**Supplementary Methods**

*Cell purification and RNA isolation*

PBMCs were collected from all subjects in the discovery and validation cohorts. To isolate PBMCs, 5 mL of peripheral blood was drawn from the ulnar vein of each person. After the blood was collected, PBMCs were immediately isolated by the Ficoll-Hypaque (Histopaque 1077, Sigma-Aldrich Corporation, Missouri, USA) density gradient centrifugation method. In brief, the whole blood was mixed with RPMI Medium 1640 basic and Ficoll-Hypaque at a ratio of 1:1:1, and the blood cells were spread over the Ficoll-Hypaque separation solution. The mixture was then centrifuged horizontally at 2,000 rpm for 20 min at room temperature. After centrifugation, the white misty cell layer was carefully collected into a new centrifuge tube and washed twice with RPMI Medium 1640 basic at 2,000 rpm for 5 min at 4 °C. The isolated PBMCs were transferred into 1.5-mL tubes, and 1 mL of TRIzol (Ambion, Thermo Fisher Scientific, Massachusetts, USA) was added for subsequent total RNA extraction. Total RNA was then extracted from the PBMCs using the TRIzol reagent (Invitrogen, California, USA) and purified with the mirVana RNA Isolation Kit (Ambion, Thermo Fisher Scientific, Massachusetts, USA) according to the manufacturer's protocol. The purity and concentration of RNA were determined from OD260/280 readings using NanoDrop ND-1000. RNA integrity was determined by 1% formaldehyde denaturing gel electrophoresis. Only RNA extracts with RNA integrity number values greater than 6 were used for further experiments.

*RNA-seq library preparation and sequencing*

To profile PBMC circRNA expression in the discovery cohort, we constructed ribosomal RNA (rRNA)-depleted RNA-seq libraries using PBMC total RNA from each person in the discovery cohort. First, mRNA and non-coding RNAs were enriched by removing rRNA from the total RNA with a Ribo-Zero rRNA Removal Kit (Illumina, California, USA). Next,

the enriched mRNA and non-coding RNAs were fragmented into short fragments (approximately 200-700 bp) using the fragmentation buffer. Then, the first-strand cDNA was synthesized with random hexamer primer using the fragments as templates. Buffer, dNTPs, RNase H and DNA polymerase I were added to synthesize the second-strand cDNA. The double-stranded cDNA was purified with the QiaQuick PCR Extraction Kit (Qiagen, Hilden, Germany) and then used for end-polishing. Sequencing adapters were ligated to the fragments, and the second strand was then degraded using Uracil-N-Glyosylase (UNG). The fragments were purified by agarose gel electrophoresis and enriched by PCR amplification. Finally, these rRNA-depleted RNA-seq libraries were sequenced using the Illumina HiSeq 4000 sequencing platform (Novogene, Beijing, China).

*CircRNA expression microarray*

The expression of PBMC circRNAs in the discovery cohort was also profiled using the CapitalBio Technology Human CircRNA Array v2 (CapitalBio Technology, Beijing, China). The CaptialBio Human CircRNA Array v2 was designed with four identical arrays per slide (4 x 180K format), and each array contained probes interrogating approximately 170,340 human circRNAs. These 170,340 circRNA sequences were collected from several public resources, including circBase [1], Deepbase [2], and all human circRNAs published by Rybak-Wolf *et al.* [3]. Each circRNA was simultaneously detected by a long probe and a short probe. The circRNA array also contained 4,974 Agilent control probes.

cDNA labeled with a fluorescent dye (Cy3-dCTP) was produced by Eberwine's linear RNA amplification method and subsequent enzymatic reaction. Double-stranded cDNAs (containing the T7 RNA polymerase promoter sequence) were synthesized from 5 μg of digested RNA using the CbcScript reverse transcriptase with cDNA synthesis system according to the manufacturer's protocol (CapitalBio) with T7 Oligo (dN). After completion

of double-stranded cDNA (dsDNA) synthesis using DNA polymerase and RNase H, the dsDNA products were purified using a PCR NucleoSpin Extract II Kit and eluted with 30 μL of elution buffer. The eluted double-stranded cDNA products were vacuum-evaporated to 16 μL and subjected to 40-μL *in vitro* transcription reactions at 37 °C for 14 hours using a T7 Enzyme Mix. The amplified cRNA was purified using the RNA Clean-up Kit.

