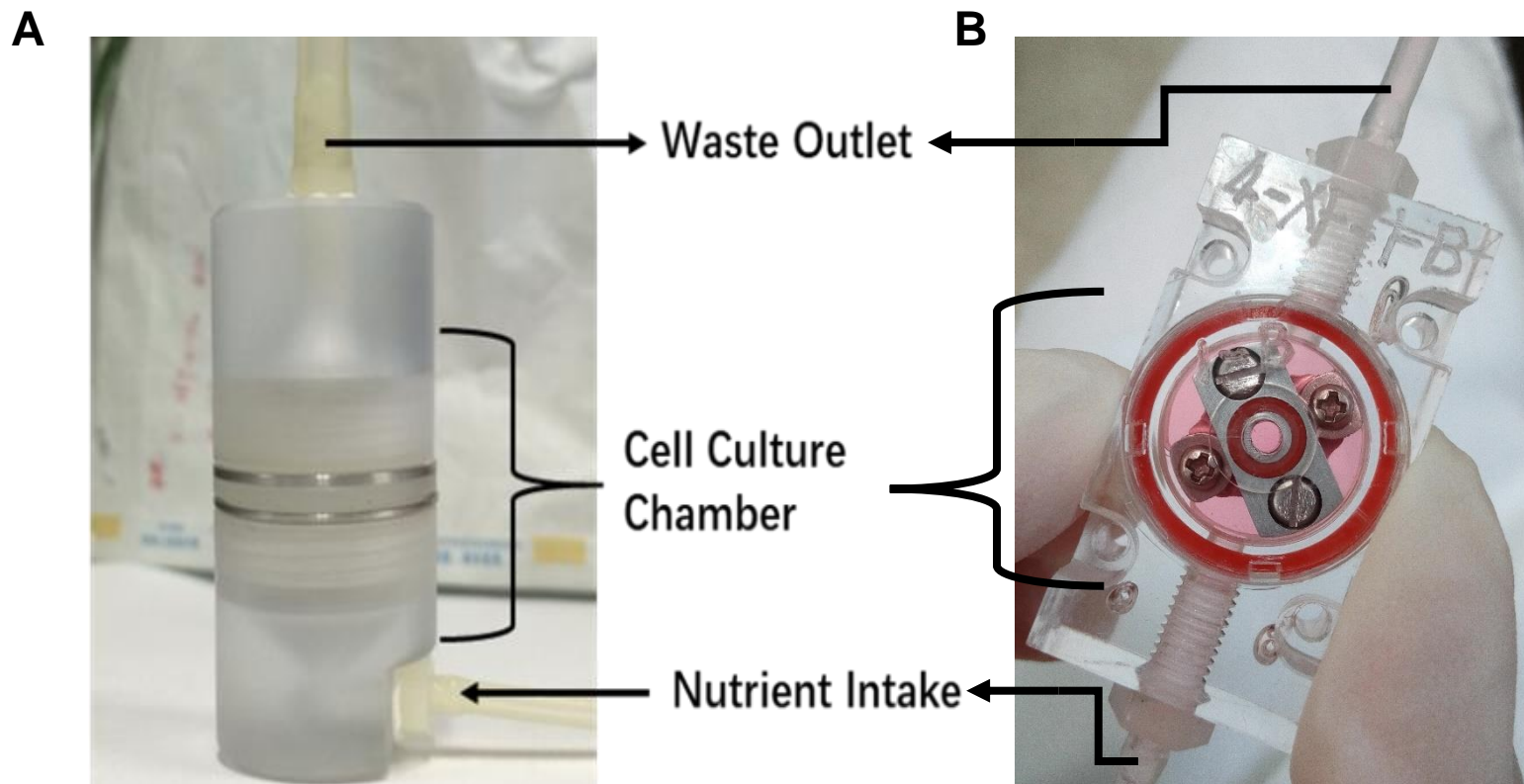


Supplementary Information

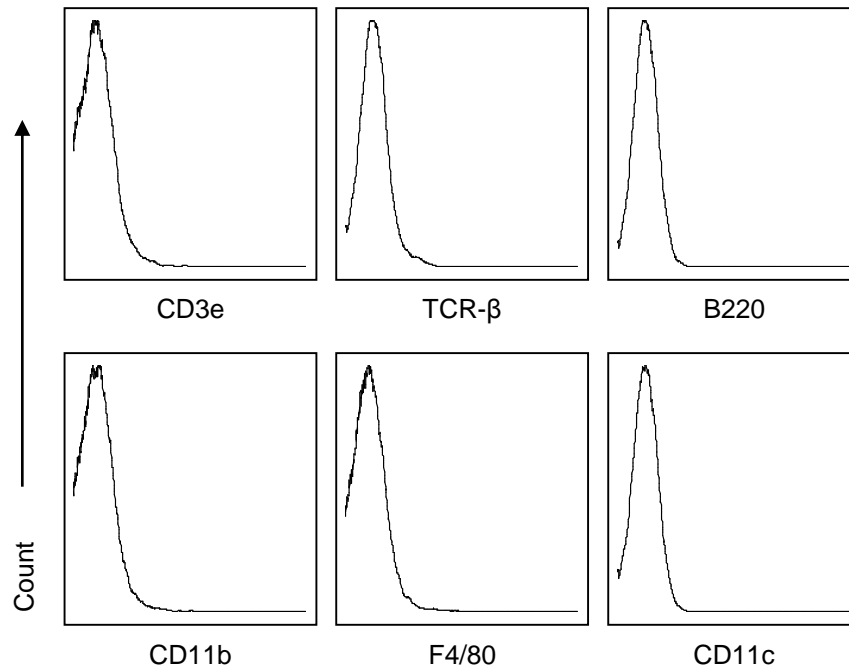
Spaceflight/Microgravity inhibits the proliferation of hematopoietic stem cells by decreasing Kit-Ras/cAMP-CREB pathway networks as evidenced by RNA-Seq assays

