RESEARCH COMMUNICATION

Incidence and Prognostic Importance of Molecular Genetic Defects in Children with Acute Myeloblastic Leukemia

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Abstract

Introduction: Acute myeloblastic leukemia (AML) accounts for 15 to 25 percent of childhood acute leukemias. The most common genetic abnormalities seen in pediatric AML patients are AML1-ETO, PML-RARα and CBFB-MYH11 genes resulting in t(8;21), t(15;17) and inv(16). These genetic defects are seen in approximately 20-25% of AML patients. Objective: We investigated in this study, incidence and prognostic significance of the AML1-ETO, PML-RARα and CBFB-MYH11 genes in children with AML. Materials and Methods: The authors analyzed 34 children with AML using the real time-polymerase chain reaction for AML1-ETO, PML-RARα and CBFB-MYH11 genes. Results: Of the patients, 8.8% were positive for t(8;21), 8.8% for t(15;17) and 3% for inv(16). There were a statistically significant differences between 48 month overall survival rates of the patients positive and negative for t(8;21), t(15;17) and inv(16). Conclusion: It was concluded that t(15;17), t(8;21) and inv(16) impact on disease prognosis positively, but comprehensive studies with larger patient series are now needed for confirmation.

Keywords: Childhood - acute myeloblastic leukemia - genetic defects - prognosis

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Introduction

Acute myeloblastic leukemia (AML) is a heterogeneous group of malign diseases characterized by uncontrolled proliferation of myeloid progenitor cells in the bone marrow. It accounts for 15-25% of all childhood acute leukemias. It has not been known any genetic predisposing factor involving in the development of AML. However, there is evidence indicating that certain genetic abnormalities play a role in AML development (Golub and Arceci, 2002).

The most commonly encountered genetic abnormalities in pediatric AML patients are AML1-ETO, PML-RARα and CBFB-MYH11 genes resulted from t(8;21), t(15;17) and inv(16) (Martinez-Climent et al., 1995). These genetic abnormalities are seen in approximately 20-25% of the patients with AML. These genetic abnormalities are detected with "real-time polymerase chain reaction" (RT-PCR). This method is so sensitive as to detect 1 leukemic cell in 100,000 to 1,000,000 (Heid et al., 1996).

In this study, it was aimed to ascertain the frequency and prognostic importance of AML1-ETO, PML-RARα and CBFB-MYH11 genes among pediatric patients with newly-diagnosed or relapsed AML at the time of diagnosis.

Materials and Methods

This study included 34 patients diagnosed with AML by cytomorphological, immunohistochemical and "*flow cytometry*" studies in the Division of Pediatric Oncology at Çukurova University Medical School. Twenty six patients were newly diagnosed with AML, eight patients had relapsed disease. The consents of the families and local ethics committee approval for the study were received. To detect t(8;21), t(15;17) and inv(16), RT-PCR method was used.

Statistical Tests

All statistical analysis were done using SPSS ver. 12.0 (statistical package for social sciences) software. Pearson Chi-Square test was used to assess the data of the patients. In addition, one-way ANOVA test and Mann-Whitney U test were used. The level of significance was set to a p value <0.05. Overall survival (OS) were calculated according to the Kaplan-Meier method. Event was defined as death for OS. Comparisons of OS between groups were based on the log-rank test.

Results

Of the 34 patients, 26 (76%) were newly diagnosed with AML and eight (24%) had relapsed AML. The ages ranged from 1 to 17 years with the mean age of 8.3±3.8 years. The female/male ratio was 1/1.4; 58.8% of the patients were boys.

