ORAL IMMUNITY
Wednesday, July 8

W.22. The Mesenteric Lymph Nodes are Essential for the Regulation of Cholera-toxin Specific IgA Responses in the Intestine, Whereas the Spleen Reacts Systemically by Specific IgM

Manuela Ahrendt, Anika Hahn, Reinhard Pabst, Ulrike Bode
Hannover Medical School, Hannover, Germany

Stimulation of the adaptive immune system in the gut is thought to be mainly initiated in the mesenteric lymph nodes (mLN) and results in an IgA secretion of plasma cells in the lamina propria. However, a detailed role of the mLN in the development of IgA immune responses is poorly understood. When the mLN of rats are removed, the lymph from the gut flows directly into the blood, whereby the spleen becomes an important lymphoid organ to initiate immune responses. Thus, cholera toxin (CT) was applied in mLN resected animals and the IgA response to CT in the gut lavage and serum and possible involvement of the spleen were examined. The CT-specific IgA in the gut lavage but also the specific IgM level in the serum were increased in mLN resected rats. The mLN are therefore critical for the regulation of a specific IgA response. In the spleen of mLN resected animals proliferation was up-regulated forming germinal centers in the follicles. However, CT-specific IgM+ cells, but no IgA+ cells were documented. Thus, the spleen is involved in the immune response against CT after mLN resection, leading to a specific IgM, but not IgA response.

W.23. Increased Number of B220+ Dendritic Cells and IL-10 Producing CD4+ T Cells in Inflamed Gingiva with Bone Loss

Tetsuro Kono[1], Ryoki Kobayashi[1], Yoshiko Fukuyama[1], Rebekah Gilbert[1], Keiko Fujihashi[1], John Ruby[1], Moriyasu Wada[2], Masafumi Yamamoto[2], Kohtaro Fujihashi[1]
[1]University of Alabama at Birmingham, Birmingham, AL; [2]Nihon University School of Dentistry at Matsudo, Matsudo, Japan

Cellular and molecular mechanisms of the immune system influencing oral bone metabolism remain to be elucidated. In this study, we characterize the mucosal immune cells in the inflamed gingiva of mice with alveolar bone reduction. A murine periodontal disease model with alveolar bone loss was established by oral infection with *Porphymonas gingivalis* (*P. gingivalis*) suspended in 100μl PBS with 2% carboxymethylcellulose 15 times over three weeks. Gingival mononuclear cells (GMCs) were isolated and cultured for analysis of IL-6 and TNF-α production. An aliquot of GMCs were subjected to FACS analysis. Significantly high level of IL-6 and TNF-α production was seen in the culture of GMCs from mice infected with *P. gingivalis*. The frequencies of ICAM-1 expressing $CD11c^+B220^+$ dendritic cells (DCs) were increased in inflamed gingival tissues with bone loss. Further, $CD4^+CD25^+$ regulatory T (Treg) cells were increased in the experimental group. Intracellular cytokine analysis revealed an increased number of IL-10 producing $CD4^+$ T cells in inflamed gingiva when compared with control group. These results show the potential roles of DCs and Treg cells for the induction and regulation of alveolar bone loss in *P. gingivalis* infection. This work is supported by NIH grants DE 12242 and AG 025873.

W.24. High-cholesterol Diet Accelerates Inflammatory Cytokine Production in Apolipoprotein E-null Mice Infected with *Porphyromonas Gingivalis*

Shinichi Sekine[1], Muneo Tanaka[1], Mio Iwasaki[1], Masahiro Toe[1], Ei Hashino[1], Kosuke Kataoka[2], Kohtaro Fujihashi[2], Satoshi Shizukuishi[1]
[1]Osaka University Graduate School of Dentistry, Osaka, Japan; [2]University of Alabama at Birmingham, Birmingham, AL

Our previous studies showed *Porphyromonas gingivalis* (*P.g.*) accelerate atherosclerotic lesion development in hyper-lipidemic animals. Several studies have reported that high-cholesterol diet affects the progression of atherosclerotic cardiovascular disease, diabetes and periodontitis. We examined the effect of high-cholesterolol diet and infection of periodontal pathogen on inflammatory cytokine production. Apolipoprotein E-deficient mice were divided into 4 groups. Two groups were fed a regular diet and the remaining two groups were fed a high cholesterol diet. One of each dietary group was inoculated with *P.g.* for 3 weeks (*P.g.*) or pyrogen-free PBS. Plasma samples were subjected to lipid assays and inflammatory cytokine-specific ELISA. Significant increases in body weight and blood total cholesterol were noted in the high-cholesterol groups regardless of their *P.g.* infection. Both *P.g.* infected with regular diet and high cholesterol with no-*P.g.* groups resulted in increased levels of IL-6 and TNF-α production in blood when compared with those of non-infected with regular diet group. Interestingly, these inflammatory cytokines were synergistically increased in the high-cholesterol diet with *P.g.* group. These results suggested that *P.g.* infection with high cholesterol diet accelerates inflammatory cytokine production that may facilitate induction of periodontitis. We are currently testing progression of atherosclerosis and periodontitis. Supported by NIH grants DE12242, AG025873 as well as Grants-in-Aid A-19209064, A-20390535, A-20890123.

W.29. Mucosal Immunity Against Candida Albicans Induced by Sublingual Immunization

Durdana Rahman, Mukesh Mistry, David Moyes, Ayesha Islam, Julian Naglik, Stephen Challacombe
King's College London, London, United Kingdom

We have shown previously in a low-oestrogen murine model of oral and vaginal Candida albicans colonisation, that intranasal (I/N) immunisation with secreted aspartyl proteinase (SAP2) resulted in concurrent inhibition of candida colonisation and elicited both salivary and vaginal IgA antibodies (Rahman *et al.*, 2007). The aim of this study was to determine whether sublingual (S/L) immunisation with SAP2 could prevent both oral and vaginal colonisation and to compare with I/N. Mice were immunised at weekly intervals at weeks 0,1 and 2 and challenged

with Candida albicans at week 5. Samples were taken at weekly intervals. Both S/L and I/N immunisation with SAP2 led to significantly reduced cfu in both sites compared with the controls and protected mice from both oral and vaginal candidiasis. Reduction in cfu orally in immunised mice was evident by week 2 and maximal by week 4 ($p<.001$). Reduction in vaginal cfu was observed at week 2 but profound by week 5 ($p<.001$). Sublingual immunisation was shown for the first time to be an effective route of mucosal immunisation against candida. These results suggest that S/L immunisation with SAP2 is at least as effective as I/N in inhibition of mucosal candida colonisation.