ABSTRACT SUPPLEMENT

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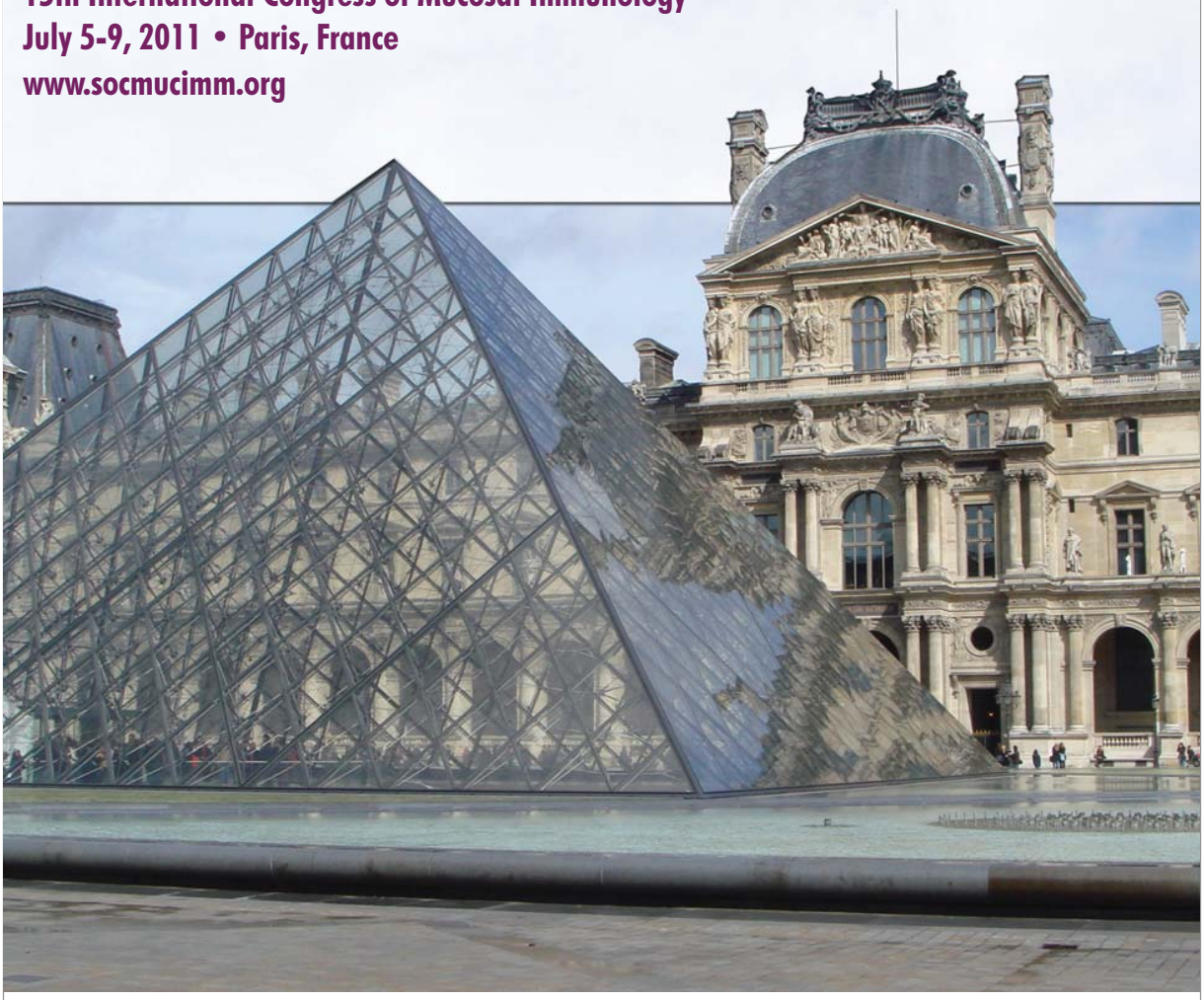

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fermented milk (PFM) in an allergy mice model. Experimental groups: normal-control (NC), Basal (B-5days-PFM); OVA-Sensitization-control (SC), Previous (P)(5d-PFM+OVA+H2O) and Continuous (C)(5d-PFM+OVA+PFM) treatment. At 7 and 15 days post-sensitization (dps) we analyzed: specific-IgE, specific-IgG and IL10 in serum. Non-specific-IgA, IL10 and IFNγ in intestinal fluid (IF), and IL2+,IL4+ and IL10+ cells in the small intestine (SI). IgA+ cells in SI and bronchus and changes in the intestinal microbiota. At 7dps, specific-IgE decreased and IL10 increased in P and C groups compared with SC. IL10+ cells were increased for 7 and 15dps, however IL10 and IFNγ release was not enhanced. The number of IL2+ cells was similar to the NC. IL4+ cells decreased for 15dps in treated groups but not for SC. For 7 and 15dps total s-IgA levels were higher than NC. IgA+ cells from SI and bronchus were increased in C group. Changes in the intestinal microbiota were observed with diminution in enterobacteria, and increases of bifidobacteria populations in the treated groups. PFM was able to regulate IgE levels, by a decrease in the number of IL4+ cells in SI and increases of IL-10. No variations in total s-IgA were found. The immune regulation observed for PFM in this allergy model could be mediated by the immune regulatory capacity exerted by bifidobacteria.

