Clinical characteristics and prognosis of acute myeloid leukemia associated with DNA-methylation regulatory gene mutations

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Supplemental Table 1. Clinical characteristics of AML patients by cytogenetics

	Cytogenetic risk group					
	Favorable		Intermediate		Adverse	
	n=60	%	n=184	%	n=51	%
Age						
Mean (y/o)	51.0		53.2		58.4	
Range (y/o)	23-80		17-86		17-81	
Sex						
Male	44	73.3%	96	52.2%	32	62.7%
Female	16	26.6%	88	47.8%	19	37.3%
WBC						
Mean (/μl)	19000		69500		51100	
Range (/μl)	900-200300		300-677000		1000-371000	
FAB classification						
M0	0	0.0%	13	7.1%	2	3.9%
M1	3	5.0%	54	29.3%	12	23.5%
M2	43	71.7%	55	29.9%	15	29.4%
M4	2	3.3%	32	17.4%	5	9.8%
M4e	12	20.0%	0	0.0%	0	0.0%
M5	0	0.0%	23	12.5%	9	17.6%
M6	0	0.0%	1	0.5%	6	11.8%
M7	0	0.0%	0	0.0%	1	2.0%
Not determined	0	0.0%	6	3.3%	1	2.0%
Stem cell transplantation						
1st CR	4	6.7%	27	14.7%	5	9.8%
2nd CR	7	11.7%	6	3.3%	0	0.0%
≥3rd CR or on disease	4	6.7%	21	11.4%	9	17.6%

CR: complete remission

Supplemental Table 2. Frequency of gene mutation, and correlation between cytogenetic prognosis and each mutations.

	All		Cytogenetic risk group					
			Favorable		Intermediate		Adverse	
	n=308	%	n=60	%	n=184	%	n=51	%
Gene mutation								
NPM1	88	28.6%	0	0.0%	83	45.1%	2	3.9%
DNMT3A	71	23.1%	1	1.7%	62	33.7%	6	11.8%
FLT3-ITD	65	21.1%	5	8.3%	55	29.9%	3	5.9%
TET2	57	18.5%	2	3.3%	42	22.8%	10	19.6%
NRAS	46	14.9%	11	18.3%	20	10.9%	14	27.5%
RUNX1	38	12.3%	5	8.3%	24	13.0%	8	15.7%
CEBPA	30	9.7%	0	0.0%	26	14.1%	2	3.9%
CEBPA dm	15	4.9%	0	0.0%	14	7.6%	0	0.0%
TP53	28	9.1%	2	3.3%	9	4.9%	13	25.5%
IDH2	28	9.1%	0	0.0%	23	12.5%	3	5.9%
WT1	27	8.8%	2	3.3%	16	8.7%	7	13.7%
KMT2A-PTD	22	7.1%	0	0.0%	16	8.7%	6	11.8%
PTPN11	20	6.5%	0	0.0%	15	8.2%	4	7.8%
FLT3-TKD	19	6.2%	1	1.7%	16	8.7%	2	3.9%
IDH1	17	5.5%	1	1.7%	14	7.6%	1	2.0%
KIT	15	4.9%	12	20.0%	2	1.1%	1	2.0%
GATA2	15	4.9%	0	0.0%	9	4.9%	6	11.8%

