

Gene Expression Profiles of Pediatric Tuberculosis Patients and Exposed Controls from India

Jeffrey Tornheim, MD MPH

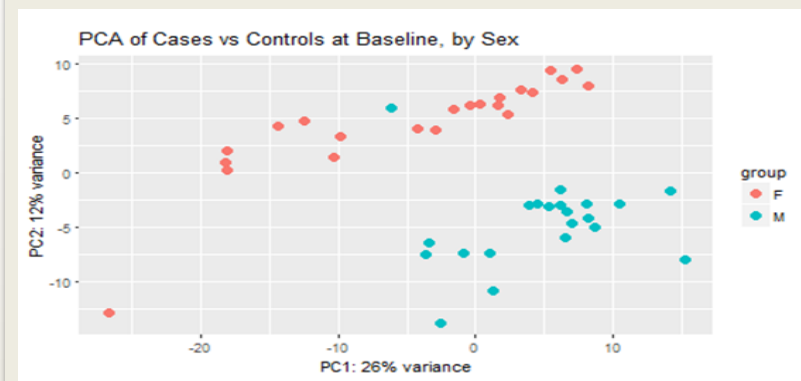
Assistant Professor of Medicine
Center for Clinical Global Health Education
Division of Infectious Diseases
Johns Hopkins University School of Medicine
tornheim@jhu.edu

Which patients have TB? When are they better?

- TB is the world's # 1 infectious disease killer. $>1/4^{\text{th}}$ cases occur in India.¹
- Current diagnostics tests are frequently inadequate for pediatric TB²
- Previously reported transcriptomic profiles associated with TB have included few children, few Indian patients, few microbiologically confirmed cases, and have primarily used microarray instead of RNAseq.
- Methods: case control study to evaluate RNAseq-derived transcriptomic signatures of TB among HIV-negative Indian children <15 with confirmed TB
- Cases were age-/sex-matched to TST & IGRA-neg household contacts of cases
- Gene signatures were evaluated for overlap with published studies

¹ WHO Global TB Report 2016.

² Perez-Velez et al. NEJM 2013

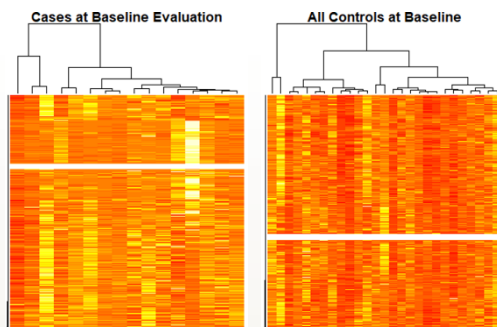


Sex and age group were identified as significant factors in principle component analysis

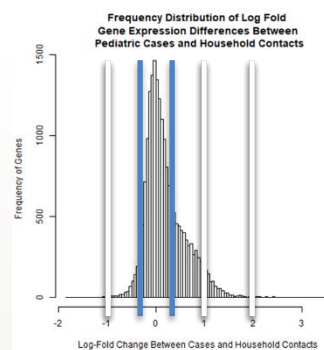
Results of Differential Gene Expression Analysis

Aim 1: Pediatric cases (N=16) vs. controls (N=32) at enrollment

Gene Counts



Fold Change Distribution

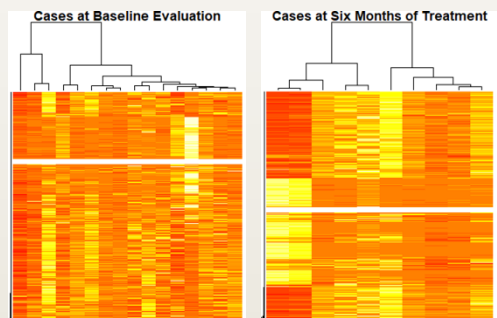


Number of Differentially Expressed Genes between Cases and Controls, by False Discovery Rate (FDR) and Log₂-Fold Change

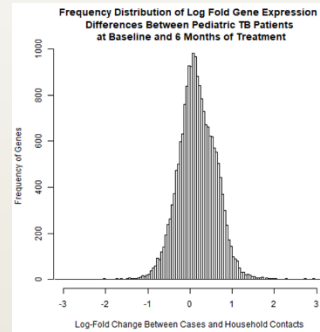
	Any	Log ₂ >0.5	Log ₂ >1.0	Log ₂ >1.5	Log ₂ >2.0
FDR=0.10	288	226	100	35	13
FDR=0.05	123	111	70	33	13
FDR=0.01	58	31	25	18	10

