Immunophenotypic changes of leukemic blasts in children with relapsed/refractory B-cell precursor acute lymphoblastic leukemia who have been treated with blinatumomab

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Table S1. Clinical and genetic characteristics of the studied patients (n=90)

n	90
Sex, m/f	55/35
Age	9,0 years (range 1 - 18 years)
Diagnosis	
BI-ALL	10
BII-ALL	72
BIII-ALL	5
BIV-ALL	2
B-lymphoblastic lymphoma	1
Chromosomal aberration	77/90 (85,6%)
t(12;21)(p13;q22)/*ETV6-RUNX1*	12
KMT2A rearranged	11
Intrachromosomal amplification of *RUNX1*	7
IgH rearranged	6
E2A rearranged	4
CRLF2 rearranged	3
t(9;22)(q34;q11)/*BCR-ABL1*	2
Complex karyotype	12
Hyperdiploid	12
Hypodiploid	3
Others aberrations (*ABL1* rearranged, monosomy 7, *PDGFRbeta* rearranged, trisomy 3)	5
No recurrent chromosomal aberrations	13
Type of therapy	
blinatumomab	23
blinatumomab->HSCT	65
blinatumomab1->HSCT->blinatumomab2	2
Blasts in bone marrow before course of blinatumomab	
<0.001%	8
≥0.001% and <5%	47
≥5%	35

Table S2. List of monoclonal antibodies used for MRD-detection. APC – allophycocyanin, PE – phycoerythrin, Cy7 – cyanin 7, Cy5.5 – Cyanin 5.5, ECD – tandem conjugate of PE with TexasRed, PerCP – peridinin-chlorophyll-protein, FITC –fluorescein isothiocyanate

Antibody	Clone	Fluorochrome	Manufacturer
Obligatory markers			
CD19	SJ25C1	APC	BD
		PE-Cy7	
	J3-119	PE-Cy7	BC
CD10	HI10a	PE	BD
		BB515	
		BV421	
	ALB1	PE-Cy5.5	BC
CD34	581	ECD	BC
	8G12	PE-Cy7	BD
		APC	
		PE-CF594	
CD20	L27	PerCP	BD
		APC-H7	
	B9.E9	APC-Alexa750	BC
CD45	2D1	APC-Cy7	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
Additional markers			
CD22	S-HCL-1	PE	BD
		PerCP-Cy5.5	
	HIB22	BV650	
CD24	ML5	BV786	BD
	ALB9	APC	BC

Table S3. Outcomes in patients who did not relapse, but had leukemic cells on MRD level by MFC in BM at least once during the follow-up period

Patient №	Blast cells in BM, %	CD19 on blast cells, %	Outcomes after MRD reappearance
Patient 1	2,98	0	Chemotherapy -> Allo-HSCT -> Death (sepsis)
Patient 2	0,01	100	MFC-MRD elimination
Patient 3	0,01	100	MFC-MRD elimination
Patient 4	0,29	100	Chemotherapy -> MRD level -> CD19 CAR-T -> MFC-MRD elimination
Patient 5	1,74	5	CD19 CAR-T (CD19+ blasts in CSF) -> Progression
Patient 6	0,51	100	CD19 CAR-T -> CD19- relapse
Patient 7	0,03	100	Allo-HSCT -> MFC-MRD elimination
Patient 8	0,01	100	Allo-HSCT -> MFC-MRD level -> 3 courses of Blinatumomab -> Progression
Patient 9	0,04	20	Chemotherapy -> Allo-HSCT is planned
Patient 10	0,17	100	CD19 CAR-T -> Neurotoxicity (MRD-status unknown)