Supplemental Table 3. Clinical characteristics of AML patients with DMRGM

	IDH1			*IDH2*			*DNMT3A*			*TET2*		
	n=17	%	*p* value	n=28	%	*p* value	n=71	%	*p* value	n=57	%	*p* value
Age												
Mean (y/o)	59.8		0.1382	58.0		0.1766	55.5		0.4141	63.5		＜0.0001
Range (y/o)	35-73			18-78			21-79			38-79		
Sex												
Male	8	47.1%	0.3232	15	53.6%	0.5538	40	56.3%	0.6809	33	57.9%	0.8827
Female	9	52.9%		13	46.4%		31	43.7%		24	42.1%	
WBC												
Mean (/μl)	46400		0.6294	67200		0.5602	86600		0.0028	80700		0.0389
Range (/μl)	300-194100			600-470500			300-677000			1000-425800		
Cytogenetic Risk group												
Favorable	1	5.9%	0.2032	0	0.0%	0.0199	1	1.4%	＜0.0001	2	3.5%	0.0087
Intermediate	14	82.3%		23	82.1%		62	87.3%		42	73.7%	
Adverse	1	5.9%		3	10.7%		6	8.5%		10	17.5%	
Unknown	1	5.9%		2	7.1%		2	2.8%		3	5.3%	
FAB classification												
M0	0	0.0%	0.6103	3	10.7%	0.1926	4	5.6%	1.0000	1	1.8%	0.2144
M1	6	35.3%	0.3810	11	39.3%	0.0649	16	22.5%	0.7541	22	38.6%	0.0096
M2	9	52.9%	0.2005	5	17.9%	0.0253	16	22.5%	0.0033	17	29.8%	0.2261
M4	1	5.9%	0.7081	6	21.4%	0.2314	15	21.1%	0.0265	5	8.8%	0.3846
M4e	0	0.0%	1.0000	0	0.0%	0.6107	0	0.0%	0.0751	0	0.0%	0.1324
M5	1	5.9%	1.0000	2	7.1%	0.7515	15	21.1%	0.0034	10	17.5%	0.0931
M6	0	0.0%	1.0000	0	0.0%	1.0000	2	2.8%	0.6634	0	0.0%	0.3560
M7	0	0.0%	1.0000	0	0.0%	1.0000	1	1.4%	0.2305	0	0.0%	1.0000
not determined	0	0.0%	1.0000	1	3.6%	0.5378	2	2.8%	1.0000	2	3.5%	0.6440
Stem cell transplantation												
1st CR	0	0.0%	0.2396	3	10.7%	1.0000	10	14.1%	0.5363	5	8.6%	0.5029
2nd CR	1	5.9%	0.5292	1	3.6%	1.0000	2	2.8%	0.7395	0	0.0%	0.1368
≥3rd CR or on disease	2	11.8%	1.0000	2	7.1%	0.7559	8	11.3%	1.0000	4	6.9%	0.2624

CR: complete remission

Supplemental Table 4. Relationship among the each mutations with EMRGM

Favorable risk	60	1	0.2458	0.2107	0	0.0639	0.0023	1	0.0431	＜0.0001	2	0.1210	0.0003	4	0.0638	＜0.0001
Intermediate risk	184	14	3.3220	0.0727	23	3.4000	0.0141	62	6.4940	＜0.0001	42	2.1490	0.0178	109	5.4780	＜0.0001
Adverse risk	51	1	0.3013	0.3241	3	0.5800	0.5930	6	0.3938	0.0442	10	1.0900	0.8441	16	0.5301	0.0633
NPM1	88	8	2.3440	0.0988	12	2.0130	0.1222	44	7.1480	＜0.0001	26	2.5570	0.0031	72	11.2100	＜0.0001
DNMT3A	71	7	2.4830	0.0788	15	4.6150	0.0002				16	1.3910	0.3834			
FLT3-ITD	65	2	0.4825	0.5406	6	1.0220	1.0000	26	2.9330	0.0005	18	2.0030	0.0465	41	2.7080	0.0007
TET2	57	0	0.1165	0.0497	1	0.1481	0.0380	16	1.3910	0.3834						
NRAS	46	1	0.3417	0.4843	4	0.9444	1.0000	5	0.3622	0.0364	3	0.2687	0.0229	11	0.3498	0.0035
RUNX1	38	3	1.5670	0.4508	4	1.2060	0.7623	6	0.5913	0.3082	6	0.8051	0.8240	14	0.7183	0.3870
CEBPA	30	2	1.2520	0.6755	1	0.3206	0.4983	3	0.3431	0.1076	10	2.4570	0.0441	13	0.9778	1.0000
CEBPA dm	15	1	1.2370	0.5820	0	0.3005	0.3768	0	0.1004	0.0265	2	0.6657	1.0000	3	0.3049	0.0651
TP53	28	1	0.6111	1.0000	3	1.2240	0.7295	8	1.3780	0.4827	3	0.5022	0.3195	9	0.5789	0.2326
IDH2	28	1	0.6111	1.0000				15	4.6150	0.0002	1	0.1481	0.0380			
WT1	27	1	0.6370	1.0000	1	0.3618	0.4889	2	0.2458	0.0535	3	0.5255	0.4371	6	0.3367	0.0241
KMT2A-PTD	22	2	1.8070	0.3468	1	0.4568	0.7056	7	1.6190	0.3021	7	2.2030	0.1481	13	1.9420	0.1804
PTPN11	20	2	2.0220	0.3039	2	1.1200	0.7010	9	2.9820	0.0254	5	1.5130	0.3878	14	3.2200	0.0190
FLT3-TKD	19	1	0.9479	1.0000	2	1.1900	0.6867	9	3.2950	0.0200	4	1.1870	0.7617	12	2.3140	0.0964
IDH1	17				1	0.6111	1.0000	7	2.4830	0.0788	0	0.1165	0.0497			
KIT	15	0	0.5097	1.0000	0	0.3005	0.3768	1	0.2276	0.2054	3	1.1060	0.7454	3	0.3049	0.0651
GATA2	15	1	1.2370	0.5820	0	0.3005	0.3768	2	0.4994	0.5336	1	0.3023	0.3200	4	0.4970	0.1932
		number	odds ratio	*p* value	number	odds ratio	*p* value	number	odds ratio	*p* value	number	odds ratio	*p* value	number	odds ratio	*p* value
		IDH1 n=17			*IDH2* n=28			*DNMT3A* n=71			*TET2* n=57			DMRGM n=135		

