Microbiology

P708
What causes recurrence of Crohn’s Disease after intestinal resection? A prospective evaluation of microbiota, smoking and anti-TNF therapy. Results from the POCER study

E. Wright1, M. Kamm1,2, J. Wagner3, S.M. Teo4, P. De Cruz5, A. Hamilton1, K. Ritchie1, M. Inouye6, C. Kirkwood1
1St Vincent’s Hospital & University of Melbourne, Gastroenterology, Melbourne, Australia, 2Imperial College London, Medicine, London, United Kingdom, 3Murdoch Children’s Research Institute, Enteric Virus Group, Melbourne, Australia, 4The University of Melbourne, Pathology, Melbourne, Australia

Background: The intestinal microbiota is the antigenic drive in Crohn’s. It is also likely to be responsible for disease recurrence after intestinal resection. We aimed to identify microbiota predictive of, or associated with, disease recurrence, remission, smoking and adalimumab therapy.

Methods: 141 mucosal samples from 34 Crohn’s disease patients were obtained at surgical resection (baseline) and from the ileum and anastomosis at colonoscopy 6 and/or 18 months post-operatively. 28 control samples were obtained; 12 colonic samples from healthy patients with a normal colon (healthy controls) and 16 ileal and anastomosis samples from 8 patients who had previously undergone right hemicolectomy for colon cancer (surgical controls). Endoscopic recurrence in Crohn’s patients was assessed using the Rutgeerts score. Mucosal 16s ribosomal profiling was performed using the MiSeq Illumina platform.

Results: Crohn’s disease was associated with reduced bacterial diversity when compared to healthy controls but not surgical controls (Shannon Diversity Index; t-test: p=0.012 and p=0.552 respectively). Bacterial composition (beta diversity) differed significantly between Crohn’s disease and both healthy (p=0.024) and surgical (p=0.038) controls, and changed within Crohn’s patients over time, but did not differ significantly between those with and without endoscopic recurrence. However significant taxonomic differences between recurrence and remission included increased Proteus (p=0.019) and decreased genera from the Firmicutes phylum including Faecalibacterium (p=0.004).

No significant differences were observed in alpha or beta diversity between smokers vs. non-smokers and between adalimumab treatment vs. no adalimumab treatment. Smoking was associated with significantly elevated levels of Proteus (p=0.013) and lower levels of Phascolarctobacterium (p=0.028) and Faecalibacterium (p=0.029). Low abundance of Faecalibacterium and smoking were both independently associated with recurrence (OR 5.5 (CI 1.8-17) p=0.002 and OR 3.3 (CI 1-11) p=0.049 respectively).

Conclusions: Crohn’s disease is associated with a microbial signature distinct from health. Surgical resection alone may be responsible for some, but not all, of the taxonomic differences observed in patients following intestinal resection in Crohn’s disease. Microbial factors, such as the presence of Faecalibacterium, and smoking may influence post-operative Crohn’s disease recurrence through independent mechanisms. The mechanism by which anti-TNF therapy prevents recurrence post-operatively does not appear to have a microbial basis.

P709
Association between genotypes of Bacteroidetes in fecal samples from patients with Crohn’s disease and its assessment using an experimental mouse model

R. Suarez1, A.P. De Felipe1, L. Urissarri2, R. Ferreiro1, A. Lorenzo1, J.E. Dominguez-Munoz2, J.M. Leiro1, M. Barreiro-de Acosta1,2
1University of Santiago de Compostela, Department of Microbiology and Parasitology, Institute of Food Research and Analysis, Santiago, Spain, 2University Hospital Santiago de Compostela, Gastroenterology, Santiago, Spain

Background: The pathogenesis of inflammatory bowel disease (IBD) involves an imbalance of the gut microbiota generating an inappropriate activation of the mucosal immune system in genetically predisposed individuals. The human commensal microbiota contains a large number of Bacteroidetes species that may cause inflammation in animal models. The aim of this study was to detect and evaluate the influence of different Bacteroidetes genotypes on the activity of Crohn disease (CD) patients. In addition, the effect of these bacteria isolated from CD patients on gut inflammation was evaluated in mice.

Methods: We performed a case control study on the intestinal bacteria of the phylum Bacteroidetes from faeces of CD and healthy controls (HC) using a polymerase chain reaction (PCR) designed to detect human-specific genetic markers targeting Bacteroidetes-like 16S rRNA genes in fecal DNA samples. The PCR products from the 16S rRNA genes were digested with HinfI, PciI, DpnII and AcI enzymes and restriction fragment length polymorphism (RFLP) were determined. RFLP and sequencing analysis indicated that a total of 6 bacterial genotypes do exist: N1, C1, C2, C3, C4 and C5 (of which N1 genotype is probably a strain of Bacteroides dorei and C1, and maybe C2, strains of B. vulgatus). The relationship between CD activity (CDAI>150) and microbiota was evaluated. Microbiota from CD patients were transplanted into mice gut to evaluate their ability to induce inflammation. Results are shown in percentages.

Results: 11 CD patients (8 with active CD -aCD- (CDAI>150), and 3 with inactive CD -iCD-), and 11 HC were included. The predominant Bacteroidetes genotype in feces from HC and iCD was N1 (present in 100% of samples), whereas this genotype was found in only 28% of patients with aCD. 18% aCD patients showed the C1 genotype, 9% the C1 and C3 genotypes together, 18% the C4 genotype, and 27% the C1 and C4 genotypes together. The transplant of bacteria from CD patients to mice led to large bowel inflammation, and the stool of the transplanted mice consisted in 30% C4 genotype and had a high level of Bacteroidetes cluster in comparison with the mice transplanted with bacteria from HC.

Conclusions: The fecal microbiota of CD patients is different from those of HC in that they present a wide variety of Bacteroidetes cluster genotypes. The C4 genotype by itself, or together with the C1