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Prognostic value of genetic mutations in adolescent and young adults with acute myeloid leukemia

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Original Article

Prognostic value of genetic mutations in adolescent and young adults with acute myeloid leukemia

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Abstract Clinical outcomes and the genetic background of acute myeloid leukemia (AML) in adolescent and young adults (AYAs) are known to differ in younger children and older adults. To clarify the impact of genetic mutations on clinical outcomes of AYAs with AML, we analyzed data from the JPLSG AML-05 and JALSG AML201 studies. AYAs aged 15 to 39 years (*n*=103) were included. *FLT3*-ITD, *KIT*, *CEBPA*, *NRAS*, *KRAS*, *WT1*, *MLL*-PTD, and *NPM1* mutations were analyzed. Overall survival (OS) of the AYAs was 61% and event-free survival was 38% at three years. *FLT3*-ITD (HR, 2.10; 95% CI, 1.07 to 4.12; *P*=0.031) and *NPM1* (HR, 0.24; 95% CI, 0.06 to 1.00; *P*=0.050) mutations were associated with risk of overall mortality in multivariate analysis. OS was significantly different according to *FLT3*-ITD and *NPM1* mutation status (*P*=0.03). Survival was 100% with *NPM1* mutations in the absence of *FLT3*-ITD and 35% (95% CI, 14-57%) with *FLT3*-ITD in the absence of *NPM1* mutations. The OS of AYAs, children (*n*=413) and older adults (*n*=124) of the AML-05 and AML201 participants were significantly different (*P*<0.0001). This is the first report to combine clinical and genetic data of AYA AML from the major Japanese pediatric and adult study groups.

Keywords

AML; AYA; genetic mutation; prognosis

Introduction

Clinical outcomes of acute myeloid leukemia (AML) are different between younger children, adolescent and young adults (AYAs), and older adults, although outcomes have improved among all the age groups during the past decades. Results of AYAs with AML in pediatric studies or pediatric and adult intergroup studies have demonstrated similar to inferior overall survival, similar to decreased relapse, and increased treatment-related mortality in the AYAs compared with children. [1-4] Pediatric protocols generally consist of intensified regimens compared with adult protocols, but it is still controversial whether an intensified pediatric AML regimen results in better survival for AYA populations.

AML is currently considered as a biologically heterogeneous disease. In addition to conventional cytogenetic abnormalities, somatic mutations have proven to be significantly prognostic in AML. Such molecular markers are becoming increasingly important as a mean of risk stratification for both children and adults with AML. However, the prevalence of each somatic mutation is known to be different between pediatric and adult populations. [5-9] Previously, the Japan Adult Leukemia Study Group (JALSG) comprehensively analyzed mutations in 51 genes among 197 adult patients with *de novo* AML who were registered in the JALSG AML 201 study. *FLT3*, *NPM1*, *CEBPA*, *DNMT3A*, and *KIT* were mutated in more than 10% of the patients aged 15 to 64 years old. Furthermore, *DNMT3A*, *MLL*-PTD, and *TP53* gene mutations clearly stratified adult AML patients into five distinct prognostic subgroups when combined with the European LeukemiaNet risk classification. [10] As for children, the Japanese Pediatric Leukemia/Lymphoma Study Group (JPLSG) analyzed somatic mutations in *de novo* AML patients who were registered in the JPLSG AML-05 study. These analyses have confirmed mutations of *KIT* in CBF-AML, *CXCR4* in low-risk AML, *CEBPA*-double, and *PRDM16* were independent prognostic factors for children with AML, respectively. [11-14]

Thus, outcomes of both children and adults with AML have been reported in relation to genetic mutations in the previous studies; however, reports on prevalence and prognostic impact of AML genetic mutations in AYAs are limited. Therefore, we analyzed data of clinical information and molecular markers using datasets of the pediatric and adult prospective trials in this study.

