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Abstracts

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Competitiveness (BES-2014-068134). P.T-C is the recipient of a fellowship from the Spanish Ministry of Economy and Competitiveness (AGL2013-48459-P). P.T-C. and M.A-G. carried out the experiments; P.T-C. and B.G-P., analyzed the data and wrote the abstract; A.F., F.J.P-C. and M.C. reviewed it.

**Keywords:** Growth factors, suckling rat, lymphoproliferation, mesenteric lymph nodes, cytokines.

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**References**


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Growth Factors Supplementation Influences the Intestinal Lymphocyte Functionality in Suckling Rats

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The newborn immune response is functionally deficient and less competent compared with adults [1]. Breast milk provides a large number of compounds that contribute to the maturation of the immune system in early life such as specific growth factors [2]. The aim of the present study was to ascertain the influence of transforming growth factor (TGF)-β2, epidermal growth factor (EGF) and fibroblast growth factor 21 (FGF21), which are present in breast milk, on the development of the intestinal immune system during suckling. For this, newborn Wistar rats were randomly distributed into four experimental groups: Reference, TGF-β2, EGF and FGF21. Rats were daily supplemented by oral gavage with these growth factors since the day of birth until the end of the suckling period (day 21). At days 14 and 21 of life, lymphocytes from mesenteric lymph nodes were isolated, cultured and their proliferative ability as well as their cytokine secretion were evaluated. The results showed that only FGF21 supplementation on day 21 was able to promote the lymphoproliferation. Regarding cytokine production, it was on day 21 when changes were more patent. EGF and FGF21 decreased IL-13 levels whereas both TGF-β2 and EGF reduced IL-10 and IL-4 production. Overall, EGF decreased the IL-10/TNFα ratio and increased the Th1/Th2 (IFNγ/IL-4) ratio when compared to the reference group. These results evidence that TGF-β2, FGF21 and mainly EGF supplementation have an immunoregulatory effect in early life promoting the switch from Th2 to Th1 responses and then contributing to the maturation of neonatal intestinal immune system.

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**Authorship:** A.F., F.J.P-C. and M.C. designed the study; P.T-C., B.G-P., and M.A-G. carried out the experiments; P.T-C. and B.G-P., analyzed the data and wrote the abstract; A.F., F.J.P-C. and M.C. reviewed it.

**Keywords:** Growth factors, suckling rat, lymphoproliferation, mesenteric lymph nodes, cytokines.

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Probiotic Properties of EPS-Producing Pediococcus Strains

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Pediococcus parvulus 2.6R [1] and Pediococcus ethanolidurans ZEp are two exopolysaccharide-producing lactic acid bacteria (LAB) isolated from Basque country cider. Both pediococci produce a homopolysaccharide (HoPS) characterized as 2-substituted (1,3)-β-D-glucan with probiotic properties [2]. In addition, P. ethanolidurans ZEp also synthesizes a heteropolysaccharide (HoPS), which is composed of glucose, galactose, N-acetylglucosamine and phosphoglycerate. Chemical mutagenesis generated the isogenic strains: P. parvulus 2.6NR [3] and P. ethanolidurans ZEXr with null or reduced 2-substituted (1,3)-β-D-glucan production, respectively.

The aim of this work was to characterize the influence of the exopolysaccharides on the probiotic properties of these LAB as well as the immunomodulatory properties of the 2-substituted (1,3)-β-D-glucan. As expected from previous results [4] for P. parvulus strains, synthesis or addition of the HoPS resulted in a significant increase of the bacterial adhesion to human Caco-2 cells, while for P. ethanolidurans strains the binding to the enterocytes was higher for ZEXr than for ZEp. The positive influence of the HoPS on P. parvulus adhesion to intestinal cells was validated using an in vivo gnotobiotic zebrafish model and fluorescently labelled 2.6R and 2.6NR strains. In addition, in this model 2.6R showed a greater ability than 2.6NR to compete with Vibrio anguillarum, protecting zebrafish against infection. Finally, in vitro/vivo studies with either human cell lines or zebrafish, plus determining levels and expression of cytokines indicated an anti-inflammatory effect of the 2-substituted (1,3)-β-D-glucan. Therefore, P. parvulus 2.6R and its HoPS seem to have potential to produce functional food designed for people with intestinal disorders.
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Authorship: A. Pérez-Ramos performed the experimental work regarding the P. parvulus strains and their HoPS and wrote the paper. M.G. Llamas performed the work related to P. ethanolidurans strains and their EPS. M.L. Mohedano contributed to the design of the interactomic studies. M.A. Pardo was responsible for the design of the experiments involving the zebra fish embryos. M.T. Dueñas designed and analysed the experiments related to P. ethanolidurans strains and their EPS. P. López contributed to the design and analysis of the experiments performed with P. parvulus strains and their HoPS.

Keywords: Lactic acid bacteria, probiotics, polysaccharides, Pediococci, β-glucan.

References


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A Multispecies Probiotic Improves Cognitive Function, Risk of Falls and Inflammatory Response in Cirrhotic Patients

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Aim: To evaluate the effect of the multispecies probiotic mixture De Simone Formulation [DSF] on cognitive function, risk of falls and inflammatory response in patients with cirrhosis.

Patients and Methods: In this double-blind placebo-controlled clinical trial (NCT01686698), we included outpatients with cirrhosis and cognitive dysfunction (Psychometric Hepatic Encephalopathy Score [PHES] <4) and/or falls in the previous year. Patients were randomized to receive DSF (Vivomixx®) one sachet containing 450 billion bacteria bid for 12 weeks or placebo. We evaluated the changes in cognitive function (PHES), risk of falls (gait speed, Timed Up & Go-test [TUG] and incidence of falls), systemic inflammatory response (CRP, TNF-α, IL-6, IL-10, neutrophil oxidative burst), bacterial translocation (serum bacterial DNA and lipopolysaccharide binding protein [LBP]), intestinal barrier (fatty acid binding protein [FABP]-6 and 2 and zonulin in serum and urinary claudin-3), and fecal microbiota.

Results: We included 36 cirrhotic outpatients. Patients treated with DSF showed a significant improvement in PHES (p = 0.01), gait speed (p = 0.03), TUG (p = 0.02) and a trend to a lower incidence of falls during follow-up (0% vs. 22.2% in the placebo group, p = 0.10). In the probiotic group, we observed a decrease in FABP-6 (p = 0.009), claudin-3 (p = 0.002), CRP (p = 0.01) and TNF-α (p = 0.01), and an increase in poststimulation neutrophil oxidative burst (p = 0.002). No significant changes in serum bacterial DNA and LBP or fecal microbiota were observed.

Conclusions: The multispecies probiotic DSF improves cognitive function, risk of falls, inflammatory response and intestinal barrier in patients with cirrhosis and cognitive dysfunction and/or previous falls.

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Financial Support: This study has been partially funded by VSL Pharmaceuticals and Mendes SRL.

Authorship: Eva Román and Germán Soriano formulated the research questions, designed the study and analysed the data and they wrote the abstract.

Juan Camilo Nieto and Silvia Vidal performed the analysis of the parameters of systemic inflammatory response, intestinal barrier and oxidative burst.

Germán Soriano and Carlos Guarner selected the outpatients in Hospital de Sant Pau.

Eva Román carried out the tests of cognitive function, risk of falls, quality of life and collected all the samples.

Chaysavanh Manichanh analyzed the fecal microbiota at the Vall d’Hebron Research Institute.

Keywords: Probiotics, cognitive function, cirrhotic patient, risk of falls, inflammatory response.