According to the French-American-British (FAB)

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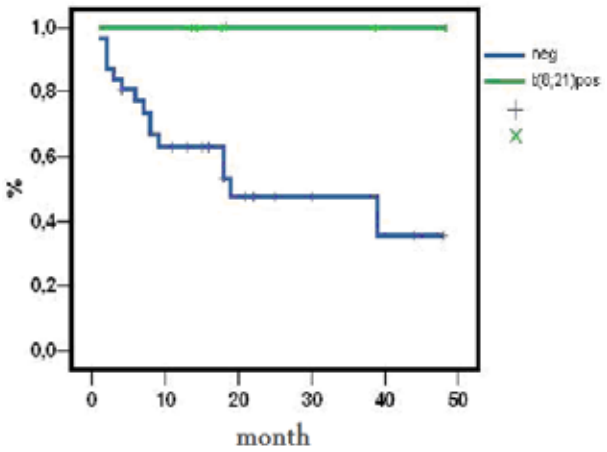

Figure 1. Overall Survival According to t(8;21) Positivity, Kaplan-Meier Test

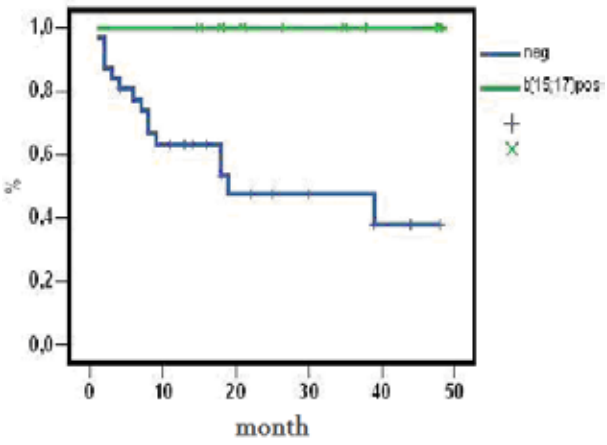

Figure 2. Overall Survival According to t(15;17) Positivity, Kaplan-Meier Test

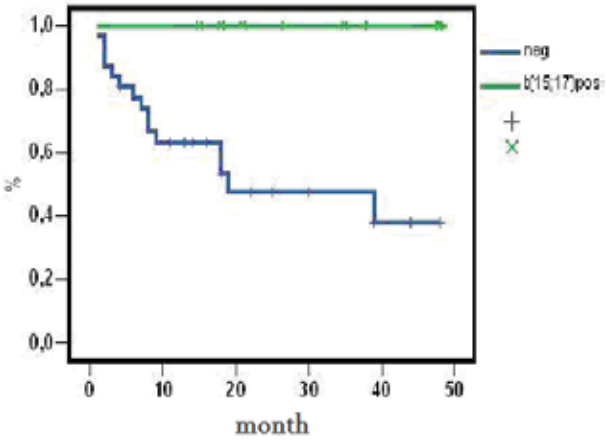

Figure 3. Overall Survival According to inv(16) Positivity, Kaplan-Meier Test

classification, morphological distribution of the patients was as follows: 8 were AML M0; 3 were AML M1; 4 were AML M2; 2 were AML M3, 6 were AML M4; 8 were AML M5 and 3 were AML M7. We did not have any patient diagnosed with AML M6. The overall survival rates of our patients were as follows: 12 month survival rate was 67%; 24 month survival rate was 58%; 36 month survival rate was 52%; 48 month survival rate was 42%.

AML1-ETO gene was detected in three (8.8%) patients. The mean age of these patients was 7.6 years. All three patients had newly diagnosed AML. They had AML-2, M3, M7 morphologies according to the FAB system. None of these three patients relapsed during follow up. There was a statistically significant difference between 48 month OS rates of the patients who were positive for and those who were negative for t(8;21) (p: 0.0036), (Figure 1).

Three patients (8.8%) were positive for PML-RARα. The mean age of these patients was 8.5 years. All three patients were newly diagnosed with the disease. They had AML-M2, -M3, and -M4 morphologies according to the FAB classification. None of these three patients relapsed. There was a statistically significant difference between 48 month OS rates of the patients who were positive for and those who were negative for t(15;17) (p: 0.0036), (Figure 2).

CBFB-MYH11 was found to be positive in only one patient (3.3%). This patient was 6 years old at the time of diagnosis and had AML-M4 morphology. The patient did not relapse during follow up. There was a statistically significant difference between 48 month OS rates of the patients who were positive for and those who were negative for inv(16) (p: 0.0024),(Figure 3).

