RESEARCH ARTICLE Indian Journal of Animal Research, Volume 55 Issue 7: 827-830 (July 2021)

Dietary *Laminaria japonica* Polysaccharide Regulates Systemic Defence Property of Weaned Piglets Improving Growth Performance under High Temperature Condition

C.W. Wang[1], Y. Jiang[1], W.N. Chen[1], Q. Liu[1], X.F. Cheng[1], Y.H. Zhang[1], D.D. Yuan[1], X.R. Jiang[2] **10.18805/IJAR.B-1340**

ABSTRACT

Background: This study was conducted to evaluate the effect of dietary LJP supplementation on growth performance, heat stress resistance and immune status of weaned piglets raised in a high-temperature environment (34 ± 3°C).

Methods: A total of 72 Barkshire × Licha Black crossbred piglets were weaned at 30 days of age (BW, 5.88 ± 0.16 kg) and fed either without or with 100, 200 or 400 mg/kg LJP in a completely randomized design, with 3 pens per diet (6 pigs per pen). On day 21, a total of 12 piglets were selected to collect blood samples.

Result: The results showed that 200 and 400 mg/kg LJP supplementation decreased the feed conversion ratio (FCR) of piglets compared to the control group (*P*=0.037) and dietary LJP linearly increased average daily gain and decreased FCR of piglets (*P*=0.037 and 0.011, respectively). In addition, adding 100, 200 and 400 mg/kg LJP reduced the serum HSP70 content compared to the control group (*P*=0.019) and dietary LJP linearly and quadratically decreased the serum HSP70 content of piglets (*P*=0.030 and 0.011, respectively). Moreover, supplementation of 200 mg/kg and 400 mg/kg LJP increased the serum (NO) and nitric oxide acid phosphatase (ACP) content compared to the control group (*P*=0.019; *P*=0.003) and dietary LJP linearly increased the serum lysozyme, NO and ACP contents (*P*=0.039; *P*=0.005; *P*=0.001). In conclusion, dietary LJP has potential to regulate systemic defence property of weaned piglets raised in a high temperature condition, accompanied by increased growth performance.

Key words: Feed conversion ratio, Heat stress resistance, Immune status, Serum HSP70.

INTRODUCTION

High temperature environment has a great negative impact on the growth performance of piglets, which reduces the intake and daily gain of piglets, reduces immunity, causes diarrhoea and even death (Johnson and Lay, 2017). The highest temperature in the summer period can exceed 40°C in the south of China, in where piglets would undergo a long-term acute or chronic heat stress and negatively influence the growth and health status of piglets.

Laminaria japonica is a large-scale economic brown alga with both edible and medicinal functions and is one of the main algae breeds in China. *Laminaria japonica* enrich in many vitamins, trace elements and growth promoting factors. *Laminaria japonica* polysaccharides (LJP) are a kind of natural bio-macromolecule substances existing intercellular and intracellular cells of *Laminaria japonica* (Zvyagintseva *et al*., 2005). Previous studies demonstrated that LJP exhibited a wide range of pharmacological properties, such as anti-oxidant, anti-inflammation, anti-virus, antithrombotic and anti-cancer (Fang *et al*., 2015; Lu *et al*., 2013; Vishchuk *et al*., 2011; Yue *et al*., 2017). In addition, previous studies reported that the inclusion of the laminaria or seaweed extract derived from *Laminaria* spp. could play a positive role in weaned piglets through increasing growth performance (Mcdonnell *et al*., 2010; O'Doherty, *et al*., 2010) and scavenging hydroxyl radical and superoxide anion radical (Zhang *et al*., 2003).

[1]College of Life Science, Jiangxi Science and Technology Normal University, Nanchang 330013, China.
[2]Key Laboratory of Feed Biotechnology of Ministry of Agriculture and Rural Affairs, Institute of Feed Research, Chinese Academy of Agricultural Sciences, Beijing 100081, China.