Red color means that the each mutations are coexistant strongly. p≤0.0500
Pink color means that the each mutations are coexistant. 0.0500＜p≤0.1000
Light blue color means that the each mutations are exclusive. 0.0500＜p≤0.1000
Blue color means that the each mutations are exclusive strongly. p≤0.0500

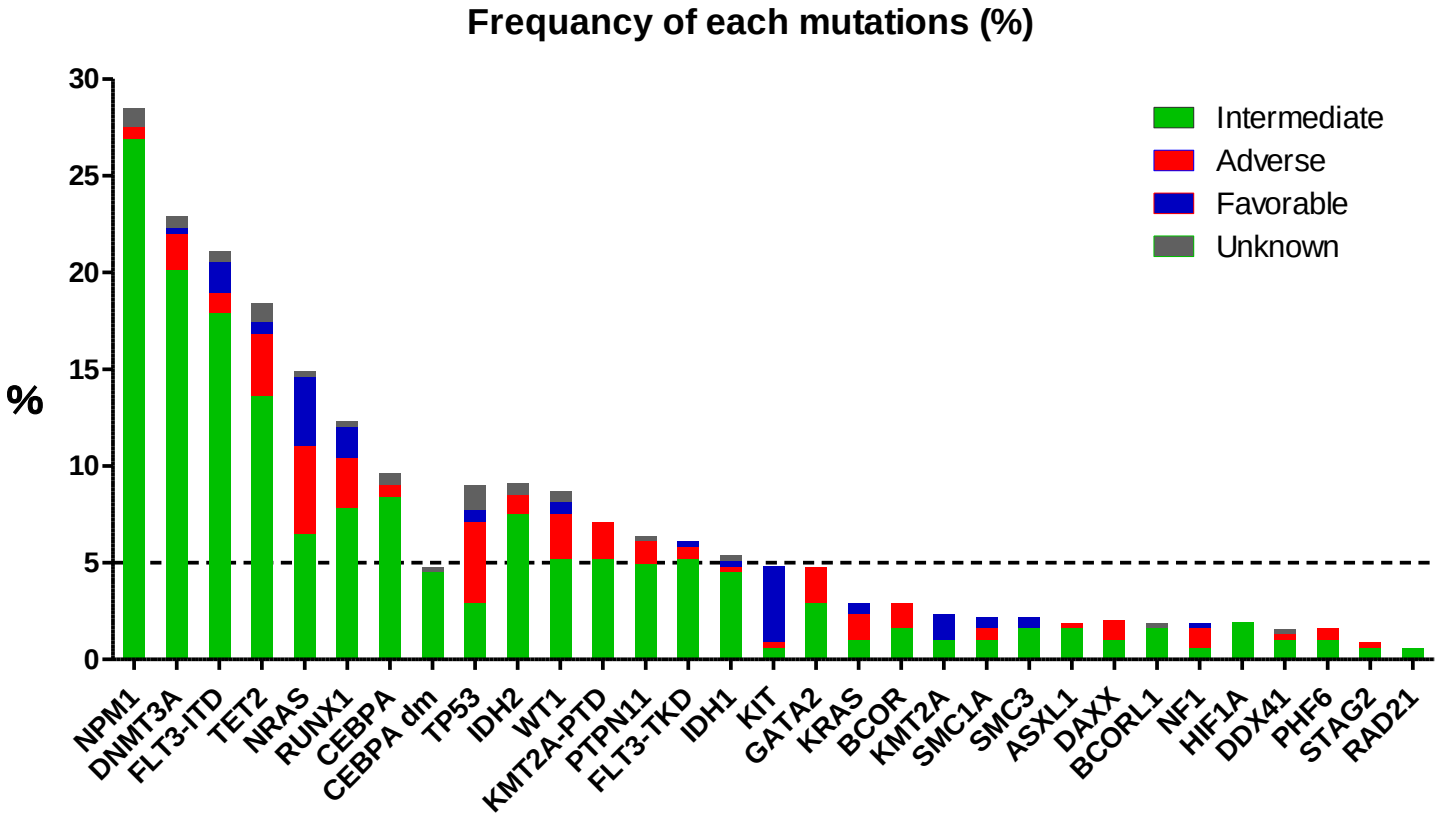
Supplemental Table 5. Prognostic factor analysis based on univariate analysis

