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Effect of *Lactobacillus delbrueckii* on Jejunum Innate Immune-Related Gene Expression in Mice

[1, 2]Jie-Lin Duan, [3]Shao-Ping He, [3]Guan Yang, [1, 2]Jie Yin, [1]Wen-Kai Ren, [3]Jian-Wen Deng, [3]Yu Gong, [3]Feng-Ming Chen, [1]Tie-Jun Li and [3]Xing-Guo Huang
[1]Key Laboratory of Agro-Ecological Processes in Subtropical Region, Chinese Academy of Sciences, Hunan Provincial Engineering Research Center of Healthy Livestock, Institute of Subtropical Agriculture, Scientific Observing and Experimental Station of Animal Nutrition and Feed Science in South-Central, Ministry of Agriculture, Changsha, 410125 Hunan, China
[2]University of Chinese Academy of Science, 100049 Beijing, China
[3]College of Animal Science and Technology, Hunan Agricultural University, Changsha, 410128 Hunan, China

Abstract: Probiotics, regarded as live microorganisms, play important roles in regulating intestinal inflammatory responses and innate immunity. Although, intestinal immunmodulation of probiotics have been enormously conducted into data, the effects of *Lact. delbrueckii* on jejunum innate immunity are rarely reported. This study aimed to investigate the effects of *Lact. delbrueckii* on serum profiles, jejunum inflammatory cytokines and innate immune-related genes expression in mouse. A total of 32 Kunming male mice were randomly divided into four groups and each group were orally administrated with saline normal, living *Lact. delbrueckii*, heat-killed *Lact. delbrueckii* and Spent Culture Supernatant (SCS). Serum profiles (IL-2, IgG) and jejunum immune-related gene expression were detected including inflammatory cytokines (INF-γ, TNF-α, IL-17, IL-13) peneth cell-sepcific gene (cryptdin 1, 4, 5, lysozyme C, CRS-1C, CRS-4C and sPLA2) and goblet cell-specific gene (mucin-2 and mucin-4). Compared with control group, orally administrated SCS significantly decreased ($p<0.05$) ADG of mice. Orally administrated living and heat-killed *Lact. delbrueckii* group significantly increased ($p<0.05$) serum IL-2 level and while SCS remarkably decreased ($p<0.05$) serum IgG level. Meanwhile, oral administration of living *Lact. delbrueckii* significantly enhanced ($p<0.05$) jejunum inflammatory cytokines (INF-γ, TNF-α, IL-17) expression. SCS significantly increased ($p<0.05$) peneth cell-sepcific gene (cryptdin 1, 4, 5 and sPLA2) gene expression. In conclusion, oral administration of *Lact. delbrueckii* affects serum profiles and jejunum innate immune-related gene expression and thereby modulates jejunum innate immunity.

Key words: *Lactobacillus delbrueckii*, jejunum, peneth cell, innate immune, CRS-1C

INTRODUCTION

Lactobacillus delbrueckii (*Lact. delbrueckii*) is a gram-positive bacteria belonging to lactic acid bacteria and conventionally has been used to ferment milk because of its great production of lactic acid that give yogurt its flavor and textural properties (Moller *et al*., 2007). Recently, many compelling studies have reported *Lact. delbrueckii* has potential probiotic function (Fernandez *et al*., 2005). Abedi *et al*. (2013) have reported that *Lact. delbrueckii* exhibits remarked anti-bacterial and anti-adhesion properties against *Escherichia coli* (*E. coli*) *in vitro* (Abedi *et al*., 2013) suggesting that *Lact. delbrueckii* may be able to prevent *E. coli* or other pathogens infection in the gut. Indeed, Dos Santos *et al*. (2011) have demonstrated this point that *Lact. delbrueckii* UFV-H2b20 protects mice from death caused by *Listeria monocyto* genes (*L. monocytogenes*) infection (Dos Santos *et al*., 2011). Besides anti-bacterial and anti-adhesion properties, numerous well-designed experiments have exemplified that *Lact. delbrueckii* exhibits versatile beneficial roles inmodulating intestinal microbiota (Mori *et al*., 2011) enhancing intestinal barrier function (Yu *et al*., 2012) and regulating intestinal immune response through modulation of Nuclear Factor kappa B (NF-κB) and Mitogen-Activated Protein Kinases (MAPKs) signaling pathway (Hegazy and El-Bedewy, 2010; Thomas and Versalovic, 2010). Additionally, *Lact. delbrueckii* enable to facilitate activation of intestinal immune system, increase number of intestinal T cell and exhibits anti-inflammation properties (Picchietti *et al*., 2009; Del Carmen *et al*., 2014).

