Paediatrica Indonesiana

p-ISSN 0030-9311; e-ISSN 2338-476X; Vol.61, No.3(2021). p.155-64; DOI: 10.14238/pi61.3.2021.155-64

Original Article

3-year survival rate in acute lymphoblastic leukemia: comparison of ALL-2006 and ALL-2013 protocols

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Abstract

Background With advances in supportive and risk-stratified therapy, the 5-year survival rate of acute lymphoblastic leukemia has reached 85.5%. The ALL-2006 treatment protocol was modified and renamed the ALL-2013 protocol, with dose and duration changes.

Objective To compare outcomes of the ALL-2006 and ALL-2013 protocols, with regards to mortality, remission, relapse, and three-year survival rates.

Methods This was retrospective cohort study. Subjects were acute lymphoblastic leukemia (ALL) patients treated from 2011 to 2018 in Mohamad Hoesin Hospital, Palembang, South Sumatera. The three-year survival rates, relapse, remission rates and comparison of ALL-2006 and ALL-2013 protocols were analyzed with Kaplan-Meier method.

Results Mortality was significantly correlated with age at diagnosis <1 year and >10 years, hyperleukocytosis, and high-risk disease status. Patients aged 1 to 10 years, with leukocyte count <50,000/mm3 and standard-risk status had significantly higher likelihood of achieving remission. Mortality was not significantly different between the ALL-2006 protocol group [70.6%; mean survival 1,182.15 (SD 176.89) days] and the ALL-2013 protocol group [72.1%; mean survival 764.23 (SD 63.49) days]; (P=0.209). Remission was achieved in 39.2% of the ALL-2006 group and 33% of the ALL-2013 group (P>0.05). Relapse was also not significantly different between the two groups (ALL-2006: 29.4% *vs.* ALL-2013: 17.9%; P>0.05). Probability of death in the ALL-2006 group was 0.3 times lower than in the ALL-2013 group (P<0.05), while that of the high-risk group was 3 times higher. Remission was 2.19 times higher in those with leukocyte <50,000/mm3 compared to those with hyperleukocytosis. In addition, relapse was significantly more likely in high-risk patients (HR 2.96; 95%CI 1.22 to 7.19). Overall, the 3-year survival rate was 33%, with 41.7% in the ALL-2006 group and 30.7% in the ALL-2013 group.

Conclusion Three-year survival rate of ALL-2006 protocol is higher than that of ALL-2013 protocol but is not statistically significant. Age at diagnosis <1 year and >10 years, hyperleukocytosis, and high-risk group are significantly correlated with higher mortality and lower remission rates. However, these three factors are not significantly different in terms of relapse. **[Paediatr Indones. 2021;61:155-64 ; DOI: 10.14238/pi61.3.2021.155-64**].

Keywords: *acute lymphoblastic leukemia, prognostic factors, survival, ALL-2006 protocol, ALL-2013 protocol*

Acute lymphoblastic leukemia (ALL) is the most common malignancy in children. With advancements in diagnosis, treatment, supportive therapy, and risk-stratified therapy, the 5-year survival rate of ALL in United States has increased from 69.3% in 1981 to 85.5% in 2010. In Indonesia, several chemotherapy protocols have been used over the years, including the *National Protocol (Jakarta), Wijaya Kusuma ALL-2000 Protocol, National ALL-2006 Protocol,* and the *ALL-2013 Protocol*. The ALL-2013 protocol is a modified version of the ALL-2006 protocol, based on an evaluation of ALL-2006 presented at the Indonesian Congress of Pediatrics, which stated that the survival rate of ALL was still low.[1,2]

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Submitted December 2, 2020. Accepted May 18, 2021.

