Review Article

Lactobacillus: Prevention and Treatment of *Salmonella* Diarrhea

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Salmonellae are gram-negative, non-spore-forming, facultative anaerobic bacilli that measure 2 to 3 by 0.4 to 0.6 μm in size. Like other *Enterobacteriaceae*, they produce acid on glucose fermentation, reduce nitrates, and do not produce cytochrome oxidase[1]. *Salmonellae* can produce both localized and disseminated systemic disease in humans and a variety of other vertebrates. There are more than 2500 known serotypes that belong to two species, namely *Salmonella enterica* (with more than 2400 serotypes) and *Salmonella bongori* (20 serotypes)[2]. The infection with *Salmonella spp, S. typhimurium* is the one of the most common causes of an infection diarrhea. *Salmonella* infections usually happen within 12 to 36 hours after eating contaminated food or water. *S. typhimurium* occurs within the first 1-3 hours of infection, massive neutrophil migration and the secretion of protein-rich exudates into the intestinal lumen do not occur until 8-10 hours following infection and diarrhea begins approximately 8-72 hours after bacterial colonization[3]. The infections contribute the homeostasis and induce the inflammatory response in human host cells[4] and induced diarrhea.

Pathophysiology of *Salmonella* diarrhea

The mechanisms of *Salmonella* infection initiation of *Salmonellae* appear to adhere and blinding toll-like receptors (TLR2 or TRL4) on cell membrane, enter the microfold cells (M cells) of the intestinal epithelium. Although invasion by normally nonphagocytic enterocytes[5]. M cells are specialized epithelial cells that sample intestinal antigens through pinocytosis and transport these antigens to lymphoid cells that underlie the epithelium in Peyer's patches[6]. *Salmonella* invade non-phagocytic cells by inducing membrane ruffles on the plasma membrane. The important mechanism for induce membrane ruffles is *Salmonella* pathogenicity island-1 (SPI-1) encodes a type III secretion system that are involved in bacterial entry into the epithelial cells. The secretory proteins of SPI-1 contribute to actin rearrangements and invasion during the entry in host cells[7]. SPI-1 has additional functions related to the activation of innate immune pathways. SPI-1 dependent inflammation appears to reflect multiple processes:[5] the induction of PMN recruitment across intestinal epithelia by the SPI-1 secreted effector protein SipA[11], the activation of NF-kB signaling by the concerted activity of SPI-1 translocated effectors, and the activation of caspase-1 mediated IL-1b/IL-18 activation and pro-inflammatory cell death by the SPI-1translocated effector SipB[8].

After entry in to host cell *Salmonella* have the survival strategies, intravascular survival and replication dependent on *Salmonella* pathogenicity island-1 (SPI-1) encoded T3SS1 (Types III secretion system I) or T3SS2 (Types III secretion system II) to modified phagosome called the *Salmonella* containing vacuole (SCV)[9]. *Salmonella* containing vacuole (SCV) contributes the homeostasis in host cells, activated of NF-κB signaling pathaway, and activated the expressions of pro-inflammatory genes and release pro-inflammatory cytokines (interleukin-6 (IL-6), interleukin -1b (IL-1b), tumor necrosis factor-alpha (TNF-α), and interferon-gamma (IFN-γ)), induces inflammatory response

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and cytotoxicity of host cells[10-14].

Probiotic treatment and prevention *Salmonella* diarrhea

Probiotic are live microorganisms naturally, which, when administered in adequate amounts, Confer a health benefit on the host[15], present in the human digestive tract which maintains a correct balance of intestinal flora[16]. Many strains of probiotic (e.g., *Lactobacillus* strain: *Lactobacillus rhamnosus* GG, *Lactobacillus reuteri,* certain strains of *Lactobacillus casei, Lactobacillus acidophilus, Escherichia coli* strain Nissle 1917, and certain *bifidobacteria* and *enterococci* (*Enterococcus faecium* SF68 and *yeast Saccharomyces boulardii*) have been can to inhibit the growth, metabolic activity and adhesion to intestinal epithelium cells of pathogenic enteric bacteria (*Salmonella, Shigella, E. coli, or Vibrio cholerae*)[17]. The potential mechanisms of probiotic for prevention and treatment of diarrhea are protection of intestinal epithelial barrier function, regulation of intestinal epithelial homeostasis, regulation of intestinal microbial environment, modifications to commensal and probiotic bacteria to enhance diarrhea prevention.

Lactobacillus bacteria are the gram-positive bacteria in the *Lactobacilla* ceae family. *Lactobacillus* can called fermentative lactobacilli or lactic acid bacteria (the properties in sugar fermentation and produce lactic acid). *Lactobacillus* is the normal flora bacteria can find in the human gastrointestinal tract and the reproductive system[18]. In food industry *Lactobacillus has the* benefit in fermentative food production and beverages such as yogurt, cheese, pickles, beer, wine, cider etc. In the medicinal *Lactobacillus* mostly use as probiotic and biotherapeutics for the prevention and treatment the disease or symptoms including gastrointestinal disease, *Salmonella* infection and diarrhea [19-21].

