

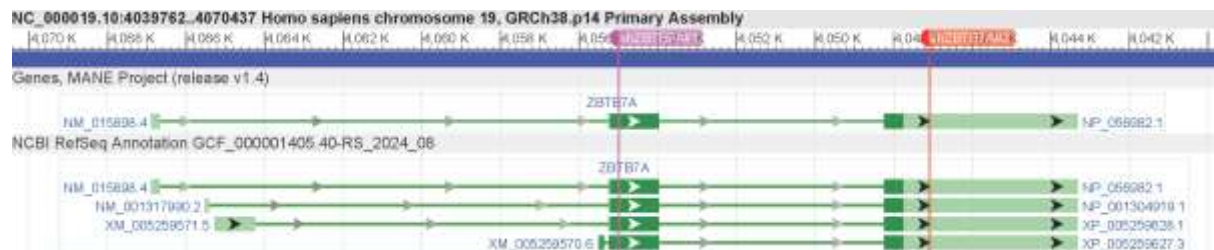
## Supplementary information

**Table S1 Primers used for RT-qPCR in this study.**

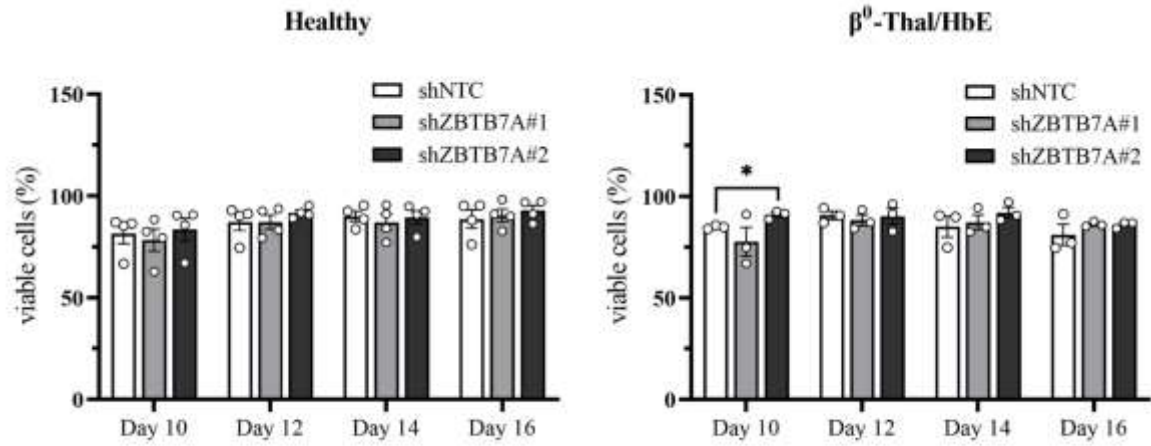
| Gene          | Primer name | Primer sequence (5'→ 3')      |
|---------------|-------------|-------------------------------|
| <i>RPS18</i>  | hRPS18-FW   | GGA TGA GGT GGA ACG TGT       |
|               | hRPS18-RV   | CTA GGA CCT GGC TGT ATT TTC   |
| <i>ZBTB7A</i> | hZBTB7A-FW  | CTT CAC CAG GCA GGA CAA       |
|               | hZBTB7A-RV  | GGT TCT TCA GGT CGT AGT TGT G |
| <i>BCL11A</i> | hBCL11A-FW  | GGG AAT TCT CGC CCG AG        |
|               | hBCL11A-RV  | GGG AAG TTC ATC TGG CAC T     |
| <i>KLF1</i>   | hKLF1-FW    | CGT ATG GCT TCT CCC CTG       |
|               | hKLF1-RV    | CAG AGG ATC CAG GTG TGA TAG   |
| <i>GATA1</i>  | hGATA1-FW   | CAG GAC AGG CCA CTA CCT AT    |
|               | hGATA1-RV   | CTG ACA ATC AGG CGC TTC TT    |
| <i>GATA2</i>  | hGATA2-FW   | CAA GGC TCG TTC CTG TTC A     |
|               | hGATA2-RV   | GCC CAT TCA TCT TGT GGT AGA   |
| <i>SOX6</i>   | hSOX6-FW    | CAG CAA CAG ATC CAG GTT CA    |
|               | hSOX6-RV    | GAA GAG GAA TCC CTG TTG GG    |
| <i>ATF4</i>   | hATF4-FW    | CCT CCA ACA ACA GCA AGG A     |
|               | hATF4-RV    | GCA TGG TTT CCA GGT CAT CTA   |
| <i>HBA</i>    | hHBA-FW     | TGG ACC CGG TCA ACT TCA AG    |
|               | hHBA-RV     | TCA CAG AAG CCA GGA ACT TGT   |
| <i>HBB</i>    | hHBB-FW     | GAA GGC TCA TGG CAA GAA AG    |
|               | hHBB-RV     | CAC TGG TGG GGT GAA TTC TT    |
| <i>HBG</i>    | hHBG-FW     | TCA CAG AGG AGG ACA AGG CTA   |
|               | hHBG-RV     | GCT TTA TGG CAT CTC CCA AG    |
| <i>HBD</i>    | hHBD-FW     | CAA AGT GAA CGT GGA TGC AG    |
|               | hHBD-RV     | CTG AGA AAA AGT GCC CTT GAG   |
| <i>HBE</i>    | hHBE-FW     | GAA TGT GGA AGA GGC TGG AG    |
|               | hHBE-RV     | GGC TTG AGG TTG TCC ATG TT    |
| <i>HBZ</i>    | hHBZ-FW     | TGA GAG GAC CAT CAT TGT GTC C |
|               | hHBZ-RV     | AAG TGC GGG AAG TAG GTC TTG   |