Both the ALL-2006 and ALL-2013 protocols use the same chemotherapy regimen, but different doses and maintenance phase time. The ALL-2013 cumulative doses are higher than that of the ALL-2006 protocol. L-asparaginase was given 6 times for 2 weeks in ALL-2006, and 9 times for 3 weeks in ALL-2013. The ALL-2013 has a longer maintenance duration of 109 weeks, while that of ALL-2006 lasts only 102 weeks.[2]

The Pediatric Hematology-Oncology Division at Dr. Mohammad Hoesin Hospital Palembang, South Sumatera, used the *ALL-2006 Protocol* for patients diagnosed from 2011 to 2014 and the **ALL-2013 Protocol** for those diagnosed from 2015 to 2018. A total of 261 patients were diagnosed from 2011-2018. A 2017 study in Dharmais Hospital, Jakarta, stated that the remission was achieved in 30% of ALL-2006 patients and 27% of ALL-2013 patients, which was not significantly different.[2] The purpose of this study was to compare outcomes of patients treated with the *ALL-2006* and *ALL-2013 Protocols*, assess for prognostic factors for mortality, remission, and relapse, as well as determine the three-year survival rate of childhood ALL.

Methods

This retrospective cohort study was conducted in Dr. Mohammad Hoesin Hospital, Palembang, South Sumatera. Subjects were pediatric ALL patients aged less than 18 years at the time of diagnosis and treated from 2011-2018 according to either the *ALL-2006* or *ALL-2013 Protocols*. The difference between *ALL-2006* and *ALL-2013 Protocols* are *ALL-2013* cummulative doses are higher. The L-asparaginase was given 6 times for 2 weeks in ALL-2006, and 9 times for 3 weeks in ALL-2013. The *ALL-2013* has a longer maintenance duration of 109 weeks, while that of ALL-2006 lasts only 102 weeks.

Patients who refused chemotherapy, were lost to follow up during therapy, or were in the maintenance phase of chemotherapy were excluded. Patients diagnosed from 2011 to 2014 were treated according to the *ALL-2006 Protocol*; patients diagnosed from 2015 to 2018 were treated according to the *ALL-2013 Protocol*. Both protocols were risk-stratified according to age at diagnosis, leukocyte counts, presence/absence of mediastinal mass, meningeal leukemia, as well as T-cell leukemia. Standard-risk criteria were age between 1 to 10 years, leukocyte count $<50,000/mm^3$, no mediastinal mass, no blast in cerebrospinal fluid, and B-cell immunophenotyping, whereas high-risk criteria were otherwise.

Patient information was collected from secondary data of patient ALL registers and medical records. The most recent condition of the patient was confirmed by medical records or by family reports. Clinical manifestation, laboratory results, morphology, immunophenotyping, risk status, and time of the first chemotherapy were collected from the ALL register. Time of death, remission, and relapse were stated by date, while duration was stated in days.

We classified patients by age group according to risk-stratification, which were <1 year and >10 years, or 1 to 10 years. Hemoglobin level was classified as anemia or not anemia, according to age. Hyperleukocytosis was defined as leukocyte counts more than $50,000/mm^3$. Thrombocytopenia was defined as platelet counts less than $100,000/mm^3$. Morphology was classified into two categories: L1-L2 and L3. Immunophenotyping was classified into either B-cell precursor or T-cell and mature B-cell. This study was approved by the Ethics Committee of Universitas Sriwijaya Medical School.

Statistical analyses were carried out using *IBM SPSS 22*. Descriptive statistics was used to characterize the patients. The three-year survival rate and comparison of protocols were analyzed with survival-analysis and Kaplan-Meier method, and compared with log-rank test. Bivariate variables of remission were analyzed with Chi-square test or Fisher's exact test. Multivariate factors were analyzed with Cox regression, with 95% confidence intervals and statistical significance set at 5% (P value <0.05).

Results

Of 261 ALL patients diagnosed from 2011 to 2018, 9 patients refused chemotherapy, 7 patients died before chemotherapy, and 6 patients were in maintenance phase chemotherapy. Of the 239 patients who met the inclusion criteria, 9 patients were lost to follow up during chemotherapy, hence, a total of 230 patients were included. Males comprised 66.5% of subjects, and 1-10-year-olds comprised 80.4% (**Table 1**). The most common clinical manifestations were pallor

(98.3%), hepatomegaly (72.6%), and fever (63.9%). Fifty-one patients received the ALL-2006 protocol and 179 patients received the ALL-2013 protocol. Morphology L1 was found in 62.6% of all subjects. Immunophenotyping was not performed in 109 (47.4%) subjects because it was not covered by the *National Health Insurance*, and not all the patients were able to afford it.