Corresponding Author: Xing-Guo Huang, College of Animal Science and Technology, Hunan Agricultural University, Changsha, 410128 Hunan, China

Interestingly, orally administrated *Lact. delbrueckii* but also exhibits anti-aging ability by slowing the aging of the T cell and elevates antimicrobial peptide human Beta-Defensin-2 (hBD2) expression (Moro-Garcia *et al*., 2013). Moreover, *Lact. delbrueckii* has been used to treat children with celiac disease (Di Cagno *et al*., 2009) type 2 diabetes (Honda *et al*., 2012) and Inflammation Bowel Disease (IBD) (Hegazy and El-Bedewy, 2010; Sengul *et al*., 2011) and experimental autoimmune encephalomyelitis (Lavasani *et al*., 2010). Considering versatile beneficial roles of *Lact. delbrueckii* it is plausible to speculate a possible role of *Lact. delbrueckii* in modulating jejunum innate immunity. However, directly evidence in favor of this hypothesis is missing.

The mucosa of the gastrointestinal tract is in directly contact with intestinal lumen contents including bacterial and antigens. Being an important harbor for microbes, the orchestrated intestinal innate immunity that protects host against pathogenic bacterial (Oswald, 2006; Ganz and Szabo, 2013; Vivier and Malissen, 2005) has been highlighted. Under normal or abnormal conditions, Pattern Recognition Receptors (PRRs) like Toll-Like Receptors (TLRs) recognize Pathogen Associated Molecular Pattern (PAMP) and then trigger TLR-MyD88 signaling pathway, leading secretion of inflammatory cytokines which perform crucial roles in regulating intestinal homeostasis (Duan *et al*., 2014; Menendez *et al*., 2013). Paneth cells that populate the crypts through the small intestine exert pivotal roles in intestinal innate immunity because of specific secreted antimicrobial peptides including α-defensins, lysozyme C and Cryptdin-Related Sequence (CRS) peptides (Selsted and Ouellette, 2005; Ouellette, 2004) which exhibit broad spectrum microbicides against gram negative and gram positive microbes by membrane-disruptive and inducing rapid microbial cell K^+ efflux (Shanahan *et al*., 2010; Santaolalla *et al*., 2011). In addition, mucins from goblet cell are indispensable components of intestinal mucus layer that separate host from lumen pathogens (Boonzaier *et al*., 2013; Johansson *et al*., 2008). Many investigations have shown that mucins from goblet cellsplay important roles in intestinal innate immunity by enhancing gut barrier function (Zarepour *et al*., 2013) thus protecting host against pathogens invasion. Although, literatures covering probiotics on intestinal innate immunity have been largely documented into data, the effect of *Lact. delbrueckii* on peneth cell and goblet cell-specific molecules and inflammatory cytokines expression in jejunum remains rarely reported. For these reasons, researchers conducted the experiment to study the effect of *L. delbrueckii* on jejunum innate immunity including serum IgG and IL-2 concentration, expression of inflammation gene, peneth cell-specific gene and goblet cell-specific gene in jejunum in mouse.