**Table S2: Antibodies for the Western blot assay used in this study.**

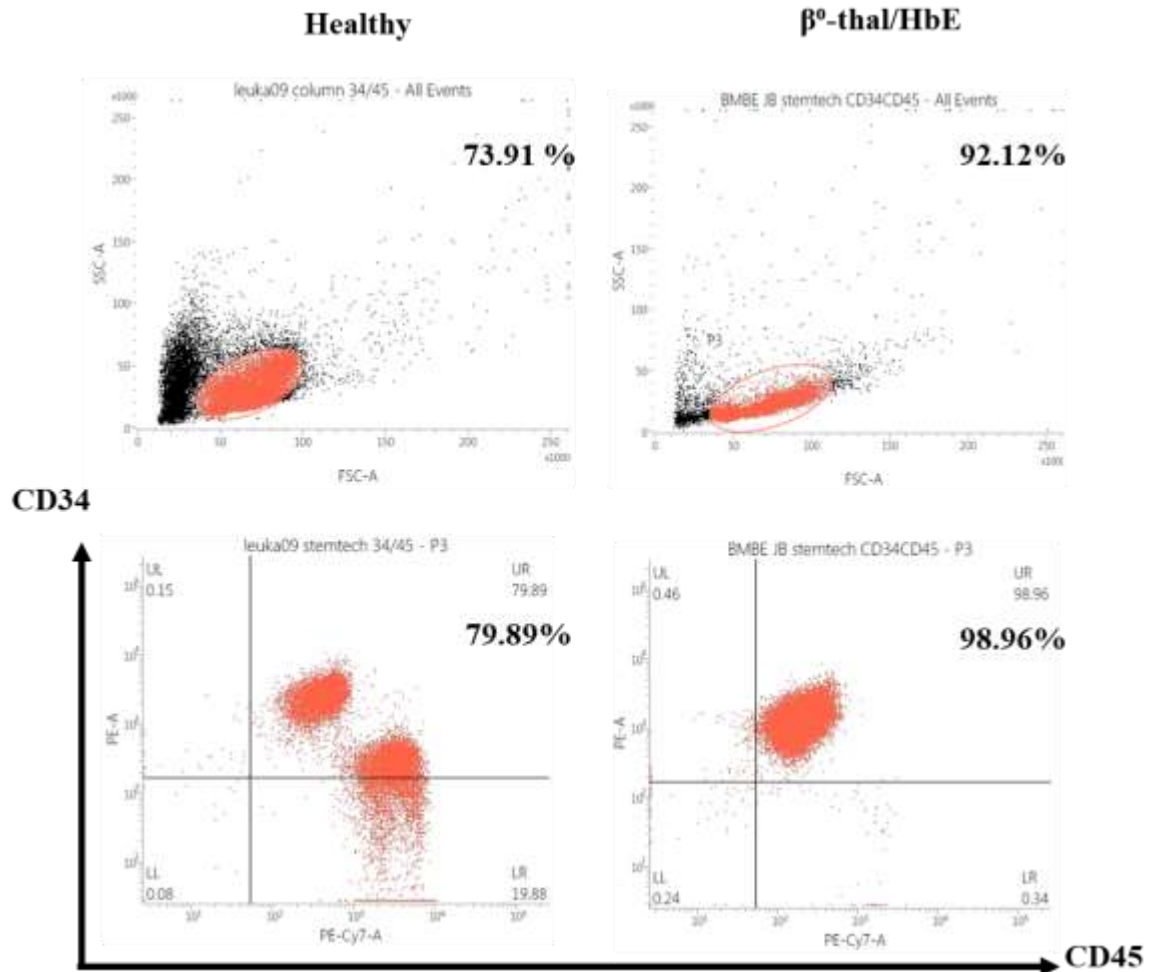
| Antibody              | source              | Manufacturer                 | RRID       | Dilution |
|-----------------------|---------------------|------------------------------|------------|----------|
| Lamin A/C             | Mouse<br>monoclonal | DSHB                         | AB_2618205 | 1:200    |
| SOX6                  | Rabbit polyclonal   | ABclonal                     | AB_2767670 | 1:2,000  |
| LRF/Pokemon           | Rabbit polyclonal   | Cell Signaling<br>Technology | AB_3697515 | 1:2,000  |
| GATA1                 | Rabbit polyclonal   | Abcam                        | AB_298635  | 1:2,000  |
| anti-rabbit IgG (HRP) | Goat polyclonal     | Abcam                        | AB_955447  | 1:10,000 |
| anti-mouse IgG (HRP)  | Horse               | Cell Signaling<br>Technology | AB_330924  | 1:10,000 |