	All patients				Age≤70, Intermediate cytogenetic risk and *FLT3*-ITD negative			
	5y OS	*p* value	5y RFS	*p* value	5y OS	*p* value	5y RFS	*p* value
All Case	34.8%	-	32.0%	-	38.8%	-	41.3%	-
Age ＞ 70 years	0.0%	＜0.0001	20.4%	0.0667	-	-	-	-
Favorable cytogenetic risk	64.0%		42.1%		-	-	-	-
Intermediate cytogenetic risk	30.5%	＜0.0001	31.9%	0.0210	-	-	-	-
Adverse cytogenetic risk	12.7%		10.2%		-	-	-	-
SCT(1stCR or 2ndCR)	71.7%	＜0.0001	-	-	67.7%	0.0283	-	-
SCT(1stCR)	66.3%	0.0005	73.8%	< 0.0001	69.8%	0.0459	80.0%	0.0008
DMRGM1-3	23.2%	0.0018	23.8%	0.1823	31.7%	0.0409	40.2%	0.8649
DMRGM2-3	19.5%	< 0.0001	20.9%	0.0244	32.7%	0.0189	23.6%	0.2041
NPM1	27.0%	0.2312	24.9%	0.1791	36.5%	0.6146	34.4%	0.7447
DNMT3A	27.0%	0.0017	28.5%	0.2300	35.2%	0.0395	38.4%	0.7062
FLT3-ITD	17.5%	＜0.0001	13.7%	＜0.0001	-	-	-	-
TET2	14.0%	0.0043	9.2%	0.0219	32.6%	0.7231	28.7%	0.8518
NRAS	42.4%	0.1775	62.5%	0.0142	43.2%	0.8808	85.7%	0.0674
RUNX1	15.9%	0.7986	19.7%	0.8665	0.0%	0.7612	66.7%	0.7315
CEBPA	40.8%	0.2263	44.3%	0.1652	49.2%	0.1780	55.8%	0.1404
CEBPA dm	66.8%	0.0282	61.2%	0.1081	62.3%	0.1314	66.8%	0.1786
TP53	8.7%	＜0.0001	0.0%	0.0011	28.6%	0.0957	0.0%	0.0068
IDH2	37.1%	0.4283	41.1%	0.8313	38.5%	0.0883	51.9%	0.9592
WT1	29.1%	0.8186	24.8%	0.4709	20.0%	0.3081	20.0%	0.0836
KMT2A-PTD	13.4%	0.0152	22.6%	0.0781	53.3%	0.7502	62.5%	0.6587
PTPN11	53.3%	0.5177	46.2%	0.1549	56.3%	0.7052	56.3%	0.2974
FLT3-TKD	24.6%	0.6154	36.7%	0.9242	55.6%	0.8011	42.9%	0.5859
IDH1	0.0%	0.3013	0.0%	0.4301	0.0%	0.3167	22.2%	0.4379
KIT	64.2%	0.1289	22.3%	0.8116	100.0%	0.3036	0.0%	0.8974
GATA2	31.9%	0.6784	26.8%	0.4802	66.7%	0.5078	80.0%	0.6117

5y: 5 years, OS: overall survival, RFS: relapse free survival, SCT: stem cell transplantation, CR: complete remission

Supplemental Figure 1. Frequency of analyzed genetic mutations in the de novo AML cohort.

Blue represents favorable cytogenetic risk, green represents intermediate cytogenetic risk, and red represents poor cytogenetic risk. Many of the mutations were detected in the intermediate cytogenetic risk group. Gene mutations with an incidence of 3% or less were excluded from further analysis as they were not sufficiently frequent to be studied as prognostic factors.



Supplemental Figure 2. DMRGM combination and prognosis

A: Overall survival rate for all cases. B: Relapse-free survival rate for all cases.
HR: Hazard ratio.