ASSESSMENT OF MICROBIOTA, INFLAMMATORY MARKERS, NUTRITIONAL AND ENDOCRINOLOGICAL STATUS IN IBD PATIENTS

MINUTE FOR IBD

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Background

Inflammatory bowel disease (IBD) is a chronic gastrointestinal disorder increasing in incidence and severely influencing affected individuals. Although the factors associated with the disease risk include immune status, gut microbiome and several environmental causes including food intake, the current scientific knowledge of specific triggers and diagnostic markers to improve interventional approaches is still scarce (1).

Additional basic and clinical research involving specific groups of patients and obtaining multilevel data are essential for the management of the disease. Investigation of mutual correlation between gut microbiota, inflammatory, endocrine and nutritional status provides integrative framework for better understanding the IBD pathophysiology.

Project scope&objectives

MINUTE for IBD project's scope is to perform the best possible systematic analysis of relevant elements and factors (gut microbiota composition, inflammatory/endocrine markers, nutritional status) contributing to the development and progress of IBD on newly diagnosed, therapy naïve IBD patients and individuals not fulfilling IBD diagnosis criteria as a control. Through an integrated translational approach, two skilled groups of experts (academic and clinical) are focused on accomplishing the main scope of the project. This will be carried out through the four main objectives.

Project objectives:

1. Analysis of microbiota composition (by 16S rRNA gene sequencing) in stools and in mucosa of 40 IBD (20 UC and 20 CD) patients and 20 non-IBD subjects
2. Quantification of relevant immune and endocrine biomarkers in blood samples
3. Determination of overall nutritional status in study participants
4. Integration of the obtained results and setting-up better guiding principles when deciding on the therapy and nutritional intervention in IBD patients

The obtained results will increase the scientific knowledge and evidence necessary to support better stratification of the patients and consequently improve the disease diagnosis, treatment and prognosis. The importance of answering such research questions is timely and of high clinical relevance for the whole research field and especially for the benefit of the patients.

Methods

In the project gut microbiota composition, blood inflammatory markers and hormones are assessed and correlated with patients' nutritional status. After enrolment subjects are completing specially designed questionnaires regarding quality of life and dietary habits. Their blood and stool samples, in addition to intestine tissue biopsies are collected and analysed for biomarkers and gut microbiota present (Figure 1). It is expected that upon complete statistical and bioinformatical data analyses, correlation of investigated factors with the disease status will be obtained for the studied group of patients. This pilot-clinical research study dealing with the cutting-edge topics and using state-of–the art sensitive methodologies, including next generation sequencing of 16S rRNA gene amplicons and proximity extension assay (Figure 2), may provide insight in the interrelationship of investigated parameters and contribute to improved patient stratification and introduce better guiding principles when deciding on the therapy and nutritional intervention in IBD patients.

Current status & results sneak peek

The current status of participant recruitment is given in Table 1. One of the main objectives of the study is to investigate the contribution of the faecal and intestinal microbiota to the disease specific phenotype. Mucosal samples taken along the colon were compared to the faecal bacterial content and the preliminary results (Figure 3) suggest there is more *Proteobacteria* in the mucosa of IBD patient when compared to the IBS control (1). Also observed is the higher abundance of *Proteobacteria* in samples collected from terminal parts of the colon for both patients.