MATERIALS AND METHODS

Preparation and culture of *Lact. delbrueckii*: *Lact. delbrueckii* was obtained from laboratory of College of Animal Science and Technology, Hunan Agriculture University. For stimulation experiment, the bacteria were anaerobically cultured for 48 h at 37°C in de Man, Rogosa and Sharp broth (MRS broth) in an anaerobic condition prior to use. *Lact. delbrueckii* lysate were harvested by centrifugation (4,000x, 10 min, 4°C) for spent culture supernatant SCS and stored at 4°C for further use. The bacterial cells in the stationary growth phase were harvested bysterile saline suspension, centrifuged (4000×g, 10 min, 4°C) washed 3 times with sterile saline and subsequently adjusted to a final concentration of 1×10^9 CFU in sterile saline. Heat-killed *Lact. delbrueckii* was obtained by maintaining cells at 56°C for 60 min for inactivation and then reaffirmed whether live *L. delbrueckii* existed by MRS plate.

Experimental design: A total of 32 Kunming male mice were obtained from the animal Laboratory Animal Center of Central South University (Hunan, China). The mices (20±2 g) were randomly assigned to four treatment groups with 8 replicates per treatment. The mice were housed in a friendly and environmentally controlled pathogen-free colony and had access to standard rodent food and water *ad libitum*. After 3 days of adaptive feeding, the four groups of mice were orally administrated with saline normal, live *Lact. delbrueckii* (1×10^9 CFU) heat-killed *Lact. delbrueckii* (1×10^9 CFU) or Spent Culture Supernatant (SCS) once daily 0.5 mL per mouse. All of the animals had free access to diets and drinking water. After 7 days, all animals were sacrificed for sample collection. All animal care, handling and surgical techniques followed protocols approved by the Hunan Agricultural University Animal Care and Use Committee before study initiation.

Average Daily Gain (ADG): All mice were weighed individually on 0, 7 days of experiment to measure body weight. The ADG (g/mouse/day) during the experimental stages was calculated as the difference between the initial and final weight.

Serum Immunoglobulin G (IgG) and Interleukin-2 (IL-2) determination: The concentration of serum Immunoglobulin G (IgG) and Interleukin-2 (IL-2) were measured measured using ELISA kit in accordance with the manufacturer's instructions (Cusabio biotech Co., Ltd. China).

Total RNA extraction and cDNA synthesis: Total RNA was isolated from liquid nitrogen frozen jejunum tissues with TRIZOL regent (Invitrogen, USA) and then treated

with DNase I (Invitrogen, USA) according to the manufacturer's instructions. For each sample, the RNA quality was checked by 1% agarose gel electrophoresis, stained with 10 μg mL^{-1} ethidium bromide. Synthesis of the first strand cDNA was performed with oligo (dT) 20 and Superscript II reverse transcriptase (Invitrogen, USA).

Quantification mRNA by real-time PCR analysis: Primers used in this study were presented in previous study (Ren *et al*., 2014). β-actin was used as an house keeping gene to normalize target gene transcript levels. Real-time PCR was performed according the previous study (Ren *et al*., 2013). Relative level of gene expression was normalized and expressed as a ratio of target gene expression to the control group.

Statistical analysis: The results were expressed as mean±Standard Error of the Mean (SEM). All statistical evaluation was performed by using SPSS 17.0 Software. Group comparisons for statistical difference were performed using the one-ANOVA's Duncan (D)-test. Differences ($p<0.05$) were considered significant.

RESULTS

Effect of oral administration of *Lact. delbrueckii* on mouse ADG: Although, the growth promotion of Lactobacillus have been extensively reported, it remains exist variable results with respect to body weight. Interestedly, Bai *et al*. (2013) have reported that dietary supplementation of probiotics can augment ADG of broiler chickens but the research from Fajardo *et al*. (2012) indicate that probiotic-supplemented exhibited no significant effect on Body Weight Gain (BWG) in the present experiment, orally administrated living and heat-killed *Lact. delbrueckii* exhibited no difference ($p>0.05$) on mouse ADG. However, SCS significantly decreased ($p<0.05$) mouse ADG (Table 1).