Methods

Patients and data

The AYA population included newly diagnosed *de novo* AML patients, aged 15 to 39 years, who were subjected to genetic alteration analyses. The patients were prospectively registered either in the JALSG AML201 study (UMIN Clinical Trials Registry [UMIN-CTR], http://www.umin.ac.jp/ctr/index.htm, number C000000157) or the JPLSG AML-05 study (UMIN-CTR, number UMIN000000511). Both AML protocols are summarized in Table S1. In detail, the JALSG AML201 study was a multi-center phase 3 randomized study, comparing high-dose daunorubicin with idarubicin in induction therapy followed by comparison of high-dose cytarabine with conventional sequential chemotherapy in post-remission therapy. Adult patients age 15 to 64 years old were registered from December 2001 to December 2005. [15, 16] The trial included 1057 patients, of whom 197 patients were subjected to comprehensive mutation analysis of the 51 genes. [10] Of these patients, 73 AYAs aged 15 to 39 years old were analyzed in this study. Data of the remaining 124 patients who were 40 years old or older were also used for comparison analysis between the AYAs and the older adults. The JPLSG AML-05 study was a Japanese nationwide multi-institutional phase 2 study for newly diagnosed pediatric AML patients, which evaluated the efficacy and safety of the risk-stratified therapy based on karyotype and/or *FLT3*-ITD status, as well as response to initial induction therapy. [17] Four hundred and forty-three children aged 18 years old or younger were registered from November 2006 to December 2010.

The cohort included 30 patients aged 15 to 18 years old. The remaining 413 patients who were younger than 15 years old were also used for comparison analysis with AYAs.

Screening for mutations were performed using high molecular weight DNA and total RNA that were extracted from either peripheral blood or bone marrow as previously described. [10-13] In our study, clinical information and mutation status of nine genes (*FLT3*-ITD, *KIT*, *CEBPA*, *NRAS*, *KRAS*, *WT1*, *MLL*-PTD, and *NPM1*) for 103 AYA patients from the combined dataset of the AML201 and AML-05 studies were analyzed. The institutional review board of Nagoya University Graduate School of Medicine approved this study.

Statistical analysis

Differences in baseline patient characteristics were compared using the Fisher's exact test or the Chi-square test for categorical variables and Kruskal-Wallis test or the Wilcoxon rank-sum test for continuous variables. Overall survival (OS) was defined as the time from the date of the trial registration until the date of death due to any cause or to the last date of follow-up. Event-free survival (EFS) was defined as the time from the trial registration to the last follow-up or event (failure to achieve remission, relapse, or death from any cause). Patients who did not achieve complete remission (CR) during induction therapy were treated as failure on the date of registration. The probability of OS and EFS was estimated according to the Kaplan–Meier method, and the groups were compared using the log-rank test. Cumulative incidence of relapse was defined as time from achieving CR to relapse; death from any cause before relapse was a competing risk. Cumulative incidence of treatment-related mortality (TRM) was defined as time from registration to death due to non-progressive disease; failure to achieve remission or relapse was a competing risk for TRM. Cumulative incidences of

the groups were compared using Gray's method. Analyses to identify risk factors for achieving CR were done with logistic regression. To compare the OS and EFS, Cox proportional hazard models were used to assess prognostic significance of the genetic mutations. [18, 19] All variables met the proportionality assumption for the Cox model. Variables with a P-value of <0.1 on univariate analysis were entered into a multivariate model using a backward selection method with a threshold P-value of less than 0.05. Interactions between the covariates in the final model were tested and there were none. Results are expressed as hazard ratio (HR) together with the 95% confidence intervals. SAS version 9.4 (SAS Institute Inc, Cary, NC), Stata version 13.1 (StataCorp, TX) and EZR statistical software (Saitama Medical Center, Jichi Medical University, Saitama, Japan) [20] were used in the analyses.

Results

Characteristics of patients

Table 1 shows characteristics of the AYAs and their disease by age subgroups. Median age of the AYAs was 23 (range, 15-39) years old. Patients were classified into favorable- (n=33, 32%), intermediate- (n=63, 61%), and adverse-risk groups (n=7, 7%) according to the refined MRC criteria [21]. There were no differences between the two age categories (15-24 versus [vs.] 25-39 years) in the background characteristics such as white blood cell (WBC) count, cytogenetic risk groups, and FAB classification. Sixty-four patients (62%) received allogeneic hematopoietic stem cell transplantation (allo-HSCT); patients who were 25 years or older more often received stem cell transplantation (n=36, 72%) compared with the patients under 25 years old (n=28, 53%, p=0.045). Comparing the clinical background of younger children, the AYAs and the older adults, the children were more likely to have poor cytogenetics and M7 or MDS related changes. AYAs were more likely to receive allo-HSCT than children or the older adults (Table S2).