**Figure S1. Position of two shRNAs on the ZBTB7A transcripts.** Four transcript isoforms of *ZBTB7A* are targeted by the shZBTB7A#1 and shZBTB7A#2 at exon 2 and 3' untranslated regions, as highlighted by pink and orange colors, respectively. The graph was acquired on the NCBI website on February 10th, 2025.

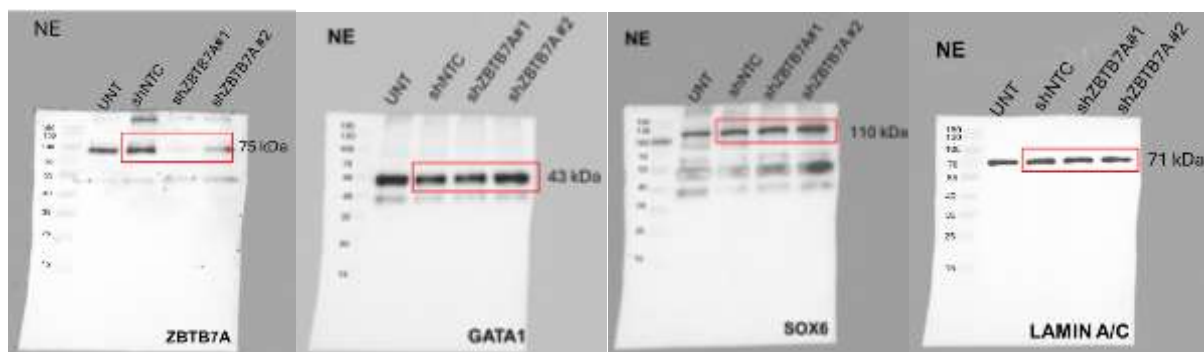


**Figure S2. Effect of *ZBTB7A* downregulation on cell viability.** Percentage of viable cells (live cells/ total cells) at various days after shRNA knockdown in erythroid cells from healthy and  $\beta^0$ -thal/HbE. Error bars represented. The data are mean  $\pm$  SEM of four individuals in each group. \* $p$ -value  $<0.05$ .