Discussion

With the studies conducted in the recent 30 years, it has been understood that genetic abnormalities play an important role in the etiology of childhood AML. These genetic defects detected in leukemias reflect the biology of the cell and thereby the clinical course and prognosis of the disease. The most common genetic abnormalities encountered in pediatric AML patients are AML1-ETO, PML-RARα and CBFB-MYH11 genes resulted from t(8;21), t(15;17) and inv(16). These genetic abnormalities were observed in approximately 20-25% of AML patients (Golub and Arceci, 2002; Martinez-Climent et al., 1995; Heid et al., 1996; Martinez-Climent, 1997).

Approximately 6-10% of all AML patients are positive for AML1-ETO gene, this frequency increases up to 90% among the patients having AML M2 morphology (Nucifora et al., 1993). In a study conducted by Andrieu et al., (1996), it was reported that it was more common among young children with AML and might occur in AML M1, AML M4 and in other AML groups except AML M2. In a study performed by Sarper et al., (2000), 3.5 year OS rate was found to be 28.5% for seven patients positive for AML1-ETO gene. As a consequence of this result, it was reported that t(8;21) was not an indicator of good prognosis. In another study conducted by Grimvade (2001), 100% remission occurred in t(8;21) positive patients, while 2 year disease-free survival rate was reported to be 50%. In our study, AML1-ETO gene was found to be positive in 3 (8.8%) of 34 AML patients. This rate is consistent with the rates reported in the literature. Our patients had AML M2, AML M3 and AML M7 morphologies according to the FAB classification. They were 6, 7 and 10 years old. It was reported the literature that it occurred in younger ages. After four-year follow up, survival rate was 100%. As a consequence of these results, it was concluded that AML1-ETO gene positive patients had good prognosis. However, it has been thought that conducting more comprehensive studies is needed

because of the few numbers of the patients.

PML-RARα gene resulted from t(15;17) is another genetic abnormality seen in childhood AML. Almost all acute promyelocytic leukemia (AML-M3) is t(15;17) positive. In the studies, PML-RARα gene was reported in a rate of 6-9% among all AML patients (Betancourt-García et al., 2009; Sazawal et al., 2009). In a retrospective study from Italy conducted by Testi et al., (2005), PML-RARα gene was found in 91 (9%) of 983 patients. PML-RARα positive patients were given ATRA + idarubicin treatment, and a survival rate of 89% was reported after 10-year follow up. In the study performed by Hu et al., (2000), 5 year survival rate was 69.9% among PML-RARα positive 70 patients with promyelocytic leukemia. In our study, PML-RARα gene was positive in three (8.8%) of 34 AML patients. This rate is consistent with the rates reported in the literature. Our patients had AML M2, AML M3 and AML M4 morphologies according to the FAB classification. The patient diagnosed with AML M3 was treated with idarubicin plus ATRA. After four-year follow up, survival rate was 100%. However, it has been thought that more comprehensive studies are needed because of the few numbers of the patients.

CBFB-MYH11 gene resulted from inv(16) is another genetic defect seen in childhood AML. This genetic defect exists in 4-7% of all AML patients. It is more common among the patients having AML M4 morphology according to the FAB classification (Ferrara et al., 2004; Ichikawa et al., 2006). In the study conducted by Liu et al., (1995), CBFB-MYH11 gene was found in 18 (6%) of 308 AML patients. In the study conducted by Poirel et al., (1995), 206 of 241 inv(16) positive patients had AML M4 morphology. In the study performed by Martin et al., (2000), it was found that 15 of 16 CBFB-MYH11 gene positive patients had AML M4 morphology. Eight of these patients relapsed within 18 months. In our study, we found CBFB-MYH11 gene in one (3%) of 34 AML patients. Morphologic diagnosis of that patient was AML M4, corresponding with the literature data. During the four-year follow up, the patient of ours did not have any problem.

In conclusion, genetic abnormalities seen in childhood acute leukemias have a significant role to assign the prognosis. It was found that the results obtained from our study were consistent with the rates of genetic abnormalities seen in AML patients, which were reported in the literature, however, the prognoses of the patients having genetic abnormalities were better. It was concluded that comprehensive studies with larger patient series are needed to achieve more reliable results.

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