LETTER TO THE EDITOR

Genome-edited peripheral blood stem cell-derived erythroid progenitor-4 cell line as the unique and beneficial panel cell for antibody identification

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https://doi.org/10.3324/haematol.2024.287187

Supplementary data

Table S1. List of antibodies and staining dyes

Primary antibody	**Reactivity**	**Remarks**
HIRO-3	RhD	Human IgG, produced in-house
HIRO-222	RhC	Human IgM, produced in-house
HIRO-38	Rhe	Human IgM, produced in-house
HIRO-59	P1	Human IgM, produced in-house
Anti-Fy^a serum	Fy^a	Ortho, human IgG
HIRO-133	Jr^a	Human IgG, produced in-house
HIRO-183	Jk^a	Human IgM, produced in-house
Anti-jk^b	Jk^b	Ortho, human IgM
CBC-3	M (GPA)	Mouse IgG, produced in-house
CBC-14	N (GPA)	Mouse IgG, produced in-house
Anti-little s serum	little s (GPB)	Ortho, human IgG
HIRO-58	Di^b	Human IgG, produced in-house
Human IgG	Isotype control	Jackson, isotype ctrl
Human IgM	Isotype control	Thermo, isotype ctrl
Mouse IgG	Isotype control	Jackson, isotype ctrl
Secondary antibody and staining dye	**Reactivity**	**Remarks**
Alexa647 anti-human IgG	human IgG	Jackson
Alexa488 anti-human IgM	human IgM	Jackson
Alexa647 anti-mouse IgG	mouse IgG	Jackson
BRIC6-FITC*	CD233	IBGRL
7-amino actonomycinD (7-AAD)	dead cell	Beckman

*used for viability staining

Table S2. CRISPR/Cas9-mediated mutations in genome-edited PBDEP-4 cell line

Gene	Amino acid replacement	Remarks
RHD	p.Gly61Alafs*37	Frameshift mutation
RHCE	p.Leu60Ilefs*38	Frameshift mutation
ACKR1	p.Gly72_Leu74del p.Val70_Gly72del	In-frame deletion
A4GALT	p.Pro16Glnfs*21	Frameshift mutation
ABCG2	p.Leu28*	Frameshift mutation
SLC14A1	p.Val230Serfs*7	Frameshift mutation
GYPA	p.Leu11Valfs*12	Frameshift mutation
GYPB	p.Leu11Cysfs*4	Frameshift mutation

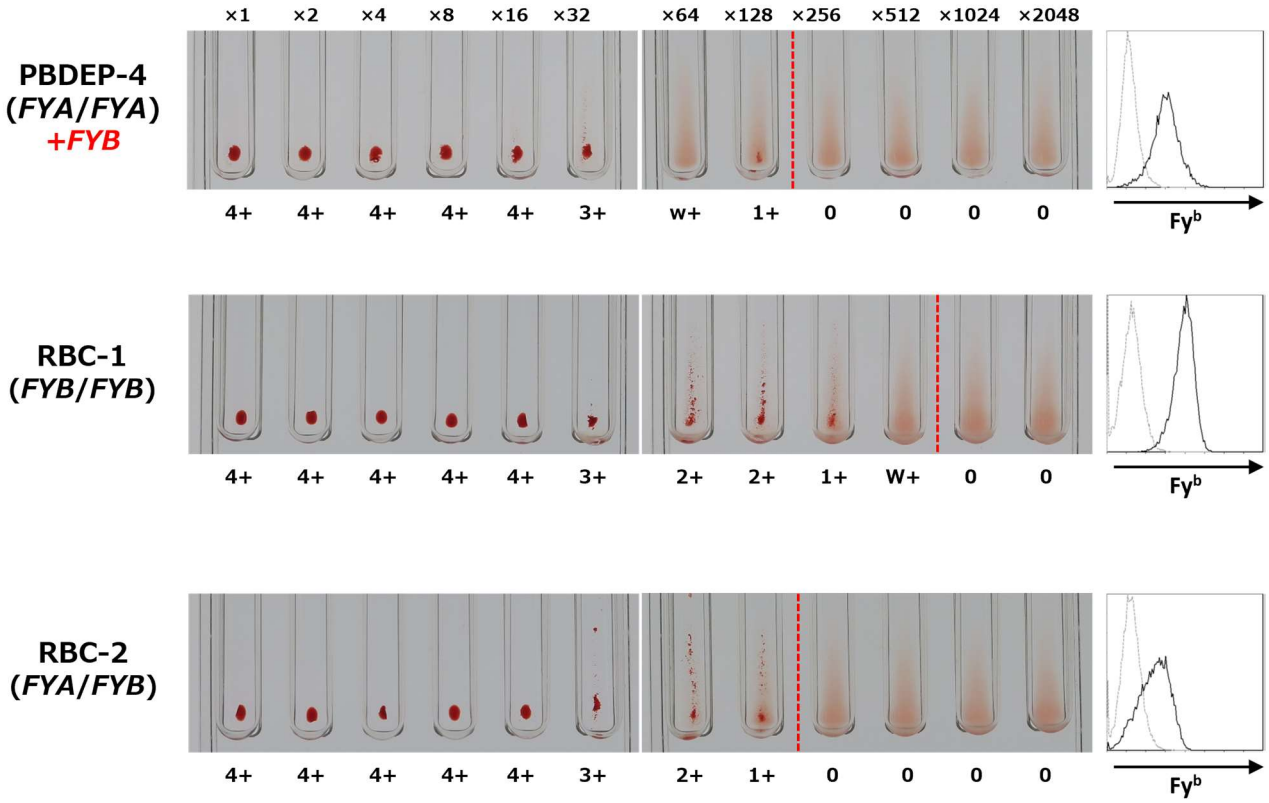

Figure S1. Agglutination assay by tube test using differentiated PBDEP-Fy^b

Agglutination images of differentiated PBDEP-Fy^b cells and RBC with Fy^b (homo and hetero) by tube test using anti-Fy^b (HIRO-299, IgM, produced in-house). A histogram of flow cytometry is adjacent. PBDEP-Fy^b was established by the transfection of the ACKR1 gene with the FYB genotype that was not expressed in parent PBDEP-4 (Fy^a/Fy^a) to evaluate the correlation between antigen expression and the strength of agglutination. RBC-1 and RBC-2 are derived from different donors with Fy^b/Fy^b and Fy^a/Fy^b phenotypes, respectively. The dilution factors of the anti-Fy^b antibodies are presented above the tube image, and the agglutination scores are below each tube image.