

# **RNA-binding protein STAT3 regulates GSDME mRNA homeostasis to mediate pyroptosis in Renal Ischemia-Reperfusion Injury**

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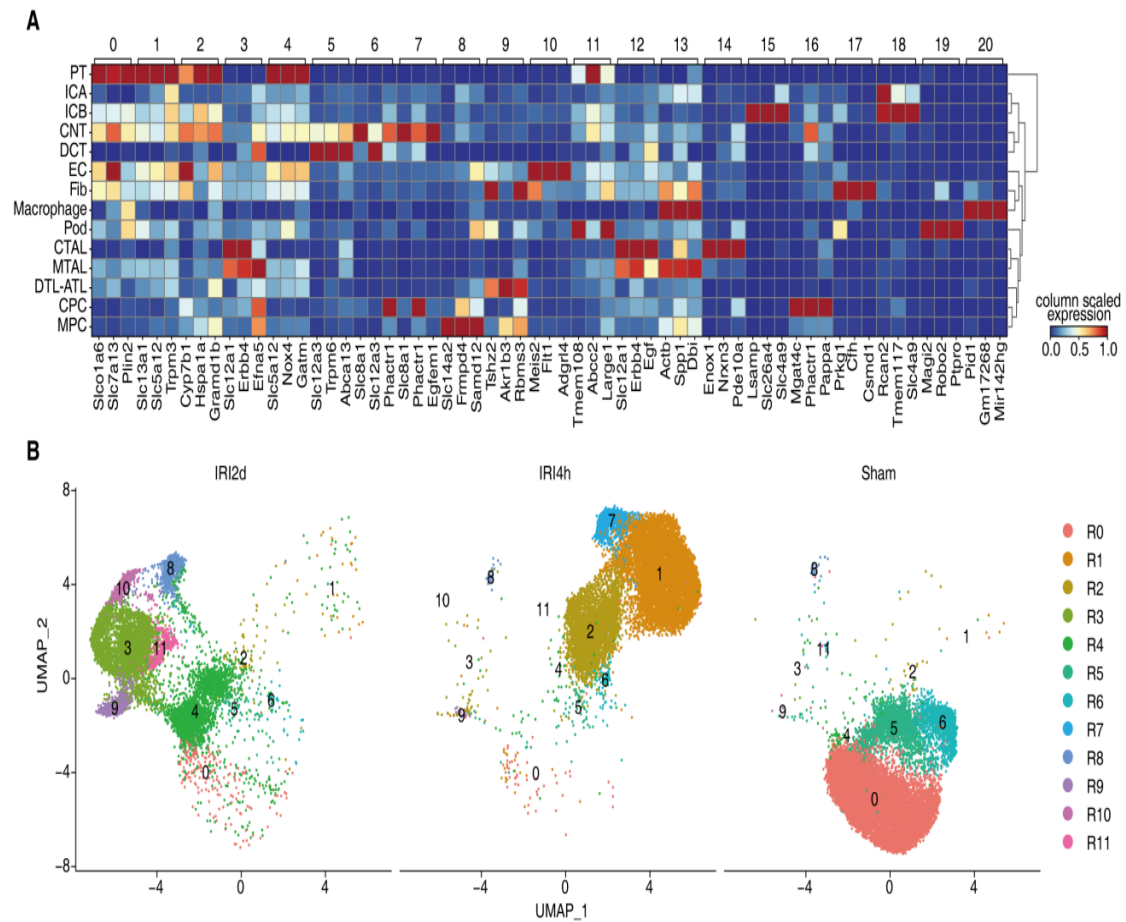