The Klenow enzyme labeling strategy was adopted after reverse transcription using CbcScript II reverse transcriptase. Briefly, 2 μg of amplified RNA was mixed with 4 μg of random nanomer, denatured at 65 °C for 5 min, and cooled on ice. Then, 5 μL of 4× first-strand buffer, 2 μL of 0.1 M DTT, and 1.5 μL of CbcScript II reverse transcriptase were added. The mixtures were incubated at 25 °C for 10 min, then at 37 °C for 90 min. The cDNA products were purified using a PCR NucleoSpin Extract II Kit and vacuum-evaporated to 14 μL. The cDNA was mixed with 4 μg of random nanomer, heated to 95 °C for 3 min, and snap cooled on ice for 5 min. Then, 5 μL of Klenow buffer, dNTPs, and Cy5-dCTP or Cy3-dCTP were added to final concentrations of 240 μM dATP, 240 μM dGTP, 240 μM dTTP, 120 μM dCTP, and 40 μM Cy-dCTP. Then, 1.2 μL of Klenow enzyme was added, and the reaction was performed at 37 °C for 90 min. Labeled cDNA was purified with a PCR NucleoSpin Extract II Kit and resuspended in elution buffer. Labeled controls and test samples labeled with Cy3-dCTP were dissolved in 80 μL of hybridization solution containing 3×SSC, 0.2% SDS, 5× Denhardt's solution, and 25% formamide. DNA in hybridization solution was denatured at 95 °C for 3 min prior to loading onto a microarray. Arrays were hybridized in an Agilent Hybridization Oven overnight at a rotation speed of 20 rpm and a temperature of 42 °C, followed by washing with two consecutive solutions (0.2% SDS, 2× SSC at 42 °C for 5 min, and 0.2× SSC for 5 min at room temperature).

After hybridization, the circRNA array data were summarized, normalized, and quality tested by the Agilent GeneSpring software V13.0.

**Reference:**

1. Glazar P, Papavasileiou P, Rajewsky N. circBase: a database for circular RNAs. *RNA*. 2014;20(11):1666-1670.
2. Zheng L-L, Li J-H, Wu J, et al. deepBase v2.0: identification, expression, evolution and function of small RNAs, LncRNAs and circular RNAs from deep-sequencing data. *Nucleic Acids Research*. 2016;44(D1).
3. Rybak-Wolf A, Stottmeister C, Glažar P, et al. Circular RNAs in the Mammalian Brain Are Highly Abundant, Conserved, and Dynamically Expressed. *Molecular Cell*. 2015;58:1-16.

**Supplementary Table S1.** The human subjects in the discovery cohort

| | Healthy control | TB patient |
|---|---|---|
| Young | Male, 23-year-old | Male, 23-year-old |
| Senior | Female, 68-year-old | Female, 75-year-old |

**Supplementary Table S2.** The human subjects in the validation cohort

| | Healthy control | TB patient | |
|---|---|---|---|
| Age | 26.7±7.6 | 32.7±14.2 | *t*-test: $P = 0.256$ |
| Gender | Male (n = 7)<br>female (n = 4) | Male (n = 7)<br>female (n =3) | $\chi^2$-test: $P = 0.757$ |