Outcomes and clinical risk factors

Overall, 84 of the 103 AYAs (82%) achieved CR. Univariate logistic analysis demonstrated that poor risk cytogenetics was an unfavorable factor for achieving CR compared with good risk cytogenetics (Odds Ratio [OR], 0.10; 95% CI 0.02 to 0.64; p=0.015). The M5 FAB classification tended to be an unfavorable factor for achieving CR compared with M2 classification (OR, 0.26; 95% CI 0.06 to 1.14; p=0.075); however, only the cytogenetic risk remained significant in the multivariate analysis. The risk of failure to achieve CR among AYAs was not different among the two age categories (15-24 vs. 25-39; OR, 0.63; 95% CI 0.23 to 1.72; p=0.37), WBC at diagnosis (≤20,000 vs. >20,000; OR, 0.78; 95% CI 0.29 to 2.11; p=0.63), or treatment groups (AML-05 vs. AML201; OR, 2.53; 95% CI 0.68 to 9.41; p=0.17).

The estimated OS for the AYA population was 61% (95% CI, 51% to 70%) at 3 years (Figure S1A). There was no difference in survival among the two age groups for OS (p=0.77) (Figure 1A). OS was significantly different according to the WBC at diagnosis (p=0.001) and the cytogenetic risk groups (p=0.004) (Figure 1B and 1C). EFS for AYAs was 38% (95% CI, 28% to 47%) at 3 years (Figure S1B). There was also no difference in EFS among the two AYA age groups (p=0.30) (Figure S2A); EFS was significantly different according to the WBC at diagnosis (p=0.008) and the cytogenetic risk groups (p=0.019). (Figure S2B and S2C). The OS of teenage patients from ages 15 to 19 was not different between AML201 and AML-05 study participants (67% [95% CI, 19% to 90%; n=6] vs. 61% [40% to 76%; n=30] at 3 years, respectively; p=0.65). EFS of teenage patients was also not different between the AML201 and AML-05 participants (33% [95% CI, 5% to 68%] vs. 53% [34% to 69%] at 3 years, respectively; p=0.46).

Cumulative incidence of relapse for the AYA patients who achieved CR following induction chemotherapy was 48% (95% CI, 37% to 59%) at 3 years (Figure S1C).

Cumulative incidence of TRM for AYAs was 5% (95% CI, 2% to 10%) at 3 years (Figure S1D). Cumulative incidence of relapse for teenage patients from age 15 to 19 was not different between AML201 and AML-05 study participants (50% [95% CI, 7% to 84%] vs. 35% [17% to 53%] at 3 years, respectively; p=0.52); TRM was also not different between the two protocols (17% [95% CI, 0.4% to 56%] vs. 7% [1% to 20%] at 3 years, respectively; p=0.43). Most deaths among the AYAs were followed by either relapse or non-CR after induction therapy. Of the 44 AYA patients who died, 14 patients (32%) were non-CR after induction therapies, 25 patients (57%) died after relapse, and 5 patients (11%) died in CR. All deaths among the older age group of AYAs (25 to 39 years, n=23) occurred after non-CR after induction therapy (n=8) or after relapse (n=15).

We further compared the survival results of the AYAs (n=103) with the younger children who were AML-05 participants under 15 years old (n=413) and the older adult AML201 participants who were 40 years or older (n=124) (Figure S1A-D). The OS of the children, AYAs and the older adults was significantly different (75% [95% CI, 70% to 79%], 61% [51% to 70%], and 52% [43% to 60%] at 3 years, p<0.0001). EFS of the children, AYAs and older adults was also different (54% [95% CI, 49% to 59%], 38% [28% to 47%] and 28% [20% to 36%], p<0.0001). Cumulative incidence of relapse was different between children, AYAs and older adults (37% [95% CI, 32% to 42%], 48% [37% to 59%] and 63% [53% to 72%], p<0.0001), whereas TRM was not different between the three different generations (1% [95% CI, 1% to 5%], 2% [2% to 10%] and 2% [1% to 8%], p=0.44).

Frequencies of mutations

FLT3-ITD, *KIT*, *CEBPA*, and *NPM1 were* mutated in more than 10% of the AYAs (Table 2). Although some of the information on *KIT* and *CEBPA* mutations was more likely to be missing in the patients aged under 24 years old, there were no significant

differences in the background characteristics of the mutation status in the AYAs among the two age groups. Overlapping mutations were frequently observed, especially for *WT1*, with *FLT3*-ITD (38%), *CEBPA* (25%) and *NRAS* (25%); for *NPM1*, with *FLT3*-ITD (36%); for *NRAS*, with *CEBPA* (29%) and *WT1* (29%); for *FLT3*-ITD, with *NPM1* (23%) (Figure 2). Comparing children, AYAs and older adults, *FLT3*-ITD was more frequently mutated in AYAs and older adults (p=0.004), *KIT* in children (p<0.001), *KRAS* in AYAs (p=0.03) and *MLL*-PTD was more frequently mutated in the older adults (p=0.001, Table S3).