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and Yong Zhao^{1, 2, 3, #}

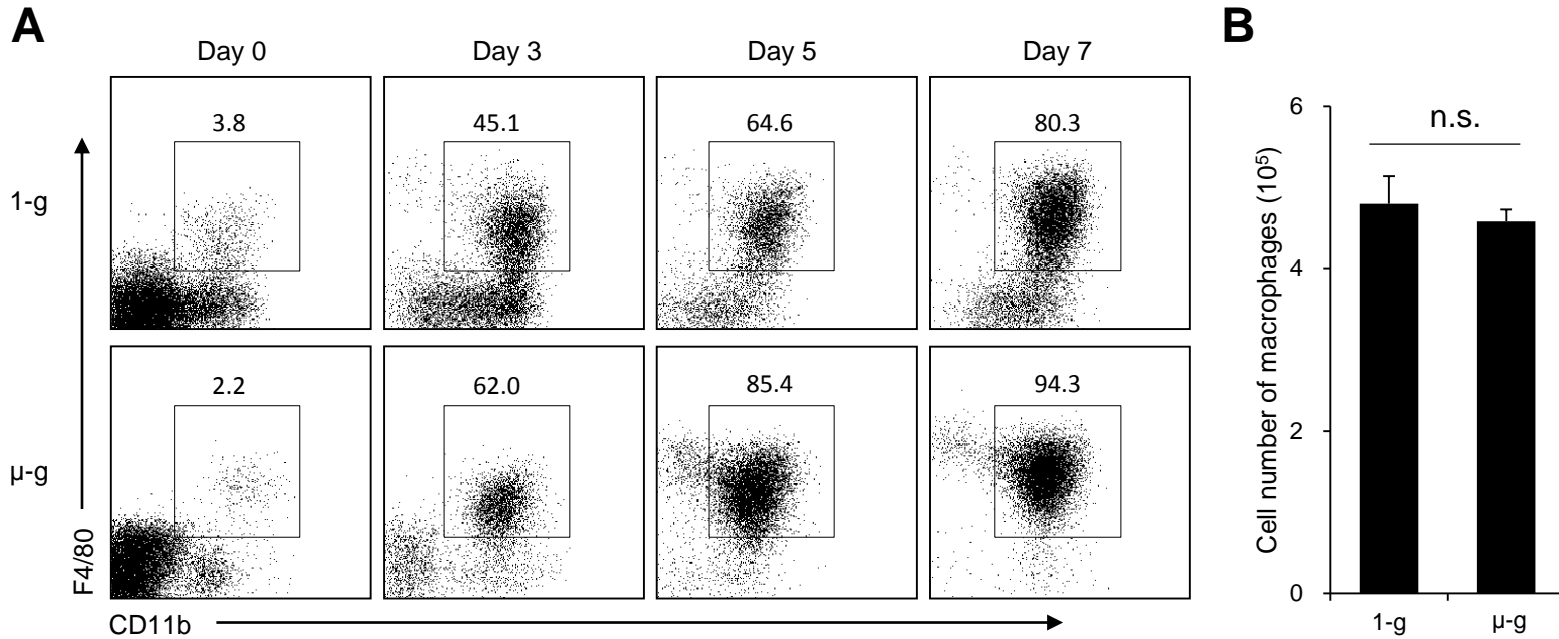


Supplementary Figure 1. The culture vessel of HSPCs on SJ-10 satellite and Tianzhou-1 cargo ship.

A, The culture vessel of HSPCs on SJ-10 satellite. **B**, The culture vessel of HSPCs on Tianzhou-1 cargo ship.