Table 1: Effect of *Lact. delbrueckii* on Average Daily Gain (ADG) in mice

Catalogue	Initial weight	Final weight	ADG (g day^{-1})
Control	24.90±0.45	31.47±1.00^{a}	0.72±0.06^{ab}
Live	25.00±0.79	31.05±1.24^{a}	0.67±0.07^{b}
Heat-kill	24.98±0.51	32.14±1.12^{a}	0.78±0.74^{a}
SCS	24.98±0.59	29.18±0.91^{b}	0.47±0.04^{c}

The values are expressed as means±MES, n = 8 and the values with different superscripts are significant ($p<0.05$) while values with same superscripts are not significant different ($p>0.05$). All groups of mice were administrated orally respectively with saline normal, live *Lact. delbrueckii* (1×10^{9} CFU) heat-killed *Lact. delbrueckii* (1×10^{9} CFU) or Spent Culture Supernatant (SCS) once daily 0.5 mL per mouse

Effect of oral administration of *Lact. delbrueckii* on serum IgG and IL-2 concentration: Immunoglobulin G (IgG) and Interleukin-2 (IL-2) indispensable components in immunologic response, play important roles in preventing pathogens from invading. In the study, compared with control group, administration of living and heat-killed *Lact. delbrueckii* exhibited no remarkable difference ($p>0.05$) on serum IgG level but SCS remarkably decreased ($p<0.05$) serum IgG level (Fig. 1). Meanwhile, oral administration of living and heat-killed *Lact. delbrueckii* significantly elevated ($p<0.05$) level of serum IL-2 while there was no significant difference ($p>0.05$) between SCS and control group (Fig. 2).

Effect of oral administration of *Lact. delbrueckii* on cytokines expression in jejunum: Intestinal inflammatory cytokines that are secreted by immune cells, e.g., lymphocyte, dentric cell play central roles in intestinal inflammation immunologic response and the expression of cytokines in jejunumis affected by various factors including commensal microbiota (Wang *et al*., 2013). Thus, researchers also detected inflammatory cytokines (INF-γ, TNF-α, IL-13, IL-17) expression in jejunum by RT-PCR. In the study, compared with control group, living *Lact. delbrueckii* significantly augmented ($p<0.05$) the mRNA abundance of TNF-α, INF-γ and IL-17. However, heat-killed *Lact. delbrueckii* and SCS exhibited no

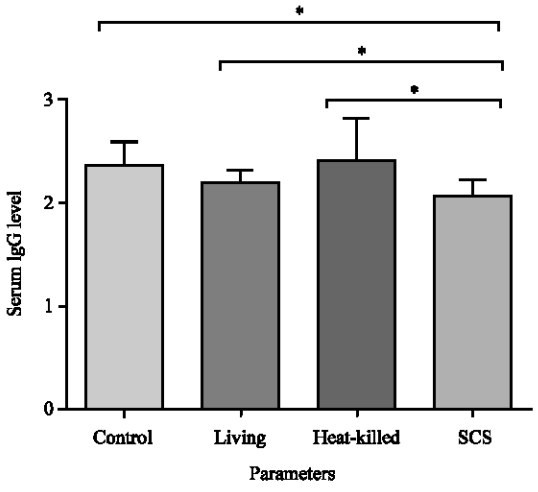

Fig. 1: Effect of *Lact. delbrueckii* on serum IgG concentration in mice. The values are expressed as means±MES, n = 8 and *$p<0.05$. All groups of mice were administrated orally, respectively with saline normal, living *Lact. delbrueckii* (1×10^{9} CFU) heat-killed *Lact. delbrueckii* (1×10^{9} CFU) or Spent Culture Supernatant (SCS) once daily 0.5 mL per mouse

significant ($p>0.05$) effects on these gene expression excepting heat-killed *Lact. delbrueckii* remarked increased ($p<0.05$) mRNA but butabundance of IL-17 (Fig. 3).