Genetic mutations and prognosis

By Fisher's exact test, *FLT3*-ITD was identified as an unfavorable factor for achieving CR (64% vs. 86% for mutation positive and negative, p=0.03); *NPM1* mutation had a high rate of achieving CR, but the difference did not reach significance (100% vs. 78% for mutation-positive and -negative, p=0.07). Univariate logistic analysis showed that only *FLT3*-ITD was an unfavorable factor for achieving CR (Odds Ratio, 0.28; 95% CI 0.09 to 0.81; p=0.019).

We analyzed the association of each mutation with the risk of mortality. *FLT3*-ITD (p=0.084) and *CEBPA* double mutation (p=0.065) were associated with increased risk of overall mortality, while *NPM1* (p=0.072) was associated with decreased risk by univariate analysis. *FLT3*-ITD (HR, 2.10; 95% CI, 1.07 to 4.12; p=0.031) remained significantly prognostic in multivariate analysis, while marginal significance was observed for *NPM1* (HR, 0.24; 95% CI, 0.06 to 1.00; p=0.050) (Table 3). As for EFS, *FLT3*-ITD (p=0.029) and *NPM1* (p=0.019) were also associated with increased and decreased risk of overall mortality by univariate analysis, respectively. In multivariate analysis, both *FLT3*-ITD (HR, 2.37; 95% CI, 1.34 to 4.21; p=0.003) and *NPM1* (HR, 0.24; 95% CI, 0.09 to 0.67; p=0.006) remained significantly prognostic (Table 3).

Figure 3 shows the survival curves according to the *FLT3*-ITD and *NPM1* mutation status. OS was significantly different according to the *FLT3*-ITD and *NPM1* mutation status (p=0.03). A favorable 3-year OS was observed for the patients with *NPM1* mutations in the absence of *FLT3*-ITD (100%) compared with the patients with *FLT3*-ITD in the absence of *NPM1* mutations (35%; 95% CI, 14-57%). EFS was also different according to the combined mutation status of *FLT3*-ITD and *NPM1* (p=0.0006), with 78% (95% CI, 36-94%) for the patients with *NPM1* mutations in the absence of *FLT3*-ITD and 11% (95% CI, 2-31%) for the patients with *FLT3*-ITD in the absence of *NPM1* mutations at 3 years.

Discussion

This intergroup study sought to identify the genetic influence on clinical outcomes in AYAs with AML, using data of clinical information and somatic mutations from the pediatric and adult prospective trials. The current analysis demonstrated that *FLT3*-ITD was associated with increased risk of mortality and *NPM1* with decreased risk of mortality for AYA AML. *FLT3*-ITD and *NMP1* are known to be important prognostic factors for AML in both children and adults. Although there are other mutations well recognized in either children or adults with AML, such as *DNMT3A* and *NUP98-NSD1*, only nine genes that had been commonly analyzed in the pediatric and adult protocols (i.e. *FLT3*-ITD, *KIT, CEBPA, NRAS, KRAS, WT1, MLL*-PTD and *NPM1*) were analyzed in this study. Age-related leukemia-associated mutational change was previously reported among blood normal controls of patients with cancer [22]; it is suggested that the mutational background of the younger adults is different from older adults. Further intergroup analyses are warranted for more detailed understanding of the AYA population in regard to mutation and prognosis [10, 23], as the improvement of outcomes with individualized molecular targeted therapy is also anticipated.