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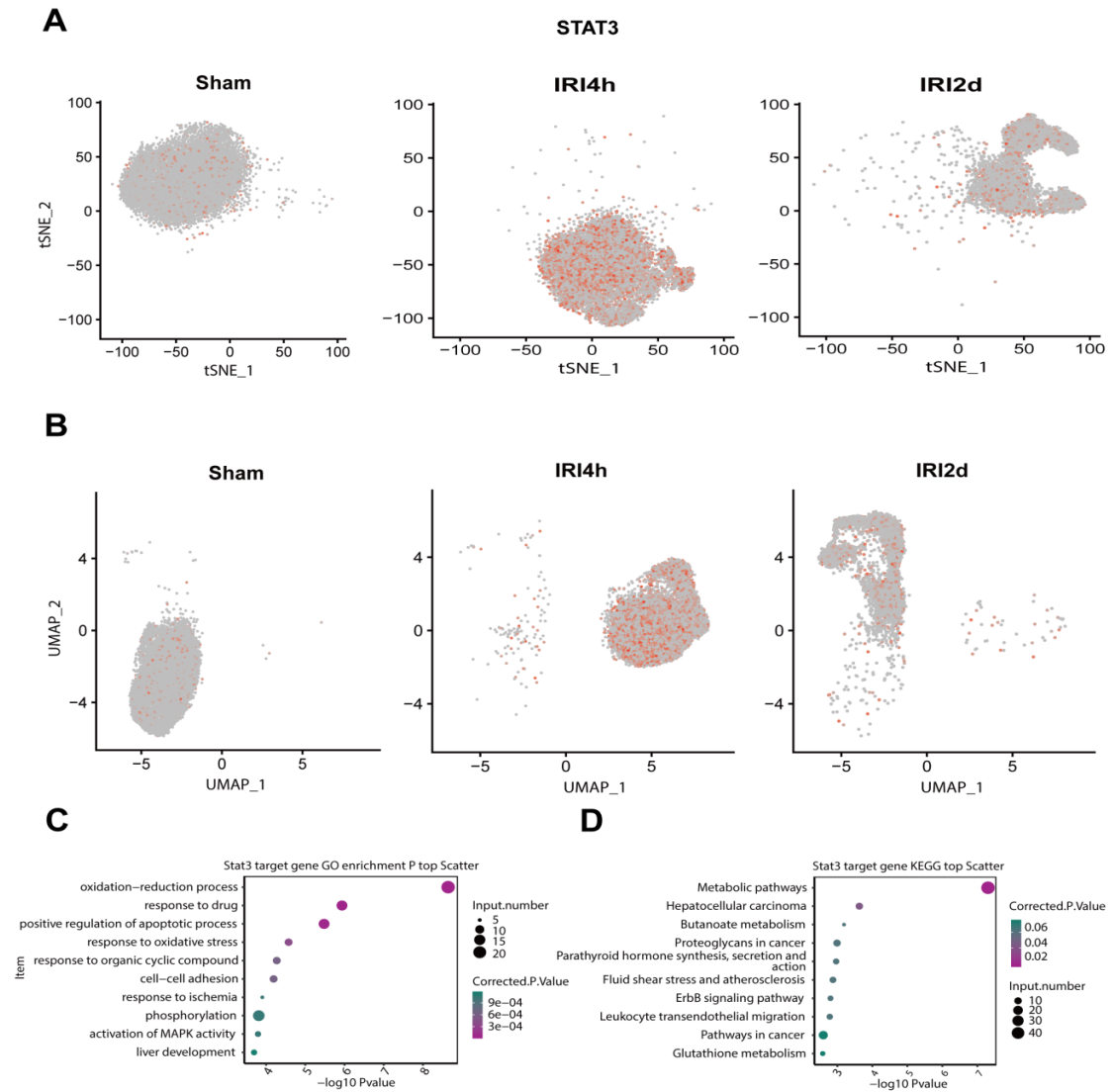
**Supplementary Table 1 - Specific primers for quantitative RT-PCR**

