکرد رشد، شاخص‌های ایمنی غیر اختصاصی و آنتی اکسیدانی میگوی پا سفید غربی گردد.

کلمات کلیدی: پلی ساکارید محلول در آب، پادینا، میگوی پاسفید غربی، آنزیم آنتی اکسیدان، پاسخ ایمنی

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