In this study, we found no difference in OS or EFS between AYA patients aged 15 to 19 years according to pediatric versus adult protocols, although there were few patients and these analyses lacked the power to test the comparisons. Pediatric regimens are generally more intensified than those of adult AML; however, it is still controversial if intensified pediatric regimens are also beneficial for AYAs with AML. It is still difficult to conclude from our study if the intensification of the regimen is preferable for teenage patients because only 6 of 36 teenage patients received treatment with the adult protocol, and all the patients who were 20 years old or older were participants in the adult trial. Woods et al. previously compared the results of the pediatric COG trials to the adult CALGB and SWOG trials, and reported that AYAs treated with pediatric protocols had better outcomes compared with those who were treated with adult protocols. However, age was also a major confounding factor of that study, and it was not clear whether the younger age for pediatric protocol participants or the regimen itself resulted in the better results of the pediatric protocol. [24] In contrast, a German intergroup study investigated outcomes of children and young adults under 30 years old, and they did not find any difference between the pediatric and adult trials in the same age group. [1] Another previous study compared the results of AYA AML treated with the pediatric protocol of the Nordic Society of Paediatric Haematology and Oncology with the patients treated in hematology departments in the Nordic countries. This study did not find any differences in outcomes of OS and EFS according to adult or pediatric protocols. [25] It is noted that the AML-05 protocol of the JPLSG used intensive central nervous system prophylaxis, which is a standard for pediatric protocols. In the contrary, adult AML201 protocol participants, who were randomized to 3 courses of high-dose cytarabine consolidation therapy, received no intrathecal chemotherapy, and those who were randomized to 4 courses of conventional standard-dose multi-agent chemotherapy received only one course of intrathecal therapy. Although children are reported to be high-risk for CNS disease at diagnosis, the

incidence generally decreases as the age increases. [26] CNS involvement is relatively rare with adult AML, and CNS prophylaxis is not generally recommended for adults without CNS symptoms. [27] We hypothesize that AYAs with AML may not need CNS prophylaxis as intense as for younger children; this needs to be tested in a future AYA study.

In the current study, OS and EFS were similar between age groups among AYAs, although the survival rates of children, AYAs and older adults were significantly different. In the previous studies, survival of AYAs was reportedly similar to or worse than younger children depending on different studies. [2, 3] A German study comparing survival of infants, children, and adolescent and young adults reported that the survival of adolescents was inferior compared with children, being the most unfavorable among young adults. [1] Although a number of reports comparing AYAs and older adults are limited, one study from the MD Anderson Cancer Center reported that survival of AYAs tended to be longer compared with that of the adults. [23] In the previous studies from Japan and other countries, risks of TRM in AYAs were generally higher than those in the younger children. [4] In this study, none of the deaths among young adult patients who were from 25 to 39 were attributed to treatment.

In our study, older patients were more likely to receive allo-HSCT. Indication of allo-HSCT was different between the pediatric and adult protocols: high-risk cytogenetics or poor response to the initial induction therapy for the JPLSG AML-05 protocol; intermediate and high-risk cytogenetics, and availability of a histocompatible donor for the JALSG AML201 protocol. Transplant indication for AYAs with AML is not yet well understood and needs to be clarified, especially for the intermediate risk group. In this study, detailed information related to stem cell transplantation, such as disease status at transplantation and second or following relapse after the transplant, was not available; therefore, relapse rate and transplant-related mortality as transplant outcomes were not analyzed. Majhail et al. reported allogenic transplant outcomes of

AYAs with AML, and compared the results to those of children and older adults. In the study, OS of AYAs were worse than children and better than older adults. [28] The other study from the University of Minnesota reported no difference in outcomes between children and AYAs. [29] Tomizawa et al. compared the transplant results of the children and AYAs of the Japanese transplant registry. [30] Similar with the results from the CIBMTR, OS was better for children than AYAs; TRM was higher for AYAs than for children under 15 years old.

To conclude, we reported here the results of AYAs with AML from the JPSLG and the JALSG protocols, and this is the first attempt to combine the clinical and genetic data of pediatric and adult AML studies from the two major Japanese nation-wide multicenter prospective study groups. Further prospective studies of AYAs with this disease are warranted.

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Disclosure of Conflicts of Interest

Dr. Miyachi reports grant from AMED and personal fees for consulting from BML. Inc. Dr. Saito reports grants from Ministry of Health, Labor and Welfare, grants from AMED, during the conduct of the study. Dr. Asou reports grants from Nippon Shinyaku Co., Ltd., grants and personal fees from Kyowa Hakko Kirin Co., Ltd., grants and personal fees from Chugai Pharmaceutical Co., Ltd., grants from Astellas Pharma Inc., grants and personal fees from Sumitomo Dainippon Pharma Co., Ltd., grants from