**Supplementary Table 2 - Primers for STAT3 knockout (siRNA sequences used for gene knockdown)**



**Supplemental Figure S1 RBP expression exhibits significant differences across various cell clusters**

(A) Matrixplot showing the expression of the top 3 marker genes of each cluster in different celltypes. (B) UMAP plot of scRNA-seq profile based on RBP expression module within each group.



## Supplemental Figure S2 Single-cell analysis reveals the expression and function of STAT3 in AKI

(A-B) t-SNE and UMAP plots of STAT3 expression in Sham, IRI-4h, and IRI-2d groups. Each point represents a cell, with cells from each group color-coded to visualize the differences in STAT3 expression and inter-group clustering. (C) GO enrichment analysis of STAT3 target genes. (D) KEGG pathway enrichment analysis of STAT3 target genes.

**Supplementary Table 1 Specific primers for quantitative RT-PCR**

| <b>Mouse Genes</b> | <b>Forward primer (5'–3')</b> | <b>Reverse primer (5'–3')</b> |
|--------------------|-------------------------------|-------------------------------|
| $\beta$ -actin     | CATTGCTGACAGGATGCAGAA         | ATGGTGCTAGGAGCCAGAGC          |
| KIM-1              | CAGGGAAGCCGCAGAAAA            | GAGACACGGAAGGCAACCAC          |
| MCP-1              | CTTCTGGGCCTGCTGTTCA           | CCAGCCTACTCATTGGGATCA         |
| TNF- $\alpha$      | CATCTTCTCAAAATTCGAGTGACAA     | TGGGAGTAGACAAGGTACAACCC       |
| IL-1 $\beta$       | GCTTCAGGCAGGCAGTAT            | ACAAACCGCTTTTCCATCT           |
| IL-6               | GAGGATACCACTCCCAACAGACC       | AAGTGCATCATCGTTGTTCATACA      |
| IL-18              | GACTCTTGCGTCAACTTCAAGG        | CAGGCTGTCTTTTGTCAACGA         |
| STAT3              | TACCATTGACCTGCCGATGT          | TCAAACGTGAGCGACTCAAAC         |
| GSDME              | GCGCACTAGCAGAAATGCCT          | CAGCAGAGGCAAACAATCGC          |

| <b>Human Genes</b> | <b>Forward primer (5'–3')</b> | <b>Reverse primer (5'–3')</b> |
|--------------------|-------------------------------|-------------------------------|
| STAT3              | TCTGTGTGACACCAACGACC          | TCCTCACATGGGGGAGGTAG          |
| GSDME              | ACATGCAGGTCGAGGAGAAGT         | TCAATGACACCGTAGGCAATG         |
| TNF- $\alpha$      | CCCAGGGACCTCTCTCTAATCA        | GCTACAGGCTTGTCACCTCGG         |
| IL-6               | CGGGAACGAAAGAGAAGCTCTA        | GAGCAG CCCCAGGGAGAA           |
| IL-1 $\beta$       | ACTACAGCAAGGGCTTCAGG          | CATATCCTGTCCCTGGAGGT          |
| MCP-1              | AGCAGCAAGTGTCCCAAAGA          | GGTGGTCCATGGAATCCTGA          |
| $\beta$ -actin     | CGCCGCCAGCTCACCATG            | CACGATGGAGGGGAAGACGG          |

**Supplementary Table 2 - Primers for STAT3 knockout (siRNA sequences used for gene knockdown)**

| Human Genes | Forward primer (5'–3') | Reverse primer (5'–3') |
|-------------|------------------------|------------------------|
| NC          | TTCTCCGAACGTGTCACGT    | ACGUGACACGUUCGGAGAATT  |
| si-STAT3-1  | TTCAGACCCGTCAACAAATTA  | UUUCAUCUGAAGACAGGACTT  |
| si-STAT3-2  | CTCAGAGGATCCCGGAAATTT  | UUCCGUUCCUGGUUCUCCTT   |
| si-STAT3-3  | TTGGGACCTGGTGTGAATTAT  | UUCAUUCUCUUCAGCUAGCTT  |