Effect of oral administration of *Lact. delbrueckii* on paneth cell and goblet cell function in jejunum: To investigate the effect of *Lact. delbrueckii* on jejunum innate immunity, researchers examined jejunum paneth cell-specific molecules (cryptdin-1, 4, 5, lysozyme C, sPLA2 but and CRS-1C, CRS-4C) and goblet cell-specific molecules (mucin-2 and mucin-4). In the study, compared with control group, SCS remarkably increased ($p<0.05$) mRNA abundance of cryptdin-1, 4 and 5 and sPLA2 (Fig. 4 and 5). Moreover, apart from cryptdin-5 in heat-

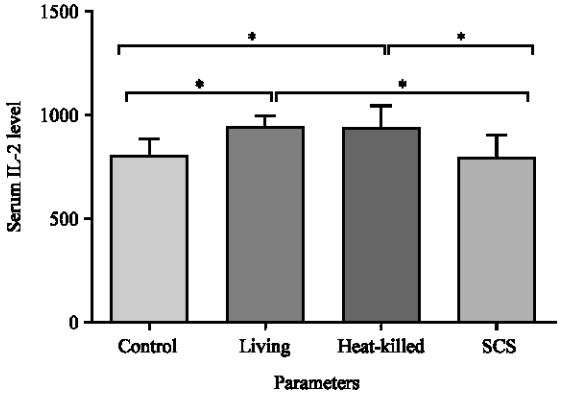

Fig. 2: Effect of *Lact. delbrueckii* on serum IL-2 concentration in mice. The values are expressed as means±MES, n = 8 and *$p<0.05$. All groups of mice were administrated orally, respectively with saline normal, live *Lact. delbrueckii* (1×10^9 CFU) heat-killed *Lact. delbrueckii* (1×10^9 CFU) or Spent Culture Supernatant (SCS) once daily 0.5 mL per mouse

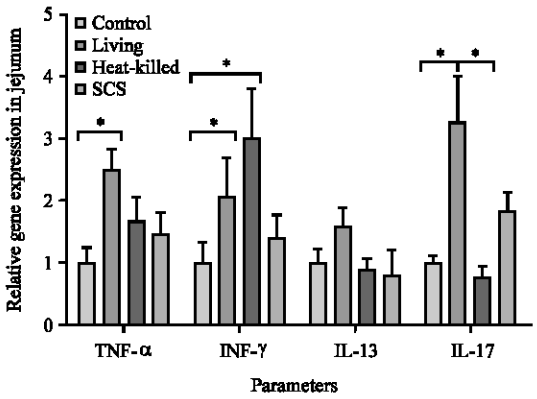

Fig. 3: Effect of *Lact. delbrueckii* on cytokines gene expression in jejunum in mice. The values are expressed as means±MES, n = 8 and *$p<0.05$. All groups of mice were administrated orally, respectively with saline normal, live *Lact. delbrueckii* (1×10^9 CFU) heat-killed *Lact. delbrueckii* (1×10^9 CFU) or Spent Culture Supernatant (SCS) once daily 0.5 mL per mouse

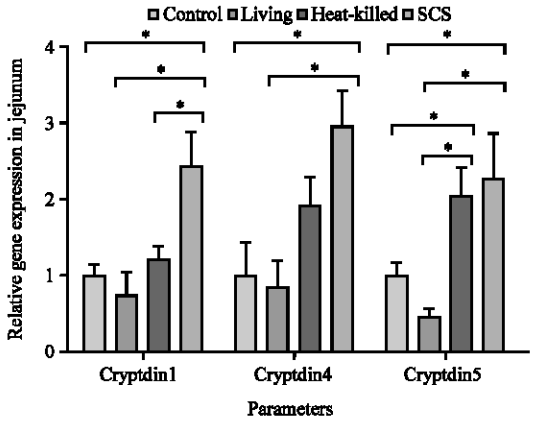

Fig. 4: Effect of *Lact. delbrueckii* on criptdins expression in jejunum in mice. The values are expressed as means±MES, n = 8 and *$p<0.05$. All groups of mice were administrated orally, respectively with saline normal, live *Lact. delbrueckii* (1×10^9 CFU) heat-killed *Lact. delbrueckii* (1×10^9 CFU) or Spent Culture Supernatant (SCS) once daily 0.5 mL per mouse

