Genes in IBD-Associated Risk Loci Demonstrate Genotype-, Tissue-, and Inflammation-Specific Patterns of Expression in Terminal Ileum and Colon Mucosal Tissue

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Disclosures

• No relevant disclosures
Background and Challenges

163 Loci

→ Need to translate these genetic findings to biology, gene function, and therapeutic targets

→ Need to understand where genes are expressed and how the genes function in the gut

→ Genetic variants in non-coding regions affect gene expression of near-by genes (e-QTLs)

→ SNPs associated with complex traits often exert a tissue-dependent effect on gene expression

→ Up to 25% of the IBD-risk loci genes were not captured in traditional microarray studies
Aim/Hypothesis

Studying the expression of genes encoded in IBD-associated risk loci by differential expression and eQTL analysis in disease-relevant tissues from IBD patients will refine candidate genes underlying the genetic associations.
Multi-center approach to address problem of sample size

593 Patients
1116 Samples (989 passed QC)

580 CD
315 UC
35 Healthy

545 Colon
385 TI

357 Inflamed; 573 Un-inflamed

178 Paired samples
-34 CD Colon
-83 CD TI
-61 UC Colon
Strategy

Genotype: ImmunoChip
RNA expression profiling: Nanostring

- Location specific Expression patterns
- Variance analysis
- Clustering analysis
- Disease and Inflammation Specific expression patterns
- Single nucleotide polymorphisms
- Expression quantitative trait loci

Do IBD risk genes enable robust discrimination between healthy and patient populations?

Can we identify novel interactions between IBD disease genes?

Are there shared mechanisms of inflammation between UC and CD?

Which key genes are regulated by genetic variation in patient population?
NanoString: Custom probeset designed to identify expression-affecting SNPs

CDK12, NEUROD2, PPP1R1B, STARD3, TCAP, PNMT, PGAP3, ERBB2, MIEN1, GRB7, IKZF3, ZPB2, GSDMB, ORMDL3, LOC728129, GSDMA, PSMD3

467 of the 1441 genes encoded in the risk loci represented on the codeset
Genes from IBD-risk loci exhibit specific geographic expression in TI and colon in healthy controls
Expression of genes from IBD-risk loci change with inflammation

Patients with UC segregate from healthy controls even in un-inflamed samples
Genes with lowest variance in expression in healthy controls show the highest variance in patients
Correlation analysis identifies a core network of co-regulated genes and predicts novel interactions

STRING analysis confirms 35 of 41 genes formed a single network of first-degree interactions

Markedly enriched for shared pathways by GSEA
Gene pairs with predicted functional interactions: SP140 and PTPRC

- SP140 – Recognizes and binds histone acetylation and methylation – may have role in immune tolerance
- PTPRC (CD45) – B and T cell receptor signaling – persistence of B cells in germinal centers; increases Treg-dendritic cell conjugation

Cis-expression quantitative trait loci demonstrate patterns of genotype specific expression

Identified 68 significant associations between 22 unique SNPs and 30 distinct genes

Twice as many eQTLs identified in uninflamed tissues compared to inflamed (43 v. 25)
**fc-eQTL: Genotype-specific gene expression variation in inflammatory context**

eQTL mapping in multiple immune subsets under various stimuli have shown SNPs have condition-specific effects on gene expression.

Risk allele count

fc-eQTL demonstrate genotype-specific variation in gene expression induced by inflammation

Cis-fc-eQTL found 43 significant associations between 32 unique SNPs and 40 distinct genes

Trans-fc-eQTL analysis found 30 significant associations
Conclusions - 1

• Expression of genes encoded in IBD-associated risk loci demonstrate:
  – specificity for tissue, disease, and inflammation status
  – low variance of expression across healthy controls

• Genes with lowest variance in expression in healthy controls have the greatest variance across patients

• Co-expression analysis connects 41 genes and suggests novel interactors (e.g. SP140 and PTPRC)
Conclusions - 2

- Identified 68 unique eQTLs with patterns of tissue, disease, and inflammation specificity
  - Replicated 24 of 36 prior eSNP associations
- `fc`-eQTL analysis demonstrates how common genetic variants change magnitudes of gene expression changes in inflammation
- Orthogonal approaches collectively prioritize candidate genes in 98 (60%) of the 163 IBD-associated risk loci
- Reinforces value of studying genes in tissue and disease context

Hulur I, et al. BMC Genomics. 2015
Kabakchiev B and Silverberg MS. Gastroenterology. 2013
Singh T, et al. Inflamm Bowel Dis. 2015
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