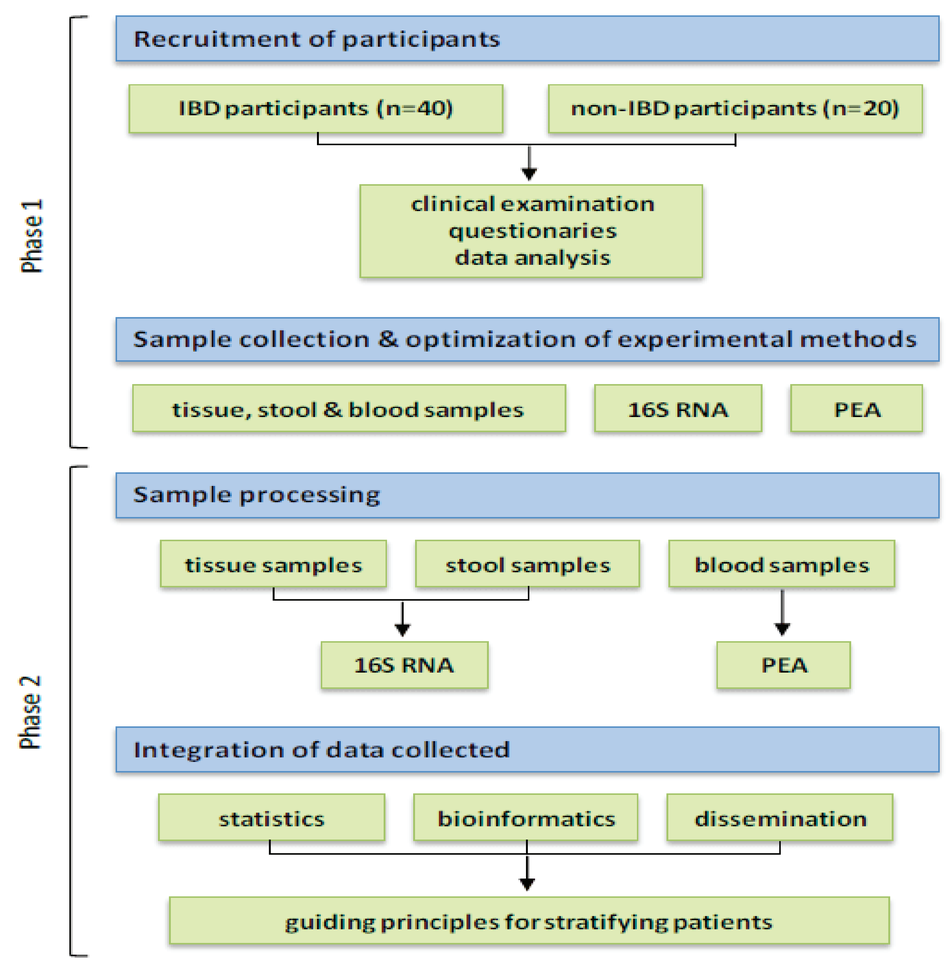

Fig. 1. Schematic representation of the MINUTE for IBD project's course

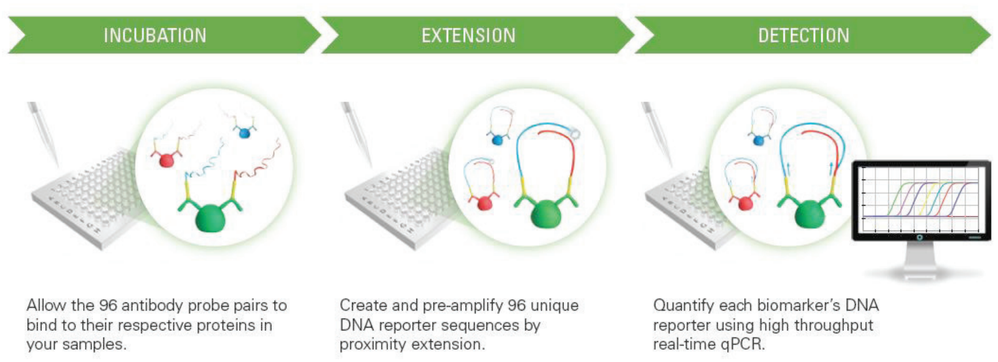

Fig 2. Principle utilised in the development of the recently adopted Proximity Extension Assay method which allows assessment of the small amount of relevant biomarkers in minimal sample volumes of different biological fluids without prior separations.

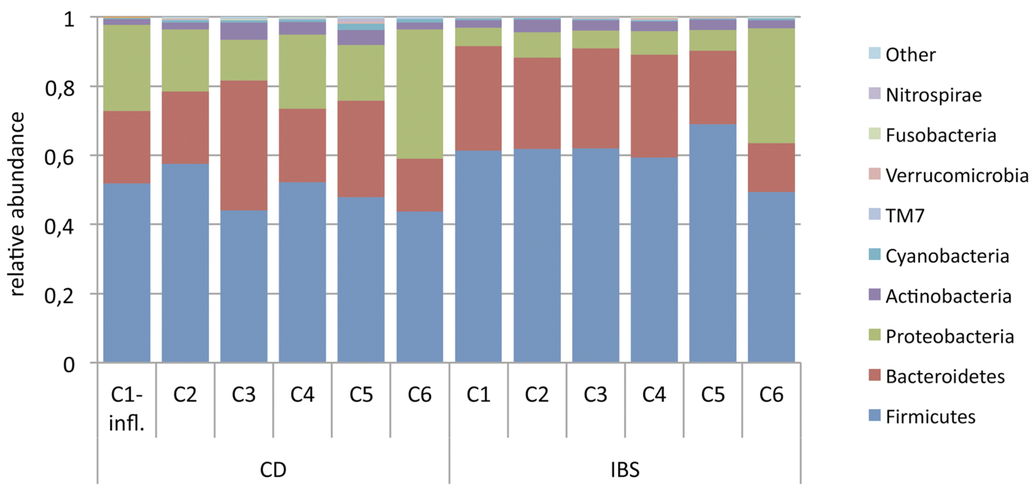

Fig 3. The relative abundance of major bacterial phyla detected by sequencing of 16S rRNA gene amplicons. Samples of colon mucosa were taken during endoscopy from different sites along the colon (from C1-terminal ileum to C6-rectum). Samples were collected from two subjects – one diagnosed with Crohn's disease (CD) and one with IBS.

Table 1. Current status of participant recruitment.

		CD	UC	IBS
n		**7**	**9**	**35**
Sex	M/F	4/3	7/2	13/22
Age (years)	range	22-56	18-34	20-44
	median	43,5	29,5	29,5

References

[1] Matijasic M et al. Modulating Composition and Metabolic Activity of the Gut Microbiota in IBD Patients. Int J Mol Sci. 2016 Apr 19;17(4). pii: E578. doi: 10.3390/ijms17040578. [2] Matsuoka K and Kanai T. The gut microbiota and inflammatory bowel disease. Semin Immunopathol. 2015 Jan;37(1):47-55. doi: 10.1007/s00281-014-0454-4.

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