Lactobacillus inhibit the growth, metabolic activity and adhesion to intestinal epithelium cells of *Salmonella* infection

Lactobacillus can produce bactericidal proteins and organic substances such as organic acids (lactic acid, acetic acid, butyric acid) that decrease the intestinal lumen pH, inhibit pathogenic adherence to intestinal epithelial cells, the study of *Lactobacillus crispatus K313 and K243* that shown can inhibit *Sal-*

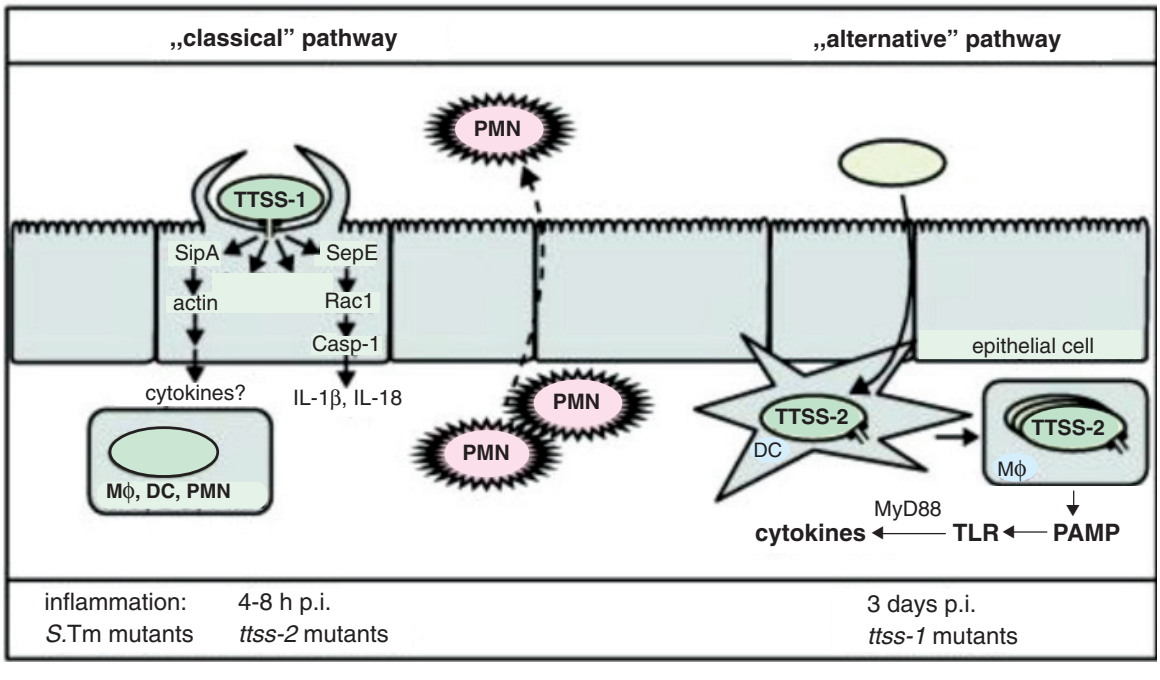

Figure 1. *S. typhimurium* can trigger gut inflammation by at least two different mechanisms: a) direct triggering of pro-inflammatroy responses by injection of bacterial virulence factors; b) by entering the mucosa via dendritic cells ($CX3CR1^{+}CD11c^{+}$ phagocytes), TTSS-2 dependent replication in the mucosal tissue and triggering of innate immune responses[4].

monella infection, anti inflammatory and high adhesion on intestinal epithelial cells by competition, exclusion or displacement the pathogenic bacteria[22], and confer enhanced protection against *S.typhimurium* infection[23-26].

Block epithelial attachment or invasion of pathogenic enteric bacteria

Lactobacillus can produce the bacteriocin and organic substance (lactic acid, acetic acid, butyric acid) which act at low pH in intestinal lumen, inhibit growth and invasion of pathogenic enteric bacteria. In 2008 the study of *Lactobacillus acidophilus* LAP5 in vitro, shown the result Lactobacillus acidophilus LAP5 was able to inhibit the invasion of S. choleraesuis to human Caco-2 cell line. *Lactobacillus acidophilus* LAP5 decrease the pH (produce organic acids and bacteriocin) which act at low pH conditions and play the role in inhibit *S. choleraesuis* invasion[27]. *Lactobacillus* blocks epithelial binding of pathogenic enteric bacteria by inducing of MUC2 expression, stimulate the mucus production and increase the releases of biofilm. The production of mucus bulling the thick layer, enhance the protection and inhibit the pathogenic invasion[28].

Improve epithelial and mucosal barrier function

The mechanism of *Lactobacillus* in improve epithelial function are enhance mucus production and increase barrier integrity to repaired and protect the epithelia[28].

Alter the immune response

Lactobacillus has the potential anti-inflammatory and immunomodulatory. Can activate innate immune response[29], decrease the inflammatory response, and decrease the expression of pro- inflammatory genes including decrease the secretion of pro- inflammatory cytokines. (e.g. IFN-γ, TNF-α). And increase the expression of anti- inflammatory genes; induced the secretion of anti- inflammatory cytokines (e.g. IL-4, IL-6, IL-8, and IL-12). The study in *vitro* of live *Lactobacillus plantarum* L2 show that *Lactobacillus* has the potential to reduced inflammatory response in TNF-α-induced Caoco-2 cell induced with TNF-α, *Lactobacillus plantarum* L2 inhibit of NF-κB translocation to the nucleus and the associated decrease in IκB. (down-regulation of IκB degradation) and inhibit the release of pro-inflammatory cytokines from TNF-α-induced cells[30]. In 2012 the study the effect *of Lactobacillus plantarum* Lp91 in colitis mouse model, suggest Lactobacillus plantarum Lp91 is the strong immunomoduratory efficacy. Induce the expression of anti-inflammatory genes (COX-1, IL-4, IL-6, IL-10 and MUC2), increase the secretion of anti- inflammatory cytokines and decrease the expression of pro-inflammatory genes (TNFα, COX-2) include decrease pro-inflammatory cytokines release. *Lactobacillus plantarum* Lp91 able to reduced inflammatory response, use as anti-inflammatory and immunomodulatory probiotic[31].

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