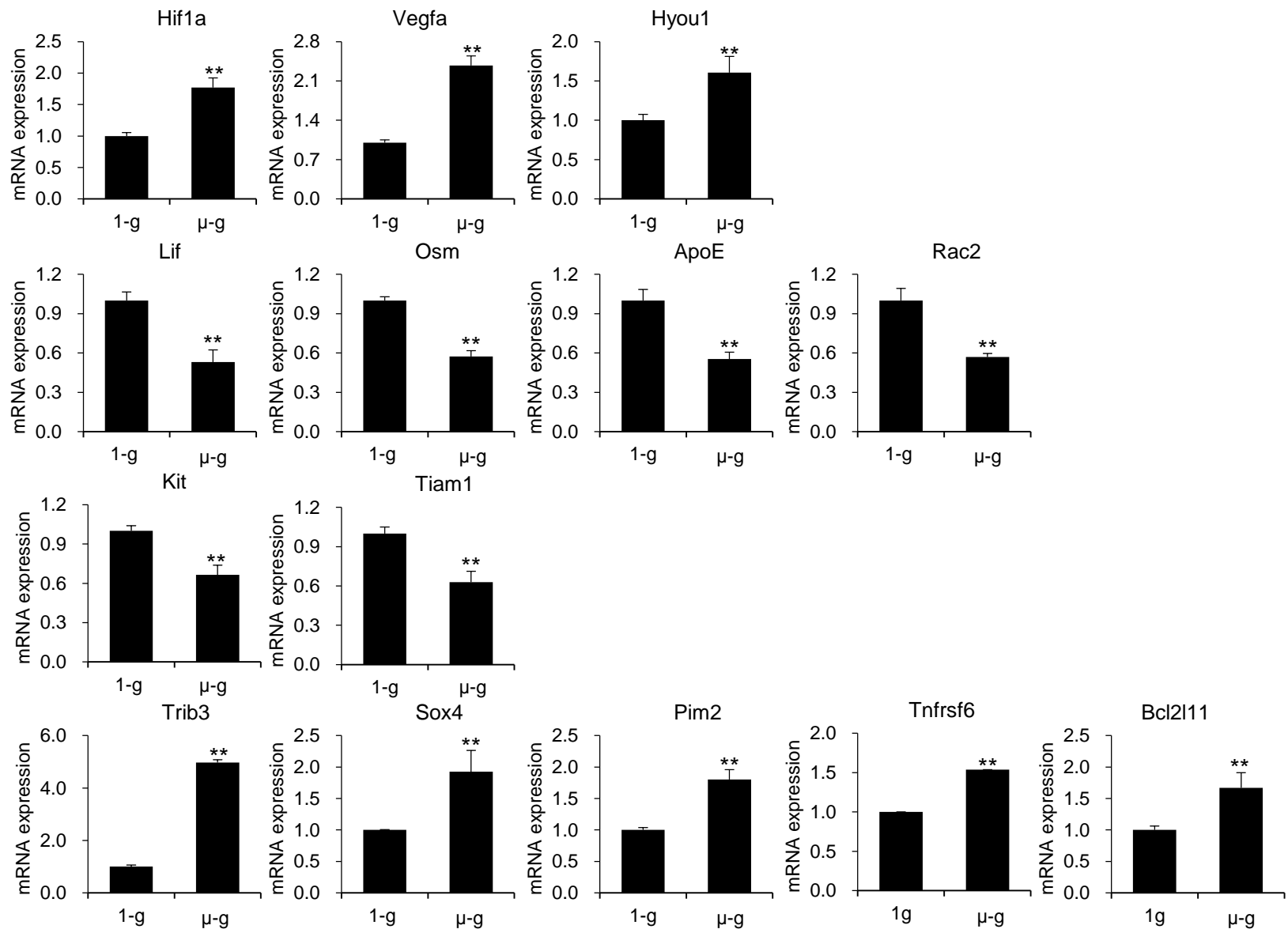


Supplementary Figure 2. The expressions of lineage specific makers in sorted HSPCs from bone marrow, including CD3 ϵ , TCR- β , B220, CD11b, F4/80 and CD11c.



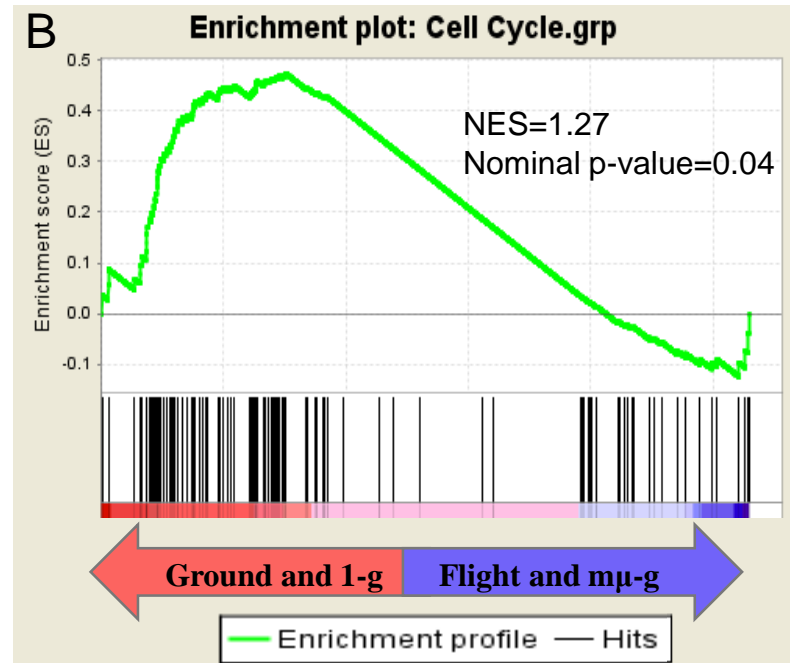
