controls (CTRL). Caco-2 cells were used as a model for CECs. All experiments were performed at least 3 times.
Results: Immunohistochemistry of biopsies from CTRL showed both nuclear and granular cytosolic OPG in CECs. However, IBD had only dense granular cytosolic OPG, not nuclear, suggesting that OPG is mobilized from the nucleus during inflammation in CECs. Immunofluorescence (IF) of Caco-2 cells revealed nuclear and granular cytosolic OPG, similar to primary CECs. The presence of nuclear OPG was confirmed by immunoprecipitation of nuclear extracts of Caco-2 cells followed by mass spectrometry. IL-1β (but not IL-18) significantly increased OPG mRNA and OPG secretion. IL-1β significantly attenuated nuclear OPG signal by IF. Secretion of OPG was polarized towards the apical side, regardless of whether IL-1β was applied apically or basolaterally. Fluorochrome-tagged OPG specifically attached to Escherichia coli and changed the flow cytometry side scatter of the bacterial population, suggesting that OPG aggregates bacteria.
Conclusions: During inflammation, nuclear OPG in CECs may be extruded from the nucleus into the cytoplasm and then towards the colonic lumen, where OPG may bind to bacteria. Therefore, OPG may be a novel innate colonic defense system triggered by inflammation.

P-010
Interleukin-10 inhibits human IFNγ-secreting effector T cells indirectly by controlling antigen-presenting cell function

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Introduction: Inflammatory bowel disease arises from abnormal T cell reactivity to commensal microbiota. Interleukin-10 (IL-10) plays a crucial role in suppressing microbiota-specific T cell responses. However, its mode of action remains unclear. We have recently identified a loss-of-function mutation in the IL-10 receptor alpha (*IL-10RA*) gene of an infantile-onset IBD patient. Before diagnosis of the genetic defect, remission of disease could be achieved with immune-suppressants allowing in-depth analysis of the mechanism by which IL-10 controls T cells.
Aim: Identify the mechanisms by which IL-10 controls inflammatory T cell responses in humans.
Materials and Methods: The phenotype of the inflammatory T cells in the absence of a functional *IL-10RA* was identified in biopsies by immunohistochemical analysis. *In vitro* assays were used to identify the mechanism by which IL-10 controls T cell responses.
Results: Lesional intestinal tissue taken at onset of disease contained high numbers of T cells displaying Th1 and Th17 characteristics. In agreement, IL-10 failed to control IFNγ and IL-17 production by activated *IL-10RA*-deficient T cells *in vitro*. By coculturing T cells and antigen-presenting cells (APC) from the *IL-10RA*-deficient patient and a healthy control, we revealed that IL-10R expression on APC, and not on T cells, is important for controlling IFNγ production by effector T cells.
Conclusion: Taken together, our data demonstrate that IL-10 inhibits effector T cell responses indirectly by controlling APC function. Our findings shed new light on the importance of APC in disease pathogenesis and provide a rationale for developing APC targeted therapies.

P-011
Differential induction of T cell tolerance in the small and large intestine

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Introduction: Intestinal antigen-presenting cells (APC) continuously sample harmless antigens from the lumen and migrate to draining lymphoid tissues to induce tolerance. Crohn's disease arises from a failure to maintain tolerance to these harmless luminal antigens. The location of inflammation varies amongst patients suggesting that the small and large intestine have different susceptibility to inflammation.
Aim: Investigate whether tolerance in the small and large intestine is imposed by distinct local regulatory mechanisms.
Materials and Methods: Mice were administered with soluble protein antigen by either the oral or rectal route. The phenotype of APC subsets and Foxp3+ regulatory T cell frequency was assessed by flow cytometry.
Results: Administration of a soluble antigen to the colon led to APC-mediated antigen presentation in the iliac lymph nodes, while orally applied antigen is exclusively presented in the mesenteric lymph nodes. After feeding antigen, CD103+CD11b+ APC actively migrated from the small intestine to mesenteric lymph nodes, whereas colonic antigen application increased the CD103+CD11b− APC population in the iliac lymph nodes. Despite the difference in draining site, both small intestinal and colonic antigen administration induced tolerance via the induction of Foxp3+ Treg cells.
Conclusion: Taken together, our data reveals that within different locations of the gastrointestinal tract tolerance is imposed by local regulatory mechanisms that are adapted to microenvironmental differences. A better understanding of the mechanism behind tolerance induction in the small and large intestine will provide crucial insights into diversity and pathogenesis of Crohn's disease.

P-012
Probiotic bacteria enhance antigen sampling and processing by dendritic cells in pediatric IBD

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Introduction and Aim: Bifidobacteria have been reported to reduce inflammation and contribute to intestinal homeostasis. However, the interaction between these bacteria and the gut immune system remains largely unknown. Because of the central role played by dendritic cells (DC) in immune responses, we examined in vitro the effects of a Bifidobacteria mixture (probiotic), on DC functionality from children with inflammatory bowel disease (IBD).
Methods: DCs obtained from peripheral blood monocytes of patients with Crohn's disease (CD; n = 12), Ulcerative Colitis (UC; n = 7), and healthy controls (HC; n = 6) were incubated with fluorochrome-conjugated particles of *Escherichia coli* or DQ-Ovalbumin (DQ-OVA®) after 24 hrs pre-treatment with the probiotic, in order to evaluate DC phenotype, antigen sampling and processing. Moreover DC culture supernatants were collected to measure TNF-a secretion by ELISAs.
Results: DCs generated from CD children showed significant higher bacteria particles uptake and DQ-OVA processing after incubation with the probiotic ($p = 0.01$ and $p = 0.01$, respectively); in contrast, DC from UC and HC showed no significant changes. Moreover, a significant TNF-a production was observed in DC from CD after exposure to *E. coli* particles ($p = 0.01$), whilst the probiotic didn't increase this pro-inflammatory cytokine ($p = 0.03$).

