Re-analysis of DNA methylation data from tuberculosis-infected individuals with a focus on target genes

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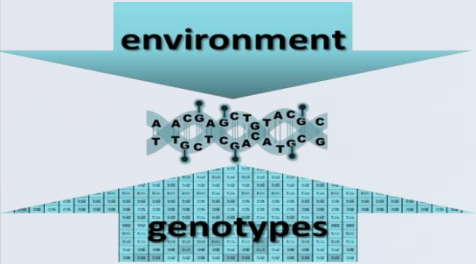
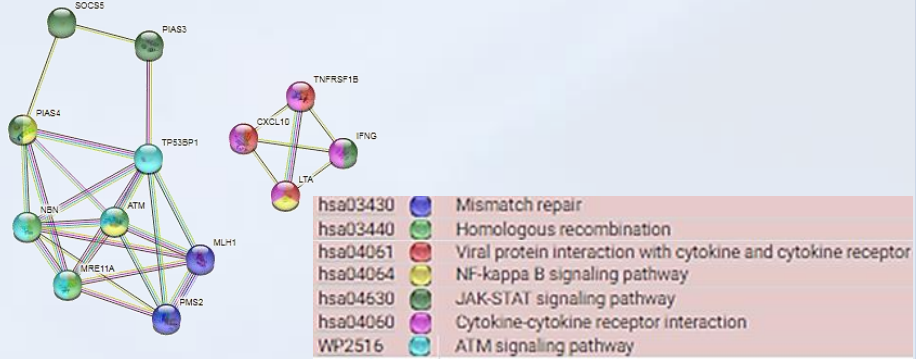


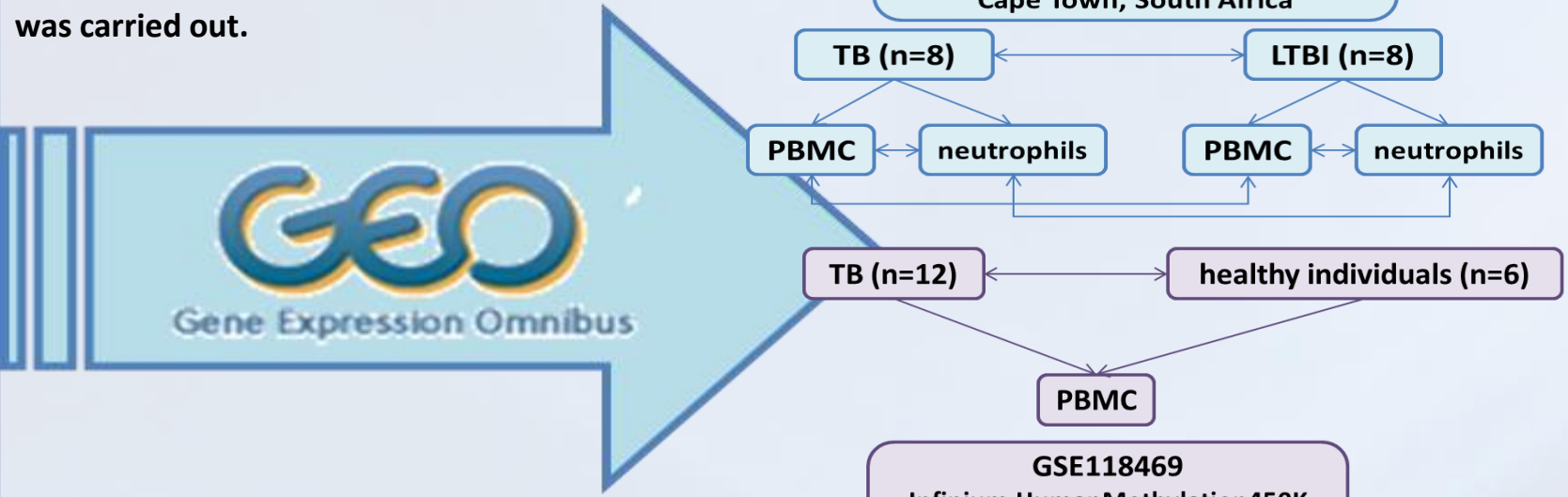
Along with the structural variability of the genome, epigenetic modifications can be significant for susceptibility to various, including infectious diseases. The aim of this study was to re-analyze the differential methylation of genes (DMG), for which associations with tuberculosis and its pathogenetically significant features were previously established, as well as for those of interest from the point of view of studying the phenomenon of reverse comorbidity of asthma and tuberculosis.