By the end of the observation period, 165/230 patients had died. There were significant differences in mortality rates with regards to age at diagnosis, hyperleukocytosis, and risk status (P<0.05). Log-rank test revealed that patients aged <1 or >10 years had a significantly shorter mean survival than those aged 1-10 years [370.976 (80.48) *vs.* 1,138.28 (101.2) days, respectively; (P<0.001)]. Similarly, patients with hyperleukocytosis had significantly shorter mean survival compared to patients without hyperleukocytosis, [622.63 (100.69) *vs.* 1098.66 (107.2) days, respectively; (P=0.005)]. Patients with high risk status significantly shorter survival compared to standard risk [583.40 (70.75) *vs.* 1,407.96 (139.81) days, respectively; (P<0.001)]. However, the remaining variables of gender, hemoglobin level, platelet count, morphology, and immunophenotyping did not result in significantly different survival rates, as shown in **Table 2**.

Table 1. Characteristics of ALL patients in year 2011-2018

Characteristics	ALL-2006 protocol (n=51)	ALL-2013 protocol (n=179)	ALL (N=230)
Age, n (%)			
< 1 year	1(2.0)	3(1.7)	4(1.7)
1-10 years	41(80.4)	144(80.4)	185(80.5)
>10-18 years	9(17.6)	32(17.9)	41(17.8)
Sex, n (%)			
Male	37(72.5)	116(64.8)	153(66.5)
Female	14(27.5)	63(35.2)	77(33.5)
Clinical manifestation*, n (%)			
Pallor	51(100)	175(97.8)	226(98.3)
Fever	31(60.8)	116(64.8)	147(63.9)
Bone pain	4(7.8)	40(22.3)	44(19.1)
Bleeding	16(31.4)	65(36.3)	81(35.2)
Seizure	0(0.0)	4(2.2)	4(1.7)
Hepatomegaly	38(74.5)	129(72.1)	167(72.6)
Splenomegaly	36(70.6)	99(55.3)	135(58.7)
Lymphadenopathy	29(56.9)	100(55.9)	129(56.1)
Hemoglobin levels, n (%)			
Normal	4(7.8)	8(4.5)	12(5.2)
Anemia	47(92.2)	171(95.5)	218(94.8)
Leukocyte count, n (%)			
Normal	28(54.9)	85(47.5)	113(49.1)
Leukopenia	6(11.8)	42(23.5)	48(20.9)
Hyperleukocytosis	17(33.3)	52(29.1)	69(30.0)
Platelet count, n (%)			
Normal	2(3.9)	13(7.3)	15(6.5)
Thrombocytopenia	49(96.1)	166(92.7)	215(93.5)
Morphology, n (%)			
L1	34(66.7)	110(61.5)	144(62.6)
L2	14(27.5)	62(34.6)	76(33.0)
L3	3(5.9)	7(3.9)	10(4.3)
Immunophenotyping, n (%)			
B-cell precursor	8(16)	92(51.4)	100(43.5)
Mature B-cell	0(0.0)	3(1.7)	3(1.3)
T-cell	1(1.7)	17(9.5)	18(7.8)
No data	42 (82.3)	67 (37.4)	109(47.4)