**Supplementary Table S3.** The RNA-seq datasets used in this study

| Sample type | SRA ID | Sample name | Library size (reads) | References |
|---|---|---|---|---|
| Anucleate cell | ERR335311 | Platelets_M1 | 104,242,054 | Kissopoulou *et al.*, 2013 [1] |
| | ERR335312 | Platelets_F | 90,831,980 | |
| | ERR335313 | Platelets_M2 | 103,456,200 | |
| | SRR2038798 | RBC | 138,396,568 | Alhasan *et al.*, 2016 [2] |
| Whole blood | SRR2537079 | H_1_wholeBlood | 57,853,921 | Memczak *et al.*, 2015 [3] |
| | SRR2537080 | H_1_rep_wholeBlood | 169,863,444 | |
| | SRR2537081 | H_2_wholeBlood | 48,035,915 | |
| | SRR2537082 | H_3_wholeBlood | 164,927,445 | |
| | SRR2537083 | H_4_wholeBlood | 171,759,972 | |
| | SRR2537084 | H_5_wholeBlood | 170,202,868 | |
| PBMC | SRR5932796 | TB young | 169,752,312 | This study |
| | SRR5932793 | Control young | 186,986,224 | |
| | SRR5932794 | TB senior | 137,311,530 | |
| | SRR5932795 | Control senior | 167,231,628 | |

[1] Kissopoulou *et al.*, Next Generation Sequencing Analysis of Human Platelet PolyA+ mRNAs and rRNA-Depleted Total RNA. *PLoS ONE* **8**, e81809 (2013).
[2] Alhasan *et al.*, Circular RNA enrichment in platelets is a signature of transcriptome degradation. *Blood* **127**, e1-e11 (2016).
[3] Memczak *et al.*, Identification and Characterization of Circular RNAs As a New Class of Putative Biomarkers in Human Blood. *PLoS ONE* **10**, e0141214 (2015).

**Supplementary Table S4.** The divergent qRT-PCR primers designed for the validation of circRNA dysregulation

| circBase ID | Left primer | Right primer |
|---|---|---|
| hsa_circ_0000681 | GATGAATGTTCCCAGCCTGT | AGGGCAGGAGAATGTGACAA |
| hsa_circ_0008797 | ATTACGGGACCCAAATGTCA | TCCCCTGGAAATATTGGTTG |
| hsa_circ_0002113 | GTTTCACACTCCACCAAGCA | CGGCAGTGAAGTCACACTCT |
| hsa_circ_0002362 | TCTGCTGAAGGTCAACGAAA | GCAAACTGTCCAAAGGGAAG |
| hsa_circ_0000414 | GGAGAAGGAGAGATTTTTGAGG | GTGCCCAGGACCAAAGTAAT |
| hsa_circ_0063179 | GATCTGGAGCGAGTGGAGTG | TGTGTTCGTATCGCATTTTCA |
| hsa_circ_0002908 | GGGGCAATGCACTAGAAAAG | TCCTTTTGGCAAATAACGAAT |

**Supplementary Table S5.** The KEGG pathways with significant circRNA dysregulation

| | | Adjusted *P*-value[1] | |
|---|---|---|---|
| Pathway ID | Pathway description | Young case-control pair | Senior case-control pair |
| hsa04060 | Cytokine-cytokine receptor interaction | 3.6E-02 | 4.2E-02 |
| hsa04062 | Chemokine signaling pathway | 3.0E-04 | 1.5E-02 |
| hsa04666 | Fc gamma R-mediated phagocytosis | 1.4E-04 | 3.5E-03 |
| hsa04722 | Neurotrophin signaling pathway | 3.7E-03 | 2.1E-02 |
| hsa05100 | Bacterial invasion of epithelial cells | 1.1E-08 | 1.3E-03 |

[1] *P*-values were adjusted by *Benjamini* and *Hochberg* procedure

**Supplementary Table S6.** Multivariable logistic regression on TB status

| | Odds ratio (log) | *P*-value |
|---|---|---|
| circRNA-based TB index: - vs. + | 2.996 | 0.038 |
| Number of cavities | 16.11 | 0.998 |
| Diameter of the largest cavity (mm) | 0.051 | 1.000 |
| Sputum smear grade | 1.255 | 1.000 |

**Supplementary Table S7.** Correlation between circRNA-based TB index and clinical indicators of TB severity

| | $\rho$[1] | *P*-value[1] |
|---|---|---|
| Number of cavities | 0.381 | 0.088 |
| Diameter of the largest cavity (mm) | 0.261 | 0.253 |
| Sputum smear grade | 0.188 | 0.416 |

[1] *Spearman*'s rank correlation test was used to compute the correlation coefficients ($\rho$) and *P*-values.