F.75. Improved Response of Intestinal Epithelial Cells Against Salmonella Enterica Serovar Typhimurium Infection in Mice Fed with Lactobacillus Casei CRL431

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Lactobacillus casei CRL431 (Lc) protected against Salmonella enterica serovar Typhimurium (ST) infection modulating the immune cells in a mouse model. Intestinal epithelial cells (IECs) participate in the early response against ST. We evaluate the IEC response to IL-6 and MPC-1in mice received Lc and were infected with ST. Lc7d-group received Lc during 7days (d), was challenged with ST, Lc administration continued until 10d post-infection (PI). IECs were isolated from treated, untreated (UC) and infected (IC) controls mice. IECs from UC were cultured with ST or Lc for *in vitro* tests. In the *ex vivo* assay IL-6 and MCP-1 release increased for Lc7d-group compared to UC and to IC after 24h PI. IC increased IL-6 levels for 7 and 10d in PI. *In vitro*, IECs cultured with Lc released higher levels of IL-6 than IECs cultured with ST. These results show the IEC as another important source of IL-6, for B-cell clonal expansion and specific-anti-ST-IgA-s production, involved in the protection against ST exerted by Lc. The results obtained for MCP-1showed the participation of macrophages in the protective effect of Lc. ST Infection increased intestinal neutrophils influx and mieloperoxidase activity, which diminished in Lc7d-group. Lc administration induced an earlier response of the IECs with IL-6 and MCP-1 secretion, increasing the protective barrier of the IECs, first step of this infection.

F.76. Effect of Precolonization with the Mixture of Lactobacillus Casei and Lactobacillus Paracasei Strains on the Development of Experimental Birch Pollen Allergy in Mice

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Disbalanced bacterial colonization in early life affects allergies development later on. We studied the effect of precolonization with the mixture of L. casei and L. paracasei on the development of allergic sensitization to the main allergic component of birch pollen Bet v 1 in mice. Originally germ-free (GF) mice were colonized with mixture of L. casei 900, L. casei 908, L. paracasei 919 strains and after 20 days colonized mice and age-matched GF controls were repeatedly immunized with Bet v 1. Allergen-specific antibody levels and transforming growth factor (TGF)-beta were determined in sera. Th1/Th2 cytokines were evaluated after 48 h cultivation of spleen and mesenteric lymph node cell cultures. Mice colonized with lactobacilli mixture showed significantly lower value of Bet v 1-specific IgG1, IgG2a and IgE and stimulated levels of total IgA in sera and intestinal lavages accompanied by increase of TGF-beta compared to the GF sensitized group. In colonized mice, proallergic IL-5 was reduced markedly and regulatory TGF-beta was found stimulated. Colonization with lactobacilli mixture inhibited the development of allergic immune responses and increased the production of regulatory cytokine TGF-beta and can be thus exploited for prevention of allergic sensitization. Supported by grants IAA500200710 and CZ.3.22/2.1.00/09.01574.

F.77. A Multi-strain Probiotic Reduces Inflammation via the Mitogen Activated Protein Kinase Pathway in HT-29 Cells Challenged with a Double-stranded RNA Viral Mimic

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Our focus was to determine the anti-inflammatory properties of a bacterial mixture, ProbioKid (PK), made with Bifidobacterium infantis R0033, B. bifidum R0071 and Lactobacillus helveticus R0052. Intestinal epithelial cells (HT-29) were challenged 3 h with the double-stranded RNA viral mimic, polyinosinic acid-poly cytidylic acid [Poly (I:C); 10 µg mL-1]. PK was added 1 h before (-1 h), simultaneously (0 h) or 1 h after (+1 h) stimulation with Poly (I:C). An expression Immune Array was used to screen innate pathways involved in HT-29 cell response to the co-challenge. Gene expression was confirmed by Q-RT-PCR and IL-8 secretion was quantified by ELISA after 3h or 6 h stimulation respectively. Results were compared to the impact of PK additions (-1 h, 0 h & +1 h) on stimulated cells response. PK reduced a number of genes in stimulated cells from MAPK signalling pathway leading to the down-regulation of the transcriptional factor AP-1 (JUN & FOS), but not from the NFκB transcriptional factor. Indeed, PK had an anti-inflammatory effect; IL-8 expression and secretion were significantly reduced in stimulated cells as were IL28B, IL29, CXCL10 and TNF-α genes. We concluded down-regulation of the MAPK signaling pathway was the most important in response to the co-challenge.