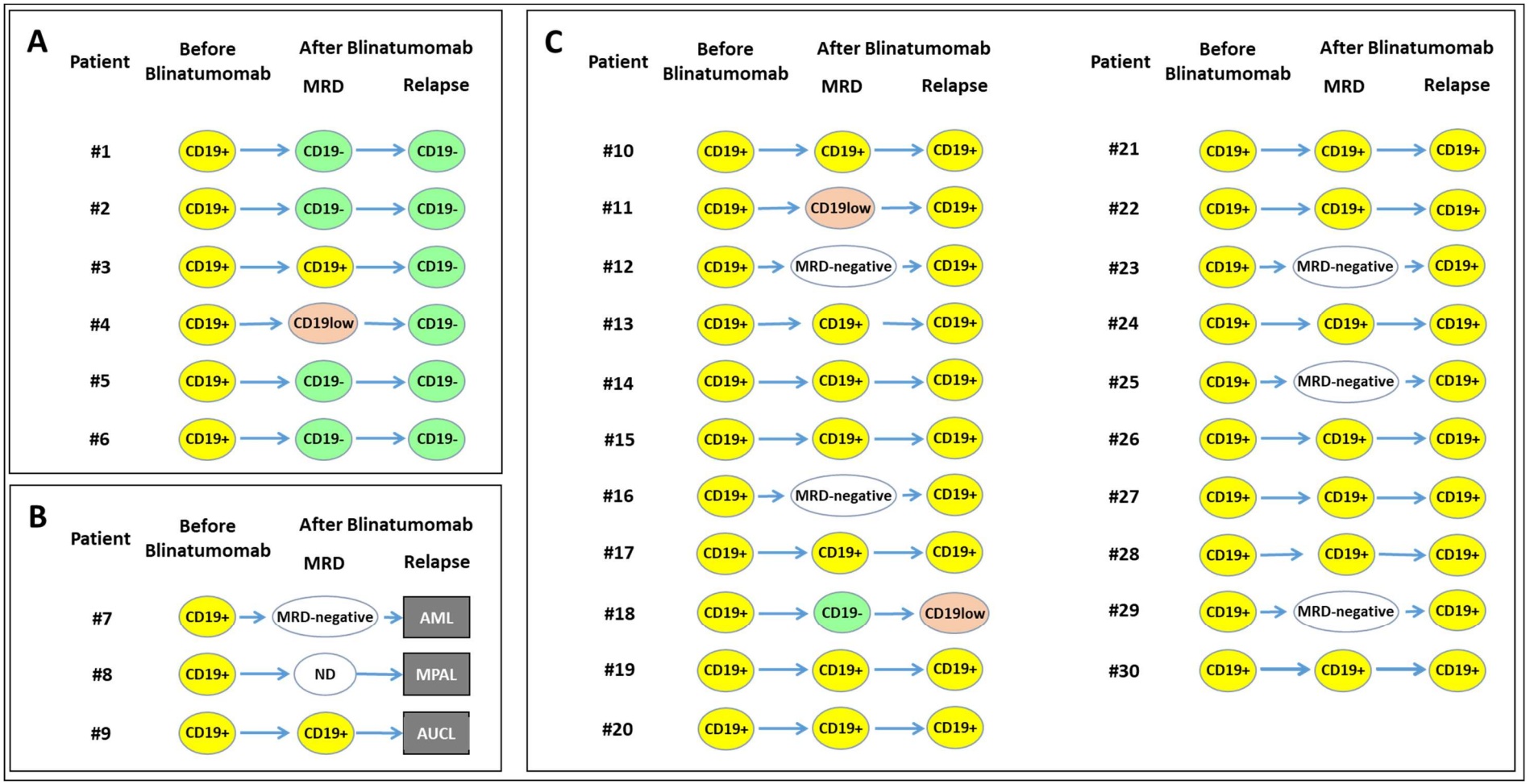

Figure S1. Changes in CD19-status of residual leukemic cells at MRD-level and at subsequent relapse in 30 patients with bone marrow relapse occurred. Panel **A** shows CD19-negative relapses (n=6), panel **B** – those who experienced "lineage switch" to acute myeloid leukemia (AML, pt #7), mixed-phenotype acute leukemia (MPAL, pt #8) and acute unclassifiable leukemia (AUCL, pt #9), while panel C – CD19-positive relapses (n= 21). 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 CD19low was dimed if number of CD19-positive leukemic blasts was between 20% and 75%. ND – no data