Conclusions: In Crohn's disease children bifidobacteria could improve the altered capacity to capture and process luminal antigens, contrasting the uncontrolled microorganisms growth and reducing inflammation in the gut.

P-013
Colitis in the graft of children treated with intestinal transplantation is associated with NOD2 mutations

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Aim: To determine the incidence of severe chronic colitis in the first 100 pediatric intestinal transplant recipients at NEM, Paris; to assess histology of the affected colons and NOD2 genotype of recipients. The study group consisted of 57 patients who received a combined small intestinal/colon transplant. Nine (15.7%) patients developed severe chronic colitis with IBD phenotype. This group presented no distinct characteristics. Their mean age at transplantation was 5.4 yrs (range: 2.2–8.5), 3 patients had a liver transplant, familial history of IBD or allergies was never mentioned. Anti-rejection strategy was similar in all patients (corticoids/tacrolimus). Colitis symptoms appeared after 4.8 yrs (range: 1–12). All presented signs of inflammation (elevated CRP), in 7/9 patients an infectious agent was identified at presentation. Endoscopy showed ulcers of variable severity. Histological findings revealed erosions or ulcerations (4/9 cases) associated with a polymorphic inflammation with an increased number of eosinophils (6/9 cases) and with crypts dedifferentiation or injury (5/9 cases). The histological findings were not pathognomonic. NOD2 genotype was obtained from 6/9 index cases and 9 controls. Three colitis patients had homozygous mutations whereas none of the controls had mutations in the NOD2 gene. Chi square analysis was significant at $p = 0.017$.
In summary, infections occur frequently after intestinal transplantation but some cases progress to severe chronic colitis with non-specific histological inflammatory infiltrates. This first report demonstrates an association with NOD2. Mutations in the NOD2 gene may put children with a colon transplant who are treated with immune suppression at risk for inflammation of the colonic graft.

P-014
Increased expression of IL-21 and co-localization with IFN gamma in inflammatory lesions of pediatric Crohn's disease

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Introduction: IL-21 is produced by activated CD4+ T helper (Th) cells and can enhance IFNγ and IL-17 production by Th1 and Th17 cells respectively. IL-21 production is increased in the mucosa of Crohn's Disease (CD) patients but location and phenotype of the IL-21-producing cells is unclear.
Aim: To investigate location and phenotype of the IL-21-producing-cells in pediatric CD.
Methods: IL-21, IFNγ and IL-17 production was assessed by immunohistochemistry on small intestinal and colonic sections. Th cells isolated from PBMCs of healthy donors were stimulated in the presence of an IL-21 blocking antibody. IFNγ production was assessed by ELISA.
Results: In all patients, increased numbers of IL-21-secreting cells were observed in inflamed compared to non-inflamed regions of the intestine. In 4 out of 5 patients, these IL-21-producing cells co-localized with increased numbers of IFNγ- but not IL-17-secreting cells whereas only 1 patient presented with IL-17-secreting cells co-localizing with IL-21-producing cells. Neutralization of IL-21 activity decreased IFNγ production by activated Th cells.
Conclusion: Increased numbers of IL-21-secreting cells are present in inflamed tissue of pediatric CD and are more frequently associated with IFNγ-producing cells than IL-17-producing cells. Neutralization of IL-21 function decreases IFNγ secretion by activated Th cells. Altogether our data show that in pediatric CD, IL-21 secretion is associated with inflammation and could promote the function of Th1 rather than Th17 cells.

P-015
Human buccal epithelium acquires microbial hyporesponsiveness at birth, a role for secretory leukocyte protease inhibitor

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Introduction: Repetitive interaction with microbial stimuli renders epithelial cells (EC) hyporesponsive to microbial stimulation. We reported that buccal EC from a subset of pediatric Crohn's disease patients are not hyporesponsive and release chemokines. To explain this phenomenon, we determined how healthy primary buccal EC become hyporesponsive to microbial stimuli after birth.
Aim: To identify kinetics and mechanisms of acquisition of hyporesponsiveness to microbial stimulation using neonatal primary human buccal EC.
Methods: Buccal EC collected directly after birth and in later stages of life were investigated. Chemokine release and regulatory pathways were studied using primary buccal EC and the buccal EC line TR146. Findings were extended to the intestinal mucosa using murine systems.
Results: Directly after birth buccal EC spontaneously produced the chemokine CXCL-8 and were responsive to microbial stimuli. Within three weeks these EC attained hyporesponsiveness, associated with inactivation of the NF-κB pathway and upregulation of the endogenous NF-κB inhibitor secretory leukocyte protease inhibitor (SLPI). Knockdown of SLPI in TR146 EC inhibited hyporesponsiveness inducing increased NF-κB activation and subsequent chemokine release. This regulatory mechanism extended to the intestine, as intestinal EC from SLPI deficient mice expressed of the murine CXCL-8 homolog CXCL-2 compared to WT mice.
Conclusion: SLPI determines acquisition of microbial hyporesponsiveness by buccal- and intestinal-epithelium in the first weeks of life.

P-016
Impact of *Campylobacter concisus* on paediatric inflammatory bowel disease

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Introduction: *Campylobacter concisus* has been identified as an organism that may contribute to IBD pathogenesis. Previous investigations have shown *C. concisus* is detected in stool more frequently in children with newly diagnosed CD[AD1] compared