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Table

Tables and figures

Table 1. Characteristics of the AYAs with AML

	15-24 years (n=53)		25-39 years (n=50)		Total (n=103)		
Characteristic	**N**	**(%)**	**N**	**(%)**	**N**	**(%)**	***P*-value**
Study protocol							<0.001
AML201	23	(43)	50	(100)	73	(71)	
AML-05	30	(57)	0	(0)	30	(29)	
WBC/μl, median (range)	15400	(1390-18000)	21450	(230-203300)	17150	(230-203300)	0.86
							0.38
≤20,000 /μl	30	(57)	24	(48)	54	(52)	
>20,000 /μl	23	(43)	26	(52)	49	(48)	
Cytogenetic risk group							0.79
good	16	(30)	17	(34)	33	(32)	
intermediate	34	(64)	29	(58)	63	(61)	
poor	3	(6)	4	(8)	7	(7)	
FAB							0.59
M0	1	(2)	2	(4)	3	(3)	
M1	8	(15)	12	(24)	20	(19)	
M2	24	(45)	23	(46)	47	(46)	
M4	11	(21)	8	(16)	19	(18)	
M5	6	(11)	5	(10)	11	(11)	
M6	1	(2)	0	(0)	1	(1)	
ND/RAEB-T	2	(4)	0	(0)	2	(2)	
Stem cell transplantation							0.045
yes	28	(53)	36	(72)	64	(62)	
no	25	(47)	14	(28)	39	(38)	

Abbreviation: ND, not determined

Table 2. Frequencies of each mutation in AYAs

	15-24 years (n=53)		25-39 years (n=50)		Total (n=103)		
Characteristic	**N**	**(%)**	**N**	**(%)**	**N**	**(%)**	***P*-value**
FLT3-ITD							0.20
wild	39	(74)	42	(84)	81	(79)	
ITD	14	(26)	8	(16)	22	(21)	
KIT							0.48
wild	27	(84)	39	(78)	66	(80)	
Mt	5	(16)	11	(22)	16	(20)	
CEBPA							0.15
wild	34	(77)	44	(88)	78	(83)	
single	7	(16)	2	(4)	9	(10)	
double	3	(7)	4	(8)	7	(7)	
NRAS							0.24
wild	48	(96)	45	(90)	93	(93)	
Mt	2	(4)	5	(10)	7	(7)	
KRAS							0.73
wild	45	(90)	46	(92)	91	(91)	
Mt	5	(10)	4	(8)	9	(9)	
WT1							0.46
wild	45	(90)	47	(94)	92	(92)	
Mt	5	(10)	3	(6)	8	(8)	
NPM1							1.00
wild	43	(86)	43	(86)	86	(86)	
Mt	7	(14)	7	(14)	14	(14)	
MLL-PTD							0.90
wild	48	(98)	40	(98)	88	(98)	
Mt	1	(2)	1	(2)	2	(2)	
FLT3-ITD/NPM1 combination							0.54
FLT3ITD-/NPM1-	32	(64)	37	(74)	69	(69)	
FLT3ITD-/NPM1+	4	(8)	5	(10)	9	(9)	
FLT3ITD+/NPM1-	11	(22)	6	(12)	17	(17)	
FLT3ITD+/NPM1+	3	(6)	2	(4)	5	(5)	

Table 3. Multivariate analysis of genetic mutations for risk of mortality of AYAs

		HR	(95% CI)	P-value
Overall survival				
FLT3-ITD	wild	1.00		
	ITD	2.10	(1.07 - 4.12)	0.031
NPM1	wild	1.00		
	mt	0.24	(0.06 – 1.00)	0.050
Event free survival				
FLT3-ITD	wild	1.00		
	ITD	2.37	(1.34 - 4.21)	0.003
NPM1	wild	1.00		
	mt	0.24	(0.09 – 0.67)	0.006

Abbreviations: HR, hazard ratio; 95% CI, 95% cumulative incidence; mt, mutated

Figure Legends

Figure 1. Overall survival of AYAs according to baseline characteristics

Overall survival according to (A) age groups, (B) WBC at diagnosis and (C) cytogenetic risk groups