Table 2. Possible risk factors of acute lymphoblastic leukemia mortality

Risk factors	Died (n=165)	Survived (n=65)	Mean survival (SD), days	HR (95%CI)	P value
Age					
<1 or >10 years	41 (91.1)	4 (8.9)	370.96 (80.48)	2.2 (1.6-3.15)	<0.001
1-10 years	124 (67.0)	61 (33.0)	1,138.28 (101.2)		
Gender					
Male	111 (72.5)	42 (27.5)	871.72 (79.82)	0.9 (0.7-1.37)	0.971
Female	54 (70.1)	23 (29.9)	1,049.42 (151.8)		
Anemia					
Yes	159 (72.9)	59 (27.1)	962.65 (88.96)	1.8 (0.8-4.1)	0.147
No	6 (50.0)	6 (50.0)	1,381.27 (319.1)		
Hyperleukocytosis					
Yes	57 (81.4)	13 (18.6)	622.63 (100.6)	1.6 (1.15-2.1)	0.005
No	108 (67.5)	52 (32.5)	1,098.66 (107.2)		
Thrombocytopenia					
Yes	154 (71.6)	61 (28.4)	874.09 (69.46)	1.1 (0.6-2.1)	0.716
No	11 (73.3)	4 (26.7)	1,102.20 (320.5)		
Morphology					
L3	9 (90.0)	1 (10.0)	447.10 (190.5)	1.6 (0.8-3.1)	0.165
L1-L2	156 (70.9)	64 (29.1)	1,016.95 (89.75)		
Immunophenotyping					
Mature B-cell, T-cell	18 (85.7)	3 (14.3)	478.36 (129.9)	1.6 (0.95-2.7)	0.073
B-cell precursor	65 (65.0)	35 (35.0)	897.37 (89.59)		
Risk status					
High	105 (84.0)	20 (16.0)	583.40 (70.75)	2.2 (1.6-3.0)	<0.001
Standard	60 (57.1)	45 (42.9)	1,407.96 (139.81)		

*Log-rank

Most patients who died did so during induction phase (81; 49%), followed by maintenance phase (16.3%), second induction after relapse (15%), intensification/consolidation phase (9.4%), second maintenance after relapse (5.8%), after chemotherapy (3%), and second intensification/consolidation after relapse (1.8%). The most common cause of death was bleeding in both protocols (37-38.8%). Of those who died during induction phase, the main causes of death were tumor lysis syndrome in the *ALL-2006 Protocol* (42.9%) and bleeding (38.8%) in the *ALL-2013 Protoco*l. Of the 81 induction phase deaths, 67 out of a total of 129 deaths (51.9%) occurred in the *ALL-2013 Protocol*, and 14 out of a total of 36 (38.9%) occurred in the *ALL-2006 Protocol*. The ALL-2006 group had fewer deaths than the ALL-2013 group, but the difference was not statistically significant (RR 0.66; 95%CI 0.363 to 1.198; P=0.166).

Seventy-nine patients had remission during the observation; 151 did not reach remission. Children aged 1 to 10 years had a 2.9 times greater chance of remission compared to age <1 year or >10 years (RR 2.959; 95%CI 1.3 to 6.3). Leukocycte counts <50,000/mm^3 showed 2 times higher chance of remission than hyperleukocytosis (RR 2.031; 95%CI 1.266 to 3.364), and standard risk had 2.5 times higher chance of remission than high risk status (RR 2.571, 95%CI 1.729 to 3.824) (**Table 3**).

A total of 47 subjects relapsed, but there were no significant differences between risk of relapse among the potential risk factors, as shown in **Table 4**. No patients with morphology L3 had relapse, but 9 of the 10 patients died. Regarding time of relapse, 31.9% had early relapse, 53.2% intermediate relapse, and 14.9% late relapse. Marrow relapse occurred in 89.4% of patients, testicular relapse in 6.4%, and meningeal relapse in 4.3%. After relapse, only 21.3% of subjects achieved remission, 2.1% had another case of relapse, and 76.6% died after relapse.

High mortality rates were observed in both the ALL-2013 and ALL-2006 groups, with 72.1% and 70.6%, respectively; (P=0.209) (**Table 5**). Despite the lack of significant mortality rate difference between the two protocol groups, the ALL-2006 group had longer survival duration than the ALL-2013 group [mean 1,182.15 (176.89) *vs.* 764.23 (63.49) days, respectively; median: 622 *vs.* 327 days, respectively] (**Figure 1**).