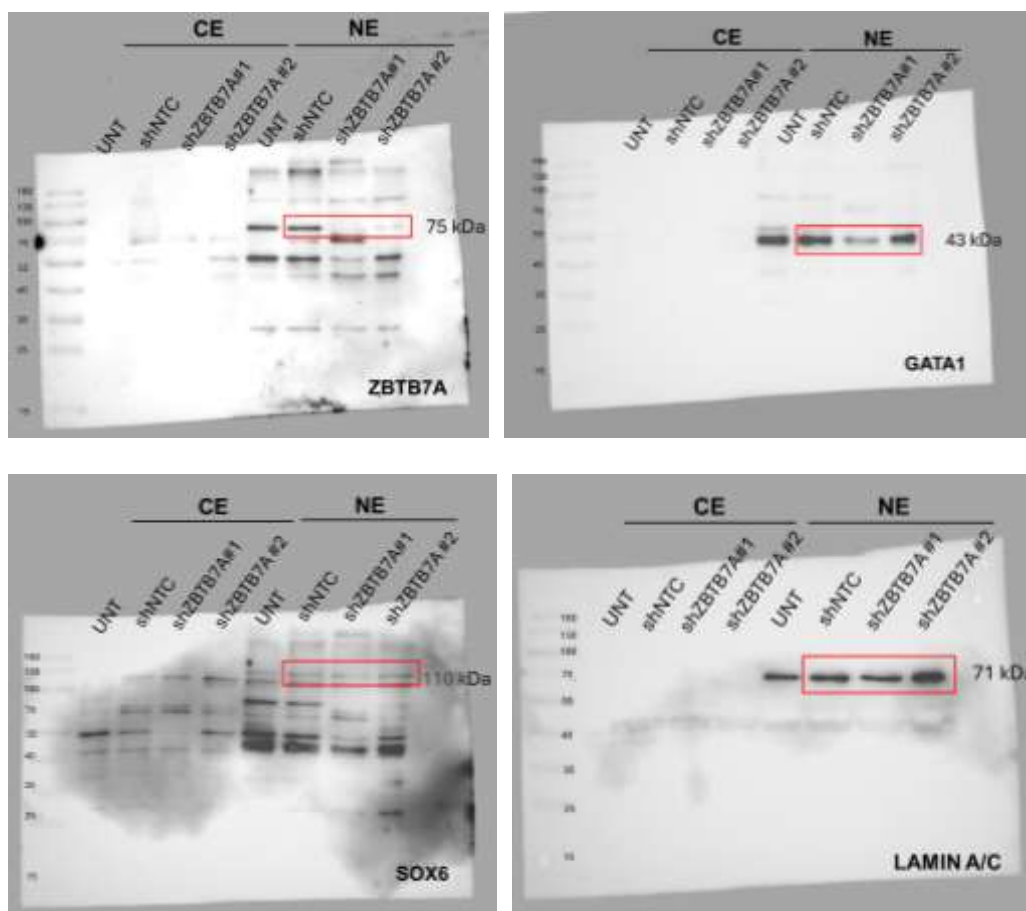


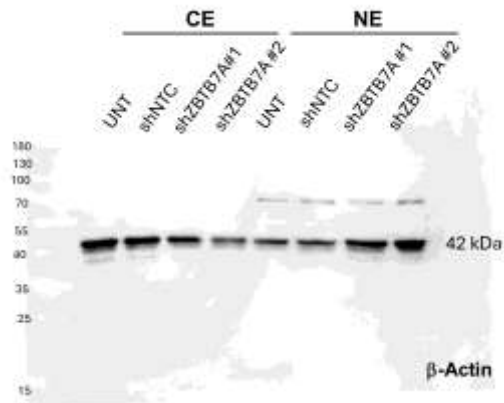
**Figure S3. Representative analysis of the purity and viability of enriched CD34<sup>+</sup> HSPCs from healthy donors and β<sup>0</sup>-thal/HbE patients.** Peripheral blood samples were collected from β<sup>0</sup>-thal/HbE patients, while mobilized peripheral blood was obtained from healthy donors via leukapheresis. CD34<sup>+</sup> HSPCs were enriched using immunomagnetic separation. For β<sup>0</sup>-thal/HbE patient samples, the purity of enriched CD34<sup>+</sup> cells typically exceeded 90%, with cell viability consistently above 90%, as assessed by trypan blue exclusion. In contrast, the purity of CD34<sup>+</sup> cells from mobilized peripheral blood of healthy donors generally exceeded 80%, with viability consistently above 80%.

A



B





**Figure S4. Original Western blot image shows ZBTB7A knockdown.** Protein levels of ZBTB7A, GATA1, and SOX6 were assessed after transduction with UNT, shNTC, shZBTB7A#1, and shZBTB7A#2 in erythroid cells from healthy donors (A) and  $\beta^0$ -thal/HbE patients (B). Lamin A/C was used as a loading control. The red boxes indicate the cropped images used in the Figures.