Genome-wide methylation profiling of Crohn’s Disease in women

A.Y.F. Li Yim1,2, N.W. Duijvīs3, J. Zhao1, W.J. de Jonge1, A.N. Mul1, M.M.A.M. Mannens1, A.A. te Velde1 and P. Henneman2*

1Department of Clinical Genetics, Academic Medical Center Amsterdam, The Netherlands; Teytig Institute for Liver and Intestinal Research, Academic Medical Center Amsterdam, The Netherlands; *Department of Genetic Epidemiology and Biostatistics, King’s College, London, United Kingdom

*Correspondence: p.henneman@amc.uva.nl

Introduction

Crohn’s disease (CD) is a complex chronic disorder, which is characterized by the inflammation of distinct parts of the gastrointestinal tract. Symptoms of CD include diarrhea, fever, abdominal pain, weight loss and anemia, resulting in a detrimental effect on the overall quality of life. Numerous genome-wide association studies (GWAS) have identified CD associated loci which explain only 20 to 25% of the heritability. A growing body of literature suggests that additional factors such as diet, the gut-microbiome and the epigenome play an important part in the development and progression of CD. Here we sought to assess how the DNA methylome of women is affected by the presence or absence of CD and compare our results with previous GWAS and epigenome-wide association studies (EWAS).

Methods and workflow

We drew peripheral blood from 15 women with histologically confirmed ileal or ileocolic CD and 28 healthy women (mean age and standard deviation: 30.5 ± 6.5 years). DNA from the blood was isolated after bisulfite conversion after which the DNA methylome was probed using the illumina HumanMethylation 450k BeadChip Array (“450k”).

Analysis of the data was done in the R statistical programming environment using the minfi Bioconductor package. Hereby we performed an initial quality control using the methylAid package which resulted in the removal of three outliers samples. The data was then normalized using the functional normalization method after which batch effects were estimated and corrected for using ruvfit, yielding a list of differentially methylated positions (DMPs). While we do not specifically correct for cell blood distribution, ruvfit corrects for hidden biological batch effects such as blood cell distribution, ruvfit corrects for hidden biological batch effects such as blood cell distribution, which reinforces the volatile nature of the HLA genes as reported previously.

Using this criterion, we found eight DMRs. Interestingly, we found a DMR upstream of HLA-J (Fig. 3a). This region encompasses the very polymorphic HLA-J locus. The nearby locus of HLA-J (Fig. 3a) which reinforces the volatile nature of the HLA genes as reported previously.

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Differentially methylated regions

Using the bumphunter function, we searched for at least four consecutive DMPs. Using this criterion, we found eight DMRs. Interestingly, we found a DMR upstream of HLA-J (Fig. 3a), which reinforces the volatile nature of the HLA genes as reported previously.

Discussion

Here we used the HumanMethylation 450k BeadChip array to assess the peripheral blood methylome of female patients with CD versus healthy controls. While we find DMPs and DMRs in CD patients versus controls, we see that the effect size is limited. Nevertheless, GO enrichment analysis indicates that the reported DMPs are significantly enriched in genes involved in pathways related to inflammation and wounding as well as bacterial response, suggesting that CD manifests itself, albeit limited, in the methylome of peripheral blood. Important to note is that the enriched pathways are not specific to CD and are generally implicated in inflammatory diseases. To this end, we reiterate the conclusions of Harris et al. (2012) where we find minimal changes within the methylome in peripheral blood that are specific to CD, but that these changes might be associated to the inflammatory nature of CD.

References