Corresponding Author: C.W. Wang; X.R. Jiang; College of Life Science, Jiangxi Science and Technology Normal University, Nanchang 330013, China; Key Laboratory of Feed Biotechnology of Ministry of Agriculture and Rural Affairs, Institute of Feed Research, Chinese Academy of Agricultural Sciences, Beijing 100081, China.
Email: 616051233@qq.com; jiangxianren@caas.cn

How to cite this article: Wang, C.W., Jiang, Y., Chen, W.N., Liu, Q., Cheng, X.F., Zhang, Y.H., Yuan, X.R., Jiang, D.D. (2021). Dietary *Laminaria japonica* Polysaccharide Regulates Systemic Defence Property of Weaned Piglets Improving Growth Performance under High Temperature Condition. Indian Journal of Animal Research. 55(7): 827-830. DOI: 10.18805/IJAR.B-1340.

Submitted: 12-12-2020 **Accepted:** 31-03-2021 **Online:** 26-06-2021

In recent years, the application of LJP in breeding industry has been explored, while the effect of LJP on physiological function of weaned piglets under high temperature environment is rarely reported. Thus, the present study was designed to investigate the effect of dietary supplementation of LJP on growth performance, heat stress resistance and immune status of weaned piglets

raised in a high temperature condition. In addition, the optimal supplementation level of LJP in the diet of piglets was evaluated, which may serve as a base for further studies.

MATERIALS AND METHODS

All the experimental procedures have been approved by the Animal Care and use Committee of the Jiangxi Science and Technology Normal University.

Experimental design and animal management

The study was carried out during summer period at a local commercial swine farm in Nanchang, China. A total of 72 healthy weaned crossbred barrows (Barkshire × Licha Black), with similar initial body weight (BW, 5.88 ± 0.16 kg) and age (30 ± 1 days), were randomly allocated to 4 treatments, with 3 replicated pens per treatment and 6 piglets per pen. All piglets were raised in the barn with high temperature (34 ± 4°C) and humidity (70 ± 5%) during the summer period (From July to August) in the south of China for a 21-days period. The dietary groups were: control group fed a basal diet without additive and LJP groups fed with 100, 200 or 400 mg/kg of LJP. The LJP product contained 80% fucoidan, which was purchased from Xi'an Zhen Lu Biotechnology Limited Company (Xi'an, Shanxi, China). Each 1.50 × 1.50 m^2 pen was equipped with a feeder and a water nipple to allow *ad libitum* consumption of feed and water. The basal diet was a typical corn-soybean diet formulated to meet the requirements of weaned piglets (NRC, 2012). Compositions of basal diet and nutrition level are presented in Table 1. The diet for the piglets was manufactured 1 week before the start of the trial without the inclusion of any antibiotic growth promoters.

Experimental observations and measurements

At the beginning (day 0) and end of the experiment (day 21), the body weight (BW) piglet was individually measured. The feed consumption was recorded daily. On day 21, average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) of piglets were calculated.

At the end of the experiment, 1 piglet per pen was randomly selected. Blood samples were taken from the jugular vein and were immediately centrifuged at 3000 r/min for 10 min at 4°C to separate the serum. Then, the serum samples were stored at -20°C until analysis. The contents of malondialdehyde (MDA) and heat shock protein 70 (HSP70) and the activities of glutathione peroxidase (GSH-PX) and superoxide dismutase (SOD) in the serum were determined spectrophotometrically using the commercial assay kits (Nanjing Jiancheng Bioengineering Institute, Nanjing, China). The activities of acid phosphatase (ACP), alkaline phosphatase (AKP) and the contents of lysozyme (LZM) and nitric oxide (NO) in the serum were determined spectrophotometrically with the commercial assay kits according to the manufacturer instructions (Nanjing Jiancheng Bioengineering Institute, Nanjing, China).

Statistical analysis

All the experimental data were analysed by one-way ANOVA using the GLM procedure of SPSS software (SPSS 24.0 version, SPSS Inc., Chicago, IL, USA). The model included the treatment effect and the pen represented the experimental unit for growth performance, while the individual piglet was the experimental unit for serum indices. The treatment comparisons were performed using Tukey's honestly significant difference test for multiple testing. Linear and quadratic contrasts of the LJP effects were also computed. Data with $P<0.05$ were considered to be significant, while those in the $P=0.05$-0.10 range were considered tendencies.

RESULTS AND DISCUSSION

Growth performance

The effect of LJP on the growth performance of weaned piglets is presented in Table 2. Ferreira (2005) suggested that the upper critical temperature for piglets between 28 and 35 days of age is 30°C. In the current study, the average temperature in the barn was 34°C that could induce decreased feed intake and weight gain of piglets as demonstrated by Dos Santos Cerqueira *et al*. (2019). Our results showed that dietary LJP did not significantly affect

Table 1: Composition and nutrient levels of the basal diet (as fed basis).

