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Lactobacillus fermentum (MTCC-5898) supplementation renders prophylactic action against *Escherichia coli* impaired intestinal barrier function through tight junction modulation

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ABSTRACT

Dietary intervention using probiotic bacteria has emerged a promising therapeutic strategy to curb gastrointestinal diseases. To test this hypothesis, present investigation was aimed to assess *in vitro* prophylactic capability of probiotic *Lactobacillus fermentum* (LF: MTCC-5898) against *Escherichia coli* (ATCC 14948) impaired barrier function using human intestinal Caco-2 cells. *L. fermentum* adhered strongly with Caco-2 cells (1850 ± 104.1 CFU/100 cells) and maintained host barrier integrity in contrast to leaky conditions induced by *E. coli* exposure characterised with high phenol red (6.73 ± 0.3%) flux concomitantly decreased TEER (0.69 ± 0.01). Intestinal cells treated with *E. coli* also displayed significantly ($p < 0.05$) reduced mRNA levels of key tight junction genes in contrast to their significantly ($p < 0.05$) elevated levels upon probiotic treatment. However, *E. coli* impaired intestinal integrity was restored significantly ($p < 0.05$) when intestinal cells were treated with *L. fermentum* either during exclusion or competition than displacement assays through increased mRNA levels of junction genes along with preserved localisation and distribution of their corresponding proteins as revealed by immunofluorescent and electron micrographs. Collectively, probiotic *L. fermentum* demonstrated *in vitro* prophylactic capability to rectify impaired barrier functions through tight junction gene modulation and therefore, could be safe and potential therapeutic agent for management of gut associated problems.

1. Introduction

Functional foods supplemented with live probiotic, prebiotic or other biologically active ingredients are gaining considerable resurgence in modern nutritional science for their potential role in the prevention and treatment of diseases associated with gastrointestinal track or other visceral organs beyond their basic nutrition value. Due to overwhelming health benefits associated with probiotic intake, a continuous growing consumption demand has enhanced the probiotic market worldwide and a keen interest has emerged in identifying such microbes (Panghal et al., 2018). Although introduced in early 20th century, science of probiotics has emerged as one of hot research area in modern therapeutic industry in past two decades. Different probiotics strains have shown ability to improve the nutrient absorption/assimilation, enhance the host ability to synthesize vitamin B and improve the calcium and lactose absorption efficacy (Raghuwanshi, Misra, & Sharma, 2018). An essential utility of probiotic microbes have been their ability to modulate the host gut microbiota composition whose imbalanced composition contributes to pathogenesis of both intestinal and extra-intestinal disorders (Carding, Verbeke, Vipond, Corfe, & Owen, 2015). Therefore, curbing the factors responsible for dysbiosed gut microbe composition could have immense impact on overall human health. Fortunately, nutritional interventions through probiotic intake in the form of fermented food product or lyophilised culture offers a suitable alternative to mitigate such health complications; however, more investigations are necessary to clearly comprehend the multifaceted probiotic actions involved in mediating host benefits.

For healthy gut functions, intestinal barrier integrity is prerequisite as it is critical for efficient defensive reactions against various challenges and hence has been often employed as a key target for pathogen invasion (Konig et al., 2016). Therefore, impaired intestinal integrity has been often observed during pathogenic exposure or even under dysbiosed guts microbiota composition (Saltzman, Palacios, Thomsen, & Vitetta, 2018). During such exposures a rigorous uncontrollable

Abbreviations: CFU, Colony forming unit; DMEM, Dulbecco's modified Eagle's medium; LF, *Lactobacillus fermentum*; SEM, Scanning electron microscopy; TJ, Tight junctions; TEER, Transepithelial electrical resistance

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