ANIMAL MODELS OF IBD 2
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OR.61. The Impact of HDAC Inhibition on T Helper Cell Activation in Chronic Intestinal Inflammation

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An anti-inflammatory potency for HDAC inhibitors was demonstrated by our group, using different models of experimental colitis. To evaluate the mechanisms involved, $CD4^+$ T cells were treated *in vitro* with the HDAC inhibitor ITF2357, resulting in a suppression of pro-inflammatory cytokines, associated with an increase in histone acetylation. However, the generation of $FoxP3^+$ cells could be enhanced while the polarization to Th17 cells was suppressed in the presence of ITF2357. To identify the responsible HDAC, the HDAC expression pattern was analyzed in the respective Th cell subsets as well as in $CD4^+$ T cells isolated from the colon, spleen or peripheral lymph nodes from controls or mice with colitis. HDAC 5 and 9 were expressed multiple times higher in Th17 and $FoxP3^+$ cells when compared to Th1 or Th2 cells. Both HDAC were similarly upregulated in $CD4^+$ T cells from the site of inflammation when compared to cells from other organs. These results were further underlined by functional data applying siRNA, where HDAC 5, 7 and 9 could be identified as critical for T cell activation. In summary, (un)specific HDAC inhibition exerts an anti-inflammatory potency, modulates T cell differentiation and thus represents a novel therapeutic target for chronic (intestinal) inflammation.

OR.62. Abnormal Antigen Presenting Cells (APCs) Alter T Cell Effector and/or Suppressive Function Leading to Colitis

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Aims**:** Deficiency in Wiskott-Aldrich Syndrome protein (WASP), a cytoskeletal regulator, leads to spontaneous colitis in mice and a subset of patients. Transfer of WT CD4+ cell into WASP/RAG double knockout mice (WTDKO mice), but not RAG KO mice, leads to severe colitis. Thus, WASP-deficient (WKO) antigen presenting cells (APCs) can induce WT T cells to become pathogenic. We aimed to determine whether disease could be prevented by increasing the regulatory T cell component of the donor CD4+ cells transferred in WTDKO mice, define the different APC subsets in various lymphoid organs in WKO mice, and identify the specific immune cell subpopulation that may lead to defective effector (Teff) and/or regulatory T cell (Treg) function. Methods and Results: WASP/RAG DKO mice that received Tregs alone demonstrated no/little disease, those that received either Teffs alone or Treg:Teff in a ratio of 1:1 developed moderate to severe disease (colitis score: 8 and 6.5 out of 8, respectively), and those that received Treg:Teff in a ratio of 2:1 had protection from colitis (colitis score: 3.2). Disease attenuation was associated with increased percentages of Tregs in all lymphoid organs. Flow cytometry of cells isolated from WKO mice demonstrated: a) increased presence of CD11bhiCD11cint population in colonic LP, b) decreased presence of CD11c+CD103+CD11b+ cells in the mLNs, and c) increased CD11c+CD11b- cells in LP, spleen, and mLNs, compared to WT. *In vitro* suppression assay, however, demonstrated WT Tregs function equally in the presence of WKO splenocytes compared to WT. Conclusions**:** WKO APCs fail to control the activation of Teffs in the LP likely by changing the balance of Teff:Treg either in number and/or function. Abnormal APCs may similarly contribute to the development of human inflammatory bowel disease.

OR.63. Granulocyte-macrophage Colony-stimulating Factor Elicits Bone Marrow-derived Cells that Promote Efficient Colonic Mucosal Healing

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Granulocyte-macrophage colony-stimulating factor (GM-CSF) therapy is effective in treating some Crohn's disease (CD) patients and protective in the mouse model of dextran sulfate sodium (DSS)-induced colitis. This study explores the possibility that intestinal mucosal repair is a main component of the beneficial effects of GM-CSF therapy. DSS colitic mice were treated with GM-CSF or saline, and clinical, histological and inflammatory parameters were evaluated. GM-CSF therapy reduced disease activity index, mitigated the release of inflammatory mediators and markedly improved mucosal repair, as highlighted by lower ulceration score in colonoscopy examinations. GM-CSF-mediated improvement of mucosal repair was accompanied by accelerated hyperproliferative response of epithelial cells in ulcer-adjacent crypts and increased mRNA expression of factors implicated in mucosal repair. GM-CSF therapy still improved colitis when hematopoietic cells exclusively were responsive to GM-CSF, as shown in lethally irradiated GM-CSF receptor-deficient mice reconstituted with wild-type bone marrow cells. Lastly, we observed that GM-CSF-induced promotion of mucosal repair is temporally associated with increased accumulation of CD11b+Gr1lo myeloid cells in colon tissues. Our study shows that GM-CSF-dependent stimulation of bone marrow-derived myeloid cells during DSS-induced colitis accelerates colon tissue repair and suggests that this property might contribute to symptomatic benefits of GM-CSF therapy observed in CD patients.

OR.64. Intestinal Microbiota Directs Infiltrate Composition and Disease Severity in a Novel Enterocolitis Zebrafish Model

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The pathogenesis of inflammatory bowel disease involves dysfunctional mucosal immune responses to commensal bacteria

in genetically predisposed hosts. The complicated interactions between innate cells and bacteria however, have left researchers with exceeding difficulties to assess the relative contribution by each of these cells to the intestinal pathology. We altered the intestinal flora of zebrafish with antibiotics and assessed the effects on the mucosal infiltrate and severity of intestinal inflammation in a novel oxazolone-induced enterocolitis model. Intestinal inflammation was evaluated by histology, flow cytometry and cytokine profiling. Zebrafish oxazolone enterocolitis is microbiota-dependent and characterized by an influx of granulocytes, epithelial damage, Goblet-cell depletion and increased expression of pro-inflammatory cytokines. Vancomycin treatment diminished the intestinal microbial load and resulted in a bacterial composition dominated by Fusobacteria. Vancomycin conferred strong protection from enterocolitis associated with reduced percentages of infiltrating neutrophils. In contrast to vancomycin, administration of colistin sulphate resulted in a predominance of γ-proteobacteria in the intestine that associated with reduced infiltration of eosinophils and lymphocytes and no significant reduction in enterocolitis. We conclude that components of the intestinal microbiota drive zebrafish oxazolone enterocolitis and directly affect the severity of disease and composition of the intestinal infiltrate.