**Gut flora specific immune responses in multiple sclerosis**

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**Objective**

The ability of intestinal microbiota to sustain an inappropriate immune reaction, such as in autoimmunity, at distant sites has been recently shown in animal models of disease. Similarly, the possibility to generate protective immune responses through the inoculation of selected commensal bacteria which induce regulatory cells has also been shown. We have found that a distinct population of cells with antibacterial activity is expanded in individuals with Multiple Sclerosis (MS). These cells, named MAIT (mucosal-associated invariant T cells) lymphocytes, preferentially home to the intestine, but they can penetrate in the brain where they can be loaded with IL17, a pathogenic cytokine. Here we studied the composition of the gut microbiota in MS patients and in homozygotic twin pairs discordant for disease; we then studied the response of MAIT cells to relevant bacterial or yeast strains, that were isolated from faecal samples from MS patients.

**Methods**

PBMCs were isolated from healthy donors and patients affected by the Relapsing-Remitting form of MS (RR-MS). Multicolor flow cytometry was used to study in detail the phenotype and the function of human MAIT cells. Fecal samples from MS patients and homozygotic twins discordant for disease were collected and analyzed for microbiota diversity. Sorted MAIT cells were challenged ex vivo with autologous antigen presenting cells (APCs) and toll-like receptor (TLR) ligands or selected bacterial strains derived from fecal samples analyzed, and cell activation and cytokine production were measured. In some experiment, the proliferative response of MAIT cells were measured following staining with CFSE after stimulation with selected bacterial strains.

**Results**

We find that the frequency of MAIT cells are significantly increase in the peripheral blood of sporadic MS patients and affected twins; these cells are equipped with the array of molecules necessary for migration in the CNS. Moreover, MAIT cells produce pro-inflammatory cytokine and proliferate when exposed to bacterial extracts, in co-culture with APCs, derived from fecal samples of affected twins and sporadic MS patients and healthy donors. We also assessed fecal samples by restriction analysis that showed that in MS patients the gut microbiota profiles are different compared to healthy volunteers.

**Conclusions**

These results are in agreement with the hypothesis that dysbiosis of the gut microbiota may determine a dysfunction of mucosal responses and consequently may favor the development of systemic inflammatory and autoimmune diseases.