Items	Content, %
Ingredients	
Expanded corn	57.50
Expanded soybean	12.00
Soybean meal	8.00
Whey powder	8.00
Lactose	8.00
Fish meal	3.00
Limestone	0.70
Calcium hydrogen phosphate	1.50
Salt	0.30
Premix[1]	1.00
Analysed composition	
CP, %	20.50
Ca, %	0.94
Total P, %	0.75
Calculated composition	
ME, MJ/kg	14.30
Lys, %	1.45
Met, %	0.45

[1]The premix provides the following per kilogram of diet: V_A l5 000 U, V_{D3} 2000 U, V_E 30.00 IU, V_K 3.75 mg, V_{B1} 2.50 mg, V_{B2} 7.00 mg, V_{B6} 2.4 mg, V_{B12} 0.03 mg, nicotinic acid 40.00 mg, calcium pantothenate 25.00 mg, Folic acid 25.00mg, biotin 0.25 mg, choline chloride 600 mg, Cu 20 mg, Fe l20mg, Zn l60 mg, Mn 100 mg, I 0.54 mg, Se 0.27 mg.

the BW and ADFI of weaned piglets (P>0.05). However, supplementation with 200 mg/kg LJP tended to increase the ADG and decreased the FCR of piglets (Contrast, P=0.100 and 0.058, respectively) and dietary LJP at 400 mg/kg significantly decreased the FCR (Contrast, P=0.046) compared to the control group. In addition, supplementation of LJP linearly increased ADG (P=0.037) and linearly decreased the FCR (P=0.011) of piglets. Similarly, previous studies reported that the inclusion of the laminaria or seaweed extract derived from *Laminaria* spp. could increase growth performance of weaned piglets (McDonnell *et al*., 2010; O'Doherty *et al*., 2010). In the present study, the potential improvement in growth performance of the LJP piglets may be due to a modulated health status under the high temperature condition, as evidenced by the alleviated response to heat stress and increased systemic non-specific immune indices.

Antioxidant and heat stress indices

Effect of dietary LJP on the heat stress indices in the serum of piglets is shown in Table 3. In this study, adding 100, 200 and 400 mg/kg LJP reduced the HSP 70 content in the serum compared to the control group (Contrast, P=0.055, 0.018 and 0.062, respectively) and dietary LJP linearly and quadratically decreased the content of HSP70 in the serum of piglets (P=0.030 and 0.011, respectively). The expression of HSP70 in the heat stress environment can maintain the self-stability of cell proteins, improve the tolerance of cells to stressors, maintain normal physiological functions and thermal balance of cells and alleviate the stress damage suffered by the body (Lee and Corry, 1998; Nakhjavani *et al*., 2010). In the current study, our results demonstrated that LJP could decrease the content of HSP70 in the serum of piglets. Previous studies evidenced that the fucoidan polysaccharide extracted from *Laminaria japonica* had the scavenging effect on hydroxyl radical and superoxide anion radical and antioxidant efficacy (Zhang *et al*. 2003). However, the LJP supplementation did not significantly affect the MDA content and the activities of GSH-PX and SOD in the serum of piglets (P>0.10), suggesting that the effect of dietary LJP on superoxide and hydrogen peroxide might be less acute than on heat stress response protein under the high temperature condition.

Table 2: Effect of *Laminaria japonica* polysaccharide (LJP) on growth performance of weaned piglets[1]

	LJP, mg/kg				SEM	*P*-value		
	0	100	200	400		ANOVA	Linear	Quadratic
BW, kg								
Day 0	5.86	5.90	5.92	5.84	0.099	0.955	0.862	0.613
Day 21	8.89	9.13	9.28	9.19	0.124	0.257	0.164	0.150
ADG, g/d	$144^{b,y}$	154^{ab}	160^{a}	$159^{ab,x}$	4.1	0.088	0.037	0.116
ADFI, g/d	201	210	214	212	5.2	0.419	0.227	0.263
FCR	1.40^{a}	1.37^{ab}	1.34^{b}	1.33^{b}	0.014	0.037	0.011	0.125

^{a,b}Means listed in the same row with different superscripts are significantly different (P≤0.10).