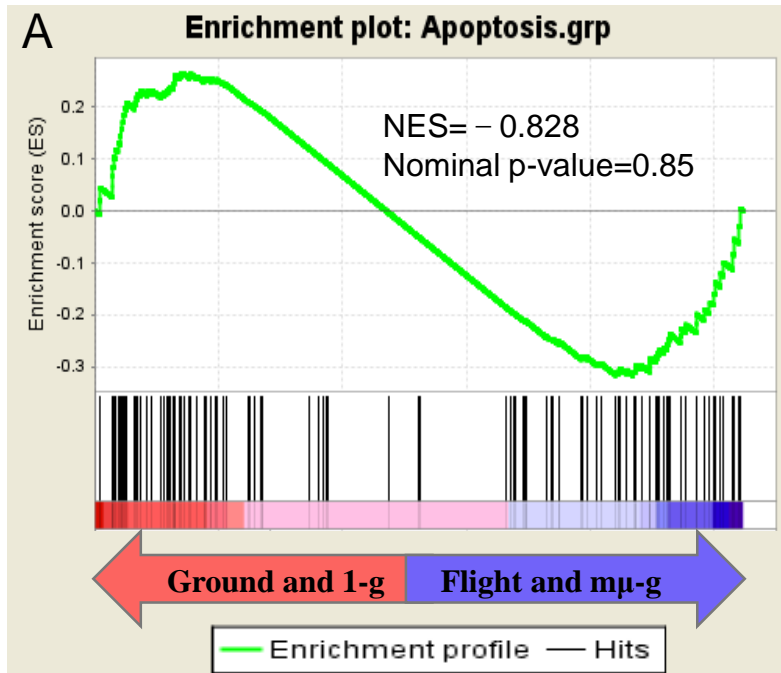
Supplementary Figure 3. There was no difference in directed differentiation of HSPCs into macrophages after culturing in normal ground control and simulated microgravity conditions for 12 days.