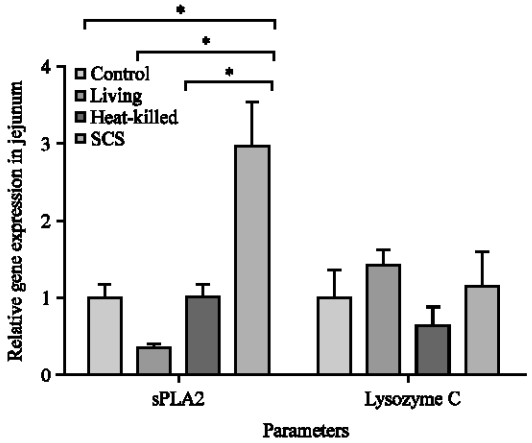

Fig. 5: Effect of *Lact. delbrueckii* on sPLA2 and lysozyme C expression in jejunum in mice. The values are expressed as means±MES, n = 8 and *$p<0.05$. All groups of mice were administrated orally, respectively with saline normal, live *Lact. delbrueckii* (1×10^9 CFU) heat-killed *Lact. delbrueckii* (1×10^9 CFU) or Spent Culture Supernatant (SCS) once daily 0.5 mL per mouse

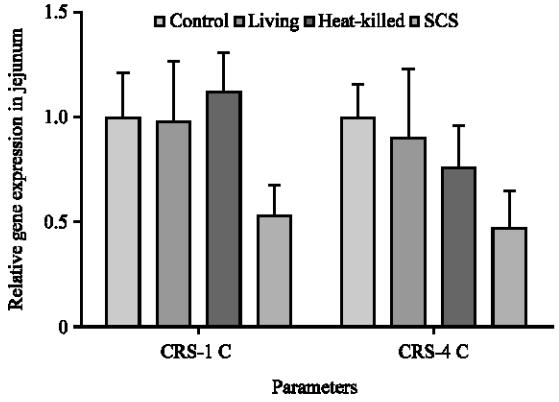

Fig. 6: Effect of *Lact. delbrueckii* on CRS-1C and CRS-4C expression in jejunum in mice. The values are expressed as means±MES, n = 8 and *p<0.05. All groups of mice were administrated orally, respectively with saline normal, live *Lact. delbrueckii* (1×10^9 CFU) heat-killed *Lact. delbrueckii* (1×10^9 CFU) or Spent Culture Supernatant (SCS) once daily 0.5 mL per mouse

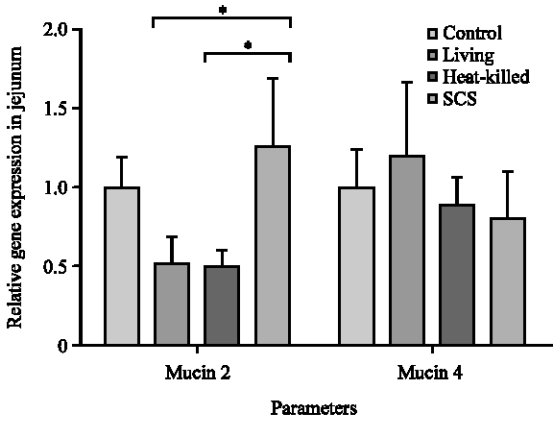

Fig. 7: Effect of *Lact. delbrueckii* on mucin 2 and 4 expression in jejunum in mice. The values are expressed as means±MES, n = 8 and *p<0.05. All groups of mice were administrated orally, respectively with saline normal, live *Lact. delbrueckii* (1×10^9 CFU) heat-killed *Lact. delbrueckii* (1×10^9 CFU) or Spent Culture Supernatant (SCS) once daily 0.5 mL per mouse

killed *Lact. delbrueckii* group, the mRNA abundance of cryptdin-1, 4, 5 and sPLA2 in SCS group were significant higher ($p<0.05$) than that inliving and heat-killed *Lact. delbrueckii* group. However, all treatments exhibited no significant ($p>0.05$) effects on lysozyme C, CRS-1C and CRS-4 C expression (Fig. 5 and 6). In addition, the abundance of mucin 2 mRNAin living and heat-killed *Lact. delbrueckii* group was significantly lower ($p<0.05$) than that in SCS group (Fig. 7) but all treatment exhibited no effect ($p>0.05$) on mucin 4 (Fig. 7) expression.