^{x,y}Means listed in the same row with different superscripts are tended to be different (0.10<P≤0.20).

Table 3: Effect of *Laminaria japonica* polysaccharide (LJP) on antioxidant and heat stress indices of weaned piglets.

	LJP, mg/kg				SEM	*P*-value		
	0	100	200	400		ANOVA	Linear	Quadratic
MDA, nmol/mL	3.34	3.40	3.47	3.24	0.326	0.981	0.851	0.734
GSH-PX, U/mL	352	359	377	371	13.4	0.721	0.403	0.546
SOD, U/mL	41.47	45.42	43.11	44.97	1.426	0.281	0.255	0.548
HSP70, ng/mL	4.55^{a}	3.28^{b}	2.95^{b}	3.31^{b}	0.278	0.019	0.030	0.011

^{a,b} Means listed in the same row with different superscripts are significantly different (P≤0.10).

Table 4: Effect of *Laminaria japonica* polysaccharide (LJP) on serum nonspecific immune indices of weaned piglets.

	LJP, mg/kg				SEM	*P*-value		
	0	100	200	400		ANOVA	Linear	Quadratic
LZM, μg/mL	1.77^{y}	2.60	2.77	3.42^{x}	0.381	0.166	0.039	0.604
NO, μmol/L	$139^{b,y}$	$134^{b,y}$	$182^{ab,x}$	197^{a}	12.1	0.019	0.005	0.801
ACP, king unit/100 mL	9.60^{b}	15.20^{b}	27.57^{a}	27.62^{a}	2.252	0.003	0.001	0.052
AKP, king unit/100 mL	33.94	38.78	39.73	43.52	3.825	0.561	0.193	0.763

LZM = Lysozyme; NO = Nitric oxide; ACP = Acid phosphatase; AKP = Alkaline phosphatase.

^{a,b}Means listed in the same row with different superscripts are significantly different (P≤0.10).

^{x,y}Means listed in the same row with different superscripts are tended to be different (0.10<P≤0.20).

Non-specific immune indices

Effect of dietary LJP on the non-specific immune indices in the serum of piglets is shown in Table 4. NO is a feature of genuine immune-system cells as well as other cells involved in immune reactions (Bogdan, 2001). Lysozyme is an important humoral component of the innate immune system, which can attack the peptidoglycan layer in the cell wall of Gram-positive and partial Gram-negative bacteria. Skouras *et al*. (2003) reported that the plasma lysozyme activity was impaired immediately under an environmental challenge. In the present study, dietary LJP at levels of 400 mg/kg increased NO production (Contrast, *P*=0.150 and 0.049, respectively) and ACP activity (Contrast, *P*=0.006; *P*=0.006) and supplementation with 400 mg/kg LJP tended to increase the LZM content (Contrast, *P*=0.126) compared to the control group. In addition, dietary LJP linearly increased the contents of LZM (*P*=0.039) and NO (*P*=0.005) and ACP activity (*P*=0.001) and quadratically increased ACP activity (*P*=0.052) in the serum of piglets. The obtained improvements are probably because LJP can bind to gastrointestinal epithelial cells and gut associated lymphocytes and downregulate the expression of intestinal inflammatory cytokines (Walsh *et al*., 2012), thereby enhancing the body's ability to resist infectious diseases.

CONCLUSION

The obtained results in this preliminary study suggested that dietary LJP had potential to improve growth performance of weaned piglets under a high temperature condition, which might be attributed to the improved systemic immune status and attenuated response to the heat stress. Considering the growth of piglets and cost of product, 200 mg/kg was an appropriate and feasible level for LJP supplementation to weaned piglets' diet. More studies on LJP application may benefit the swine industry.

ACKNOWLEDGEMENT

The authors gratefully appreciate the financial support provided by the Doctoral Research Initiation Foundation Program of Jiangxi Science and Technology Normal University (to C.W. Wang) and the Science and Technology Youth Program of Jiangxi Provincial Department of Education (GJJ150801).

REFERENCES

Bogdan, C. (2001). Nitric oxide and the immune response. Nature Immunology. 2(10): 907-16.

