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## β-Galactomannans protect epithelial barrier function disruption induced by Salmonella Enteritidis in human intestinal Caco-2 cells

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The gastrointestinal epithelium is a selective barrier that allows the absorption of nutrients, electrolytes and water, but restricts the passage of larger, potentially injurious compounds such as allergens, toxins and pathogens. A defective epithelial barrier has been reported as an important pathogenic factor in many infectious, ischemic, and immune-mediated intestinal diseases and to significantly contribute to water loss in diarrhoea. Mannan oligosaccharides are mannose rich substrates, firstly obtained from yeast *Saccharomyces cerevisiae* cell walls, which are described to agglutinate Gram negative bacteria thus exerting beneficial effects in the prevention of intestinal infections. Recently, we have observed that  $\beta$ -galactomannans obtained from the carob bean gum of the *Ceratonia silliqua* tree, are able to reduce *Salmonella* Typhimurium adhesion to ileal pig cells *in vitro* and the expression of proinflammatory cytokines and chemokines induced by *Salmonella* infection<sup>(1)</sup>.

The aim of this study was to investigate the potential role of different products containing  $\beta$ -galactomannans derived from *Ceratonia silliqua* and *Cassia obtusifolia* to prevent the disruption of epithelial barrier function induced by the colonization of *Salmonella enterica* subsp. *enterica* serovar Enteritidis (*Salmonella*) in intestinal human Caco-2 cells in culture.

Differentiated intestinal Caco-2 cells (ATCC) were incubated with *Salmonella* (provided by Ignacio Badiola, Centre Recerca en Sanitat Animal, CReSA; IRTA-UAB, Bellaterra, Spain) in the apical compartment during 3 h (MOI: 10-100). To test the effect of  $\beta$ -galactomannans, the products (apical compartment:  $10-100\,\mu\text{g/ml}$ , ITPSA, Barcelona) were preincubated during 30 min before *Salmonella* infection. Cell viability was assessed from lactate dehydrogenase (LDH) determination in the culture medium and epithelial barrier function, from transepithelial electrical resistance (TER) and D-mannitol fluxes as previously described.

To establish the experimental conditions of epithelial barrier disruption, first, TER and D-mannitol fluxes were determined in the presence of increasing MOI and  $\beta$ -galactomannan concentrations in parallel to LDH release to the incubation medium. In infected cells, the results showed a significant decrease in TER and increase in D-mannitol fluxes, thus indicating an increase in paracellular permeability of the monolayer. Moreover, the addition  $\beta$ -galactomannans was able to prevent the effects of *Salmonella* on TER and D-mannitol fluxes. In conclusion, the results obtained give an additional value to mannan oligosaccharides due to its ability to prevent epithelial barrier function disruption in *Salmonella* infection.

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1. Badia R et al. (2012) Clin Vaccine Immunol 19:368-376.