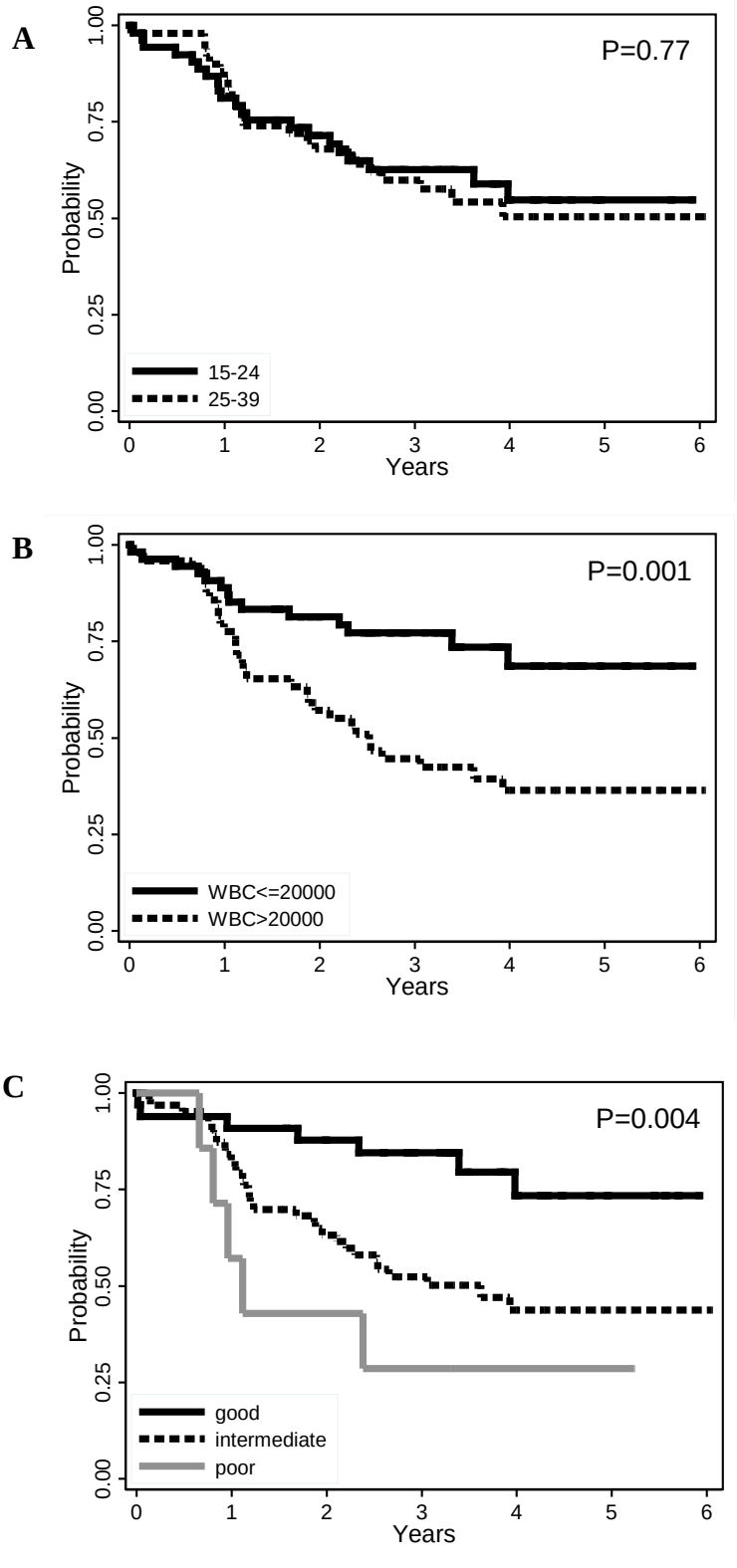
Figure 2. Frequencies of gene mutations

Association of mutated genes is shown by Circos plot. [31]

Figure 3. Survival difference according to NPM1 and *FLT3*-ITD mutation status

(A) Overall survival and (B) event free survival curves are categorized by combinations of *NPM1* and *FLT3*-ITD mutation status. The curves are stratified into wild type for both mutations (wt/wt), FLT3-ITD-/NPM1+, FLT3-ITD+/NPM1- and FLT3-ITD+/NPM1+.