Dos Santos Cerqueira, L.G., Schinckel, A.P., Silveira, H., Kuribayashi, T.H., Moreira, R.H.R., *et al.* (2019). Spray-dried porcine plasma improves feed intake of weaned piglets subjected to heat stress. Journal of Animal Physiology and Animal Nutrition (Berl). 103(3): 836-45.

Fang, Q., Wang, J.F., Zha, X.Q., Cui, S.H., Cao, L. and Luo, J.P. (2015). Immunomodulatory activity on macrophage of a purified polysaccharide extracted from *Laminaria japonica*. Carbohydrate Polymers. 134: 66-73.

Ferreira, R.A. (2005). Maior produção com melhor ambiente para aves, suínos e bovinos. Aprenda Fácil, pp 371.

Johnson, J.S. and Lay, D.C. (2017). Evaluating the behavior, growth performance, immune parameters and intestinal morphology of weaned piglets after simulated transport and heat stress when antibiotics are eliminated from the diet or replaced with L-glutamine. Journal of Animal Science. 95(1): 91-102.

Lee, Y.J. and Corry, P.M. (1998). Metabolic oxidative stress-induced HSP70 gene expression is mediated through SAPK pathway-role of Bcl-2 and c-Jun NH2-terminal kinase. Journal of Biological Chemistry. 273(45): 29857-63

Lu, J., You, L., Lin, Z., Zhao, M. and Cui, C. (2013). The antioxidant capacity of polysaccharide from *Laminaria japonica* by citric acid extraction. International Journal of Food Science and Technology. 48(7): 1352-8.

Mcdonnell, P., Figat, S. and O'Doherty, J.V. (2010). The effect of dietary laminarin and fucoidan in the diet of the weanling piglet on performance, selected faecal microbial populations and volatile fatty acid concentrations. Animal. 4(4): 579-85.

Nakhjavani, M., Morteza, A., Khajeali, L., Esteghamati, A., Khalilzadeh, O., Asgarani, F. and Outeiro, T.F. (2010). Increased serum HSP70 levels are associated with the duration of diabetes. Cell Stress Chaperones. 15(6): 959-64.

NRC - National Research Council. (2012). Nutrient requirements of swine. 11th ed. Washington, DC, USA: National Academies Press;

O'Doherty, J.V., Dillon, S., Figat, S., Callan, J.J. and Sweeney, T. (2010). The effects of lactose inclusion and seaweed extract derived from *Laminaria* spp. on performance, digestibility of diet components and microbial populations in newly weaned pigs. Animal Feed Science and Technology. 157(3-4): 173-80.

Skouras, A., Broeg, K., Dizer, H., von Westernhagen, H., Hansen, P.D. and Steinhagen, D. (2003). The use of innate immune responses as biomarkers in a programme of integrated biological effects monitoring on flounder (*Platichthys flesus*) from the southern North Sea. Helgoland Marine Research. 57(3): 190-8.

Vishchuk, O.S., Ermakova, S.P. and Zvyagintseva, T.N. (2011). Sulfated polysaccharides from brown seaweeds *Saccharina japonica* and *Undaria pinnatifida*: Isolation, structural characteristics and antitumor activity. Carbohydrate Research. 346(17): 2769-76.

Walsh, A.M., Sweeney, T., O'Shea, C.J., Doyle, D.N. and O'Doherty, J.V. (2012). Effects of supplementing dietary laminarin and fucoidan on intestinal morphology and the immune gene expression in the weaned pig. Journal of Animal Science. 90 (Suppl 4): 284-6.

Yue, Y., Li, Z., Li, P., Song, N., Li, B., Lin, W. and Liu S. (2017). Antiviral activity of a polysaccharide from *Laminaria japonica* against enterovirus 71. Biomed Pharmacother. 96: 256-62.

Zhang, Q.B., Li, Z., Xu, Z.H., Niu, X.Z. and Zhang, H. (2003). Effects of fucoidan on chronic renal failure in rats. Planta Medica. 69(6): 537-41.

Zvyagintseva, T.N., Shevchenko, N.M., Nazarenko, E.L., Gorbach, V.I., Urvantseva, A.M., Kiseleva, M.I. and Isakov, V.V. (2005). Water-soluble polysaccharides of some brown algae of the Russian Far-East. Structure and biological action of low-molecular mass polyuronans. Journal of Experimental Marine Biology and Ecology. 320(2): 123-31.