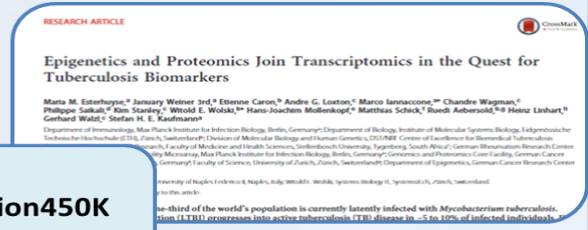
We were primarily interested in genes that, according to our previously data, were associated with infectious diseases, in particular thirteen genes *IFNG, SOCS5, TNFB, TNFRSF1B, PIAS3, PIASY, CXCL10, ATM, NBN, MRE11, MLH1, PMS2,* and *TP53BP1*.



Methods and Algorithms:

Re-analysis of two DMG datasets presented in the public repository of functional genomic data Gene Expression Omnibus was carried out.




RESEARCH ARTICLE

Epigenetics and Proteomics Join Transcriptomics in the Quest for Tuberculosis Biomarkers

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University of Naples Federico II, Naples, Italy; Wolski, Wolski, Systems Biology IT, Systemtech, Zürich, Switzerland

to this article.

...e-third of the world's population is currently latently infected with *Mycobacterium tuberculosis*. ...tion (LTBI) progresses into active tuberculosis (TB) disease in ~5 to 10% of infected individuals. ...

GSE72338
Infinium human methylation450K BeadChip (Illumina)
Cape Town, South Africa

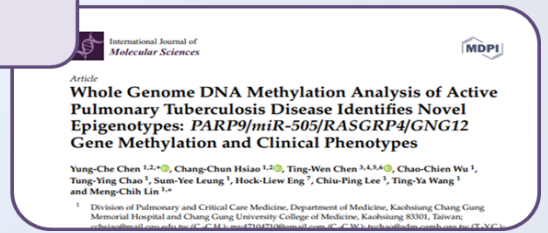
TB (n=8) ↔ **LTBI (n=8)**

PBMC ↔ **neutrophils** | **PBMC** ↔ **neutrophils**

TB (n=12) ↔ **healthy individuals (n=6)**

PBMC

GSE118469
Infinium HumanMethylation450K BeadChip v1.2
Taiwan


International Journal of *Molecular Sciences*

MDPI

Article

Whole Genome DNA Methylation Analysis of Active Pulmonary Tuberculosis Disease Identifies Novel Epigenotypes: *PARP9/miR-505/RASGRP4/GNG12* Gene Methylation and Clinical Phenotypes

Yung-Che Chen [1,2,*], Chang-Chun Hsiao [1,2], Ting-Wen Chen [3,4,5,6], Chao-Chien Wu [1], Tung-Ying Chao [1], Sum-Yee Leung [1], Hock-Liew Eng [7], Chiu-Ping Lee [1], Ting-Ya Wang [1] and Meng-Chih Lin [1,*]

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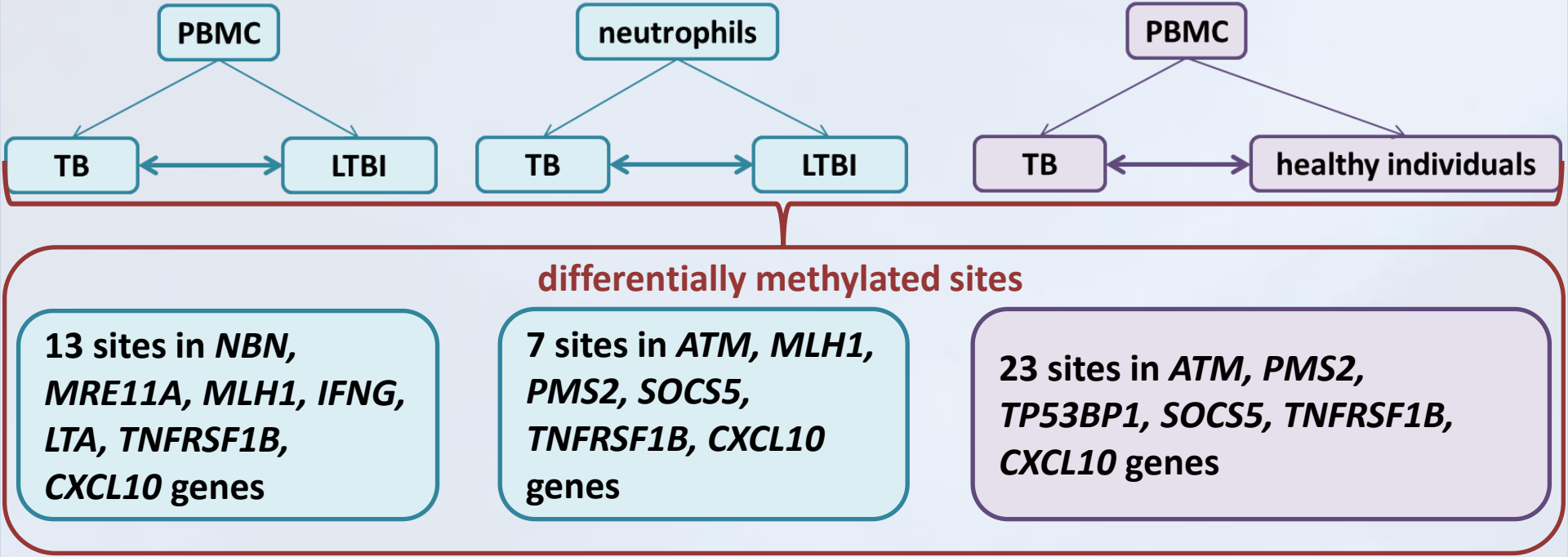
Results:

- Taking into account the FDR correction no significant differences were obtained in any of the comparisons. Further, we carried out the analysis, focusing on the initial level of significance achieved, without introducing any corrections.

- Only the *SOCS5* gene (out of 13 genes) included in the Top-50 DMG list, topped the DMG list when comparing neutrophils from TB and LTBI patients (p = 0.00000226)



Results:

UCSC Genome Browser

We annotated **differentially methylated sites** with the UCSC resource.

In two datasets, 39 CpG-sites were identified in target genes that were differentially methylated between TB patients and healthy controls. The position of three CpG-sites in the genome allows them to be attributed to the sphere of influence of two genes: cg11588932 (*ATM, NPAT*), cg13846866 (*MLH1, EPM2AIP1*), cg23492613 (*MRE11A, ANKRD49*).

There are 32 sites in the gene body (targeted or co-localized) (4 in exons and 28 in introns); 7 sites are located in the intergenic space near the promoter region of the gene.

localization in :

- **POLR2A binding region - 8 CpG-sites**
- **open heterochromatin - 12 CpG sites**
- **CpG-islands - 5 sites**
- **near CpG-islands - 21 sites**
- **near the promoters of target (and co-localized) genes - 26 sites**

- **Only cg21288207 is included directly in the promoter of the target gene (*PMS2*).**
- **2 CpG-sites are included in the co-localized genes promoter sequence:**
  - **cg23492613 - ANKRD49**
  - **cg16280132 - ATP6V1G2**

Results:

UCSC Genome Browser

9 CpG-sites are located in transcription factor binding regions.

CpG-site	target gene	Region of binding TF
cg05033322	*ATM,NPAT* (ex 1)	E2F6, PHF8, TAF1, E2F1, MYC, RUNX3
cg13846866	*MLH1* (in 1), *EPM2AIP1*	E2F1, MAX, E2F6, PHF8, REST
cg23492613	*MRE11* (in 1), *ANKRD49*	MAX, TAF1, PHF8, E2F4
cg21288207	*PMS2* (ex1), *AIMP2*	PHF8, MAX, SP1, E2F6, TAF1, **ELF1#**, GABPA, E2F4
cg19570749	*PMS2* (in 7)	**GATA3#**, JUN
cg03651680	*SOCS5* (in 1)	PHF8, TAF1, EGR1, ZNF263
cg17428954	*TNFRSF1B* (in 8)	CTCF, RAD21
cg24682307	*TNFRSF1B* (ex1)	PHF8, TAF1, **IRF1***, E2F6, MAX
cg03617487	*TP53BP1*	CTCF, RAD21

*** IRF1 - activates the transcription of genes involved in the response to viruses and bacteria**
 # ELF1 and GATA3 - regulate gene expression during the development of T-lymphocytes

Conclusion:

- ✓ **Thus, these findings indicated that DNA methylation is important in TB development.**

- ✓ **The differential value of DNA methylation of 13 target genes associated with TB, as well as the localization of these sites in the binding regions of transcription factors involved in the development of the immune response to infection, confirm the involvement of protein products of target genes in the pathogenesis of TB.**

The work was carried out within the framework of the State Task of the Ministry of Science and Higher Education No. 122020300041-7.

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