Lact. delbrueckii that has been wildly used to ferment milk is an important probiotic bacterial and recently has been used to treat some diseases because of its immunomodulation. In recent years, growing focuses have been located at immunomodulation of *Lact. delbrueckii*. Although, increasing numbers of research literatures have shown that *Lact. delbrueckii* enable to enhance intestinal barrier function, balance intestinal immune response and regulate intestinal homeostasis (Valdovinos-Diaz, 2013; Persborn *et al*., 2013; Yang and Sheu, 2012; Howarth and Wang, 2013) the impact of *Lact. delbrueckii* on jejunum innate immunity remains obscurity and needs to be investigated. In the study, oral administration of *Lact. delbrueckii* increased serum IL-2 concentration and stimulated inflammatory cytokines expression and affected paneth cell-specific molecules and goblet cell-specific molecules expression in jejunum in mice.

Although, the growth promotion effects of Lactobacillus have been extensively reported, it remains exist variable results. Interestedly, Bai *et al*. (2013) have reported that dietary supplementation of probiotics augment ADG of broiler chickens but the research from Fajardo *et al*. (2012) indicates that probiotic-supplemented have no difference effect on Body Weight Gain (BWG) (Fajardo *et al*., 2012). In the current research, living and heat-killed *Lact. delbrueckii* exhibited no significant effect on mice ADG, corresponding with Giang *et al*. (2010) report that supplementation of lactic acid bacterial has no effect on performance of weaning piglets. However, there existed other researchs suggesting that probiotics may be improve animal growth performance since supplementation with probiotic bacterial enable to enhance intestinal function and modulate intestinal microbial composition which are important for nutrients absorption (Wang *et al*., 2012). In addition, growth performance effects of probiotics link to strains and dosage of probiotics. Mountzouris *et al*. (2010) have reported that probiotic inclusion levels exhibit a significant effect on broiler growth performance. According to these factors in the study, the differences with other research may be link to the effect of *Lact. delbrueckii* on intestinal bacterial composition and also may be related to strains or dosage of *Lact. delbrueckii*.

The mucosal membrane of the gastrointestinal tract is the first barrier against foreign antigens such as natural

toxins, pathogenic microflora (Oswald, 2006). Thus, intestinal innate immunity is orchestrated to protected host against pathogens invasion, guaranteeing to sustain intestinal homeostasis (Santaolalla *et al*., 2011). Intestinal inflammatory cytokines that are secreted by activated T lymphocytes, macrophages but and decentric cell, plays critical roles in intestinal innate immunity (Wang *et al*., 2013; Heneghan *et al*., 2014). Many studies have shown that supplementation with *Lact. delbrueckii* modulate intestinal innate immunity (Tohno *et al*., 2011) and remarkably stimulate intestinal cytokines production and balance Th1 and Th2 immune response in health or pathological model (Hegazy and El-Bedewy, 2010; Rocha *et al*., 2014; Elmadfa *et al*., 2010) through the NF-κB and MAPK signaling pathways (Thomas and Versalovic, 2010). In this study, administration with living *Lact. delbrueckii* elevate djejunum TNF-α, INF-γ and IL-17 expression, suggesting that orally administrated *Lact. delbrueckii* may be able to induce jejunum Th1-immune response and jejunum inflammatory response. Similarly, many compelling studies also have demonstrated that *Lact. delbrueckii* mediates Th1 cytokines production and stimulate inflammatory response *in vivo* and *in vitro* (Castanheira *et al*., 2007; Neumann *et al*., 2009). Moreover, elevated serum IL-2 concentration in living and heat-killed *Lact. delbrueckii* further confirms this point because IL-2 signaling is involved in proliferation of T lymphocytes cell and Th1 development (Fujimura *et al*., 2013). These results may be link to TLRs-MyD88 mediate various signaling pathway because TLRs-blocking antibodys reduce cytokines production induced by *Lact. delbrueckii* (Cai *et al*., 2010). However, some studies also have reported that administration with *Lact. delbrueckii* significantly inhibits intestinal pro-inflammatory or inflammatory cytokines production, especially in pathological models such as Dextran Sulfate (DSS) induced colitis (Hegazy and El-Bedewy, 2010; Santos Rocha *et al*., 2012). These different results may be linked to different experimental model and experimental conditions. However, to elucidate this point, further studies need to be carried out.