Aim 2: Cases at baseline and 6 months of treatment

Gene Counts



Fold Change Distribution



Number of Differentially Expressed Genes between Cases and Controls, by False Discovery Rate (FDR) and Log₂-Fold Change

	Any	Log ₂ >0.5	Log ₂ >1.0	Log ₂ >1.5	Log ₂ >2.0
FDR=0.10	1014	683	28	1	1
FDR=0.05	568	487	25	1	1
FDR=0.01	0	0	0	0	0

Concordance with Published Gene Signatures for TB

Using a False Discovery Rate of <0.05 and a log2 Fold-Change of >1.0

CTRIUMPh	TB vs. Not TB					TB vs. LTBI		Inflammation	Interferon
	Berry ¹ 393 Genes	Jenum ² 12 Genes	Lee ³ 267 Genes	Maertzdorf ⁴ 4 Genes	Sweeney ⁵ 3 Genes	Anderson ⁶ 42 Genes	Kaforou ⁷ 27 Genes	Obermoser ⁸ 74 Genes	Obermoser ⁸ 79 Genes
AIM2	X								X
ANKRD22	X						X		
C1QB	X						X		
C1QC							X		
DEFA3						X			
DHRS9	X								X
FAM26F	X								
FCGR1A	X	X	X				X		
FCGR1B	X		X				X		
GBP1	X			x					X
GBP5	X				X	X			X
GBP6	X					X	X		X
GPR84	X								
MPO							X		
PRRG4	X								
SEPT4	X								X

- 16 / 70 genes overlapped with other published signatures.
- The most overlap was seen with Sweeney (33.3%) and Kaforou (25.9%)

¹ Berry, et al. Nature 2010

⁵ Sweeney, et al. Lancet Resp Med 2016

² Jenum, et al. Sci Reports 2016

⁶ Anderson, et al. NEJM 2014

³ Lee et al. BMC Bioinformatics 2016

⁷ Kaforou, et al. PLoS One 2013

⁴ Maertzdorf, et al. EBMO Mol Med 2016

⁸ Obermoser, et al. Immunity 2014

Acknowledgements

JHU team

Amita Gupta

BJMC Team

Mandar Paradkar

Nikhil Gupte

Vandana Kulkarni

Neeta Pradhan

Chhaya Valvi

Rewa Kohli

Vidya Mave

Institute of Bioinformatics Team

Anil Madungundu

Sreelakshmi Sreenivasamurthy

Remya Raja

Akhilesh Pandey

NIRT Team

Padmapriyadarsini Chandrasekaran

Shri Vijay Bala Yogendra Shivakumar

Funding

NIH/DBT RePORT India Consortium

Indo (DBT)-US (NIH) (USB1-31147-XX-13 CRDF/NIH)

NIH/NIAID (UM1AI069465)

Fogarty International Center BJGMC JHU HIV TB Program (D43TW009574)

UJMT Fogarty Global Health Fellows Program (R25TW009340)

Ujala Foundation, Gilead Foundation, Wyncote Foundation, and Persistent Systems

Summary of Published Signatures for Comparison

Author	Age	Application	Sequencing Method	# of Genes in Signature	False Discovery Rate	Log Fold Difference in Expression
Anderson (NEJM 2014)	Children	TB vs. LTBI	Microarray	42	--	0.5
Berry (Nature 2010)	Adults	TB vs. Not TB	Microarray	393	0.01	1
Bloom (PLoS One 2012)	Adults	Post Treatment	Microarray	320	0.01	1
Jenum (Sci Reports 2016)	Children	TB vs. Not TB	dcRT-MLPA	12	--	--
Kaforou (PLoS One 2013)	Adults	TB vs. Not TB	Microarray	53	--	0.5
Kaforou (PLoS One 2013)	Adults	TB vs. LTBI	Microarray	27	--	0.5
Lee (BMC Bioinformatics 2016)	Adults	TB vs. LTBI	Microarray	169	0.05	1
Lee (BMC Bioinformatics 2016)	Adults	TB vs. Not TB	Microarray	267	0.05	1
Maertzdorf (EMBO Mol Med 2015)	Adults	TB vs. Not TB	RT-PCR	4	--	--
Obermoser (Immunity 2014)	Adults	Inflammation	Microarray	74	0.1	1
Obermoser (Immunity 2014)	Adults	Interferon γ	Microarray	79	0.1	1
Sweeney (Lancet Resp Med 2016)	Mixed	TB vs. Not TB	Public Data	3	0.01	0.58
Zak (Lancet 2016)	Children	Incident LTBI	RNAseq	16	--	--

- Published signatures employed different methods from each other, specifically with respect to FDR and thresholds of differential expression.