A, Typical FACS plot of macrophages expressing CD11b and F4/80 for HSPCs simulated by M-CSF for 0, 3, 5, 7 days under different gravity conditions. **B**, The number of macrophages differentiated by HSPCs after seven days under different gravity conditions. 1-g: Normal gravity. μ-g: Simulated microgravity.



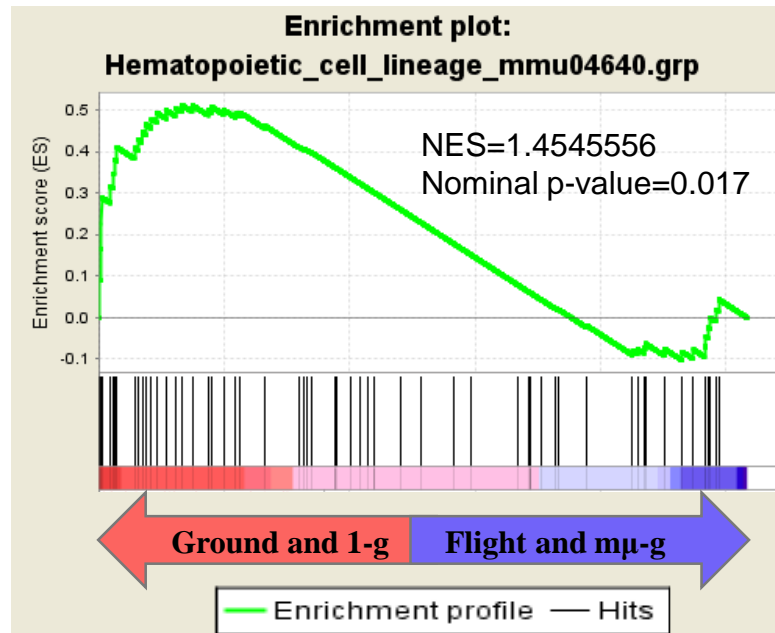
Supplementary Figure 4. The mRNA expressions of genes associated with processes of hypoxia stress, cell proliferation and cell apoptosis.

These genes include *Hif1 α* , *Vegfa*, *Lif*, *Osm*, *ApoE*, *Rac2*, *Kit*, *Tiam1*, *Trib3*, *Sox4*, *Pim2*, *Tnfrsf6* and *Bcl2l11*.



Supplementary Figure 5. GSEA analysis of DEGs between the control and microgravity-treated samples (including spaceflight and simulated microgravity experiments).

A, Apoptosis pathway; **B**, Cell cycle pathway. Profile of the running Enrichment Score and positions of gene set members on the ranked list of genes are shown. NES, normalized enrichment score.



Supplementary Figure 6. GSEA plot: Hematopoietic cell lineage pathway (mmu04640) analysis between the control and microgravity-treated samples.

Profile of the running Enrichment Score and positions of gene set members on the ranked list of genes are shown. NES, normalized enrichment score.