## **SUPPLEMENTARY APPENDIX**

Immunophenotypic changes in leukemic blasts in children with relapsed/refractory B-cell precursor acute lymphoblastic leukemia after treatment with CD19-directed chimeric antigen receptor (CAR)-expressing T cells

Ekaterina Mikhailova, Olga Illarionova, Larisa Shelikhova, Elena Zerkalenkova, Olga Molostova, Yulia Olshanskaya, Galina Novichkova, Alexey Maschan, Michael Maschan and Alexander Popov

Dmitry Rogachev National Medical Research Center of Pediatric Hematology, Oncology and Immunology, Moscow, Russian Federation Correspondence:

ALEXANDER M POPOV - uralcytometry@gmail.com doi:10.3324/haematol.2021.279677

 Table S1. Characteristics of patients

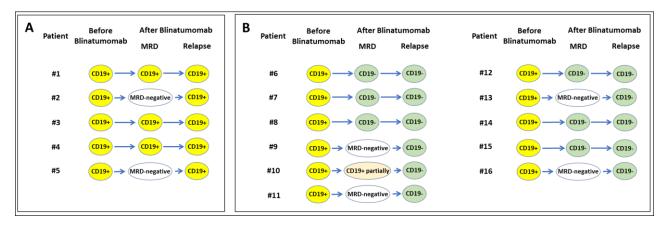
_	20		
n Sov. m/f	39		
Sex, m/f	25/14		
Age Diagnosis	9,0 years (range 0,6 - 20,0 years)		
Diagnosis	5		
BI-ALL	5		
BII-ALL	32		
BIII-ALL	1		
BIV-ALL	1		
Chromosomal aberration	34/37 (91,9%)		
KMT2A rearranged	7		
TCF3 rearranged	4		
t(12;21)(p13;q22)/ETV6-RUNX1	5		
IgH rearranged	3		
CRLF2 rearranged	3		
Complex kariotype	2		
T T T T T T T T T T T T T T T T T T T			
Hyperdiploid	6		
Hypodiploid	2		
	2		
Other aberrations (Intrachromosomal amplification of <i>RUNX1</i> , del9p)	2		
No well-established chromosomal aberrations	3		
No data	2		
Previous therapy			
no HSCT/blinatumomab	15		
blinatumomab only	4		
HSCT only	5		
blinatumomab + HSCT	15		
Blasts in bone marrow before CD19 CAR-T infusion			
< 0,01%	0		
$\geq 0.01\%$ and $< 5\%$ *	14		
≥ 5%**	25		
1. 0.0530/ 0.057 3.3350/	•		

<sup>\*</sup> median 0.952%, range 0.057 – 3.235%; \*\* median 34.122%, range 6.414 – 99.112%.

Table S2. List of antibodies used in the study

Antibody name	Clone	Fluorochrome	Manufacturer	
CD19	010501	APC	— BD	
	SJ25C1	PE-Cy7		
	J3-119	PE-Cy7	BC	
CD10	HI10a	PE		
		BB515	BD	
		BV421		
	ALB1	PE-Cy5.5	BC	
CD34	581	ECD	BC	
	8G12	PE-Cy7		
		APC	BD	
		PE-CF594		
CD20	L27	PerCP	— BD	
		APC-H7		
	B9.E9	APC-Alexa750	BC	
CD45	2D1	APC-Cy7	BD BD	
		PerCP		
	J.33	Krome Orange	BC	
		APC-Alexa750		
CD38	HIT2	APC-R700	BD	
		BV510		
	LS198-4-3	APC-Alexa700	BC	
CD58	AICD58	FITC	BC	
	3C1	FITC	BD	
CD22	S-HCL-1	PE		
		PerCP-Cy5.5	BD	
	HIB22	BV650		
CD24	ML5	BV786	BD	
	ALB9	APC	BC	
CD79a	HM47	APC	BD	

BD – Becton Dickinson, SJ, US; BC – Beckman Coulter, FL, US.



**Figure S1.** Changes in CD19-status of residual leukemic cells at MRD-level and at subsequent relapse in 16 patients with bone marrow relapse occurred. Panel A shows CD19-positive relapses (n=5), panel B – CD19-negative relapses (n= 11). CD19-negativity was defined as less than 20% of tumor cells found to be CD19-positive, CD19-positivity – as more than 75% of leukemic blasts are CD19-positive and CD19+ partially was dimed if number of CD19-positive leukemic blasts was between 20% and 75%.