Figure 1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0.75
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15-24
25-39
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2
3
4
5
6
Years
B
P=0.001
Probability
0.00
0.25
0.50
0.75
1.00
WBC<=20000
WBC>20000
0
1
2
3
4
5
6
Years
C
P=0.004
Probability
0.00
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1.00
good
intermediate
poor
0
1
2
3
4
5
6
Years

Figure 2

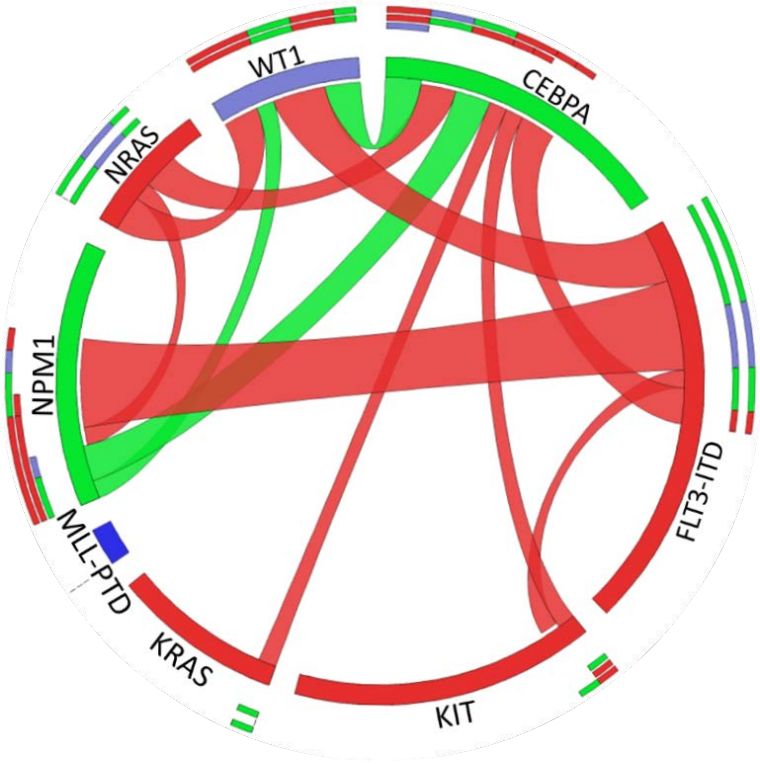
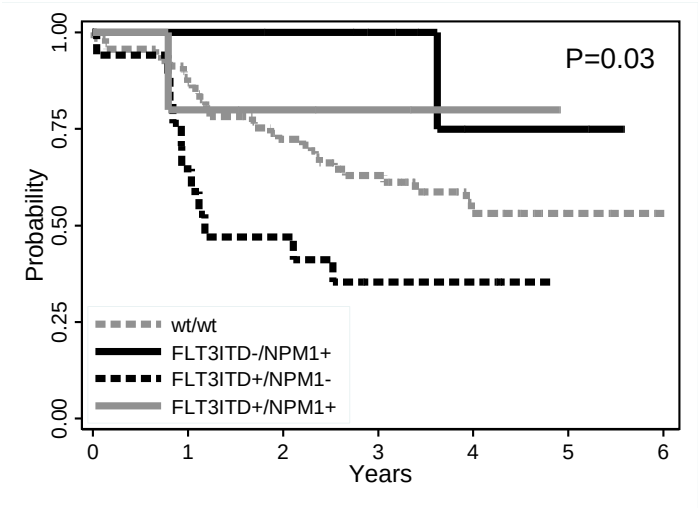


Figure 3

A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2
3
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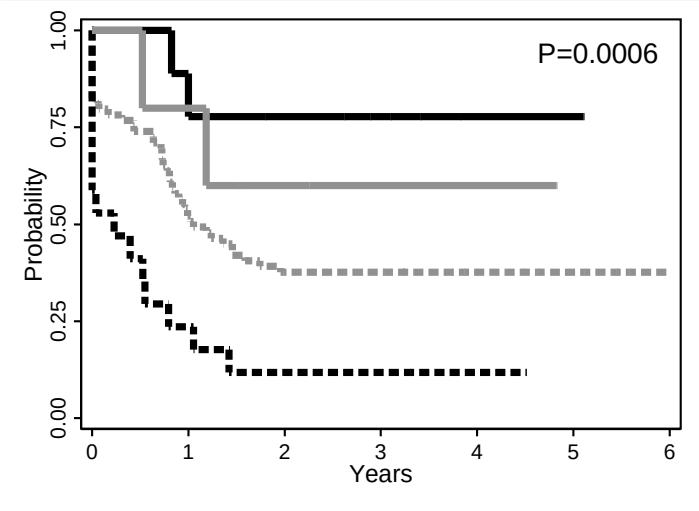
5
6
Years
wt/wt
FLT3ITD-/NPM1+
FLT3ITD+/NPM1-
FLT3ITD+/NPM1+

B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3
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Years