Moreover, antimicrobial peptides from paneth cells and mucins from goblet cells play pivotal roles in intestinal innate immunity. Many studies have shown that paneth cells secrete specific antimicrobial peptides butinto intestinal lumen (Selsted and Ouellette, 2005; Ouellette, 2004; Zhao and Lu, 2014) and exhibits broad spectrum microbicides by membrane-disruptive (Santaolalla *et al*., 2011; Boonzaier *et al*., 2013) therefor protecting host against microbe invasion. In addition, mucins from goblet cellare important components of intestinal mucus layer and play important roles in intestinal innate immunity by enhancing gut barrier function (Johansson *et al*., 2008; Zarepour *et al*., 2013). Many well-designed experiments have shown that probiotics affect intestinal α-defensins, CRS-4C and mucins expression in human and mouse (Underwood *et al*., 2012; Schlee *et al*., 2008; Mondel *et al*., 2009). Indeed, in the current study, orally administrated SCS changed jejunum cryptdin-1, 4 and 5 and sPLA2 expression. However, living and heat-killed *Lact. delbrueckii* exhibited no significant effects on antimicrobial peptide and mucins expression which were different with Moro-Garcia *et al*. (2013) research that *Lact. delbrueckii* could increase human beta definsin-2 expression. Meanwhile, Wehkamp *et al*. (2004) also have demonstrated that probiotic, *Escherichia coli Nissle* 1917, up-regulates human beta definsin-2 through activation of NF-κB and AP-1. As a result, researchers speculate that the different results may be ascribed to bioactive metabolites of *Lact. delbrueckii* such as hydrogen peroxide and lactic acid largely produced by *Lact. delbrueckii* (Batdorj *et al*., 2007; Strus *et al*., 2009). In addition, gut microbial composition also affects intestinal antimicrobial peptide and mucins expression (Salzman *et al*., 2007). Thus, immuno modulation of *Lact. delbrueckii* on jejunum paneth cells and goblet cell function may link to the effect of *Lact. delbrueckii* on intestinal microbial composition (Hemarajata and Versalovic, 2013). Thus, to elucidate this point many researches should be carried out.

Intriguingly, in this study, different treatment with living, heat-killed *Lact. delbrueckii* and SCS exhibited different effects on jejunum inflammatory cytokines and paneth cell-specific molecules expression. Although, living *Lact. delbrueckii* induced remarkable inflammatory response, heat-killed *Lact. delbrueckii* and SCS exhibited no significant effects on inflammatory cytokines expression. These interested results maybe link to the effects of *Lact. delbrueckii* on intestinal microbial composition and the bacteria-mediated fecal metabolites such as Short-Chain Fatty Acids (SCFAs) (Mori *et al*., 2011) which play important roles in regulating differentiation of intestinal T cell population (Furusawa *et al*., 2013). Additional, SCS significantly increased paneth cell-specific antimicrobial peptide expression but other treatment failed to up-regulate these genes expression. With regard to these intriguing results, researchers speculate the possible reason is that bioactive metabolites such as hydrogen peroxide and lactic

acid largely produced by *Lact. delbrueckii* (Strus *et al*., 2009). However, many researches should be conducted to elucidate this point.

CONCLUSION

Researchers found that oral administration of *Lact. delbrueckii* affects serum IL-2 levels, stimulate jejunum inflammatory cytokines expression and affect jejunum paneth cell-specific molecules and goblet cell-specific molecules expression in mice, suggesting that *Lact. delbrueckii* may be able to regulate jejunum innate immunity.

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