Table 3. Potential factors contributing to remission of ALL

Parameters	Remission (n=79)	No remission (n=151)	RR (95% CI)	P value
Age, n (%)				
1-10 years	73 (39.5)	112 (60.5)	2.959 (1.3 to 6.3)	0.001[a]
<1 or >10 years	6 (13.3)	39 (86.7)		
Gender, n (%)				
Female	27 (35.1)	50 (64.9)	1.03 (0.7 to .5)	0.871[a]
Male	52 (34.0)	101 (66.0)		
Anemia, n (%)				
No	7 (58.3)	5 (41.7)	1.766 (1 to 2.9)	0.114[b]
Yes	72 (33.0)	146 (67.0)		
Hyperleukocytosis, n (%)				
No	65 (40.6)	95 (59.4)	2.031 (1.2 to 3.6)	0.002[a]
Yes	14 (20.0)	56 (80.0)		
Thrombocytopenia, n (%)				
No	5 (33.3)	10 (66.7)	0.968 (0.46 to 2)	0.932[a]
Yes	74 (34.4)	141 (65.6)		
Morphology, n (%)				
L1-L2	77 (35.0)	143 (65.0)	1.75 (0.5-6.1)	0.501[b]
L3	2 (20.0)	8 (80.0)		
Immunophenotyping, n (%)				
B-cell precursor	39 (39.0)	61 (61.0)	2.048 (0.8 to 5.1)	0.082[a]
Mature B-cell and T-cell	4 (19.0)	17 (81.0)		
Risk status, n (%)				
Standard	54 (51.4)	51 (48.6)	2.571 (1.7 to 3.8)	<0.001[a]
High	25 (20.0)	100 (80.0)		

[a]Pearson's; Chi-square; [b]Fisher's exact test

Table 4. Potential risk factors for relapse in ALL

Parameters	Relapse (n=47)	Non-relapse mortality/ relapse-free survival (n=183)	Mean relapse (SD), days	P value* HR (95%CI)
Age, n (%)				
<1 or >10 years	8 (17.8)	37 (82.2)	1,044.02 (168.6)	0.066
1-10 years	39 (21.1)	146 (78.9)	1,611.82 (86.88)	HR=2 (0.9-4.3)
Gender, n (%)				
Male	36 (23.5)	117 (76.5)	1,385.29 (85.77)	0.119
Female	11 (14.3)	66 (85.7)	1,750.0 (139.90)	HR=1.7(0.8-3.3)
Anemia, n (%)				
Yes	45 (20.6)	173 (79.4)	1,551.33 (85.09)	0.476
No	2 (16.7)	10 (83.3)	1,360.85 (163.9)	HR=1.6(0.4-6.8)
Hyperleukocytosis, n (%)				
Yes	12 (17.1)	58 (82.9)	1,554.11 (168.4)	0.758
No	35 (21.9)	125 (78.1)	1,539.68 (94.04)	HR=1.1 (0.5-2.1)
Thrombocytopenia, n (%)				
Yes	43 (20.0)	172 (80.0)	1,576.63 (85.92)	0.643
No	4 (26.7)	11 (73.3)	1,194.71 (1962)	HR=0.7 (0.8-2.2)
Morphology, n (%)				
L3	0 (0.0)	10 (100.0)	-	-
L1-L2	47 (21.4)	173 (78.6)	1,343.12(103.12)	
Immunophenotyping, n (%)				
Mature B-cell and T-cell	2 (9.5)	19 (90.5)	1,652.63 (239.4)	0.760
B-cell precursor	19 (19.0)	81 (81.0)	1,542.21 (101.2)	HR=0.8(0.2-3.4)
Risk status, n (%)				
High	23 (18.4)	102 (81.6)	1,464.57 (127.5)	0.224
Standard	24 (22.9)	81 (77.1)	1,595.8 (110.9)	HR=1.4(0.8-2.5)

*Log-rank

Table 5. Mortality comparison of the ALL-2006 and ALL-2013 protocol groups

Group	Died n (%)	Survived n (%)	Mean survival (SD), days	P value
ALL-2006 (n=51)	36 (70.6)	15 (29.4)	1,182.15 (176.89)	0.209
ALL-2013 (n=179)	129 (72.1)	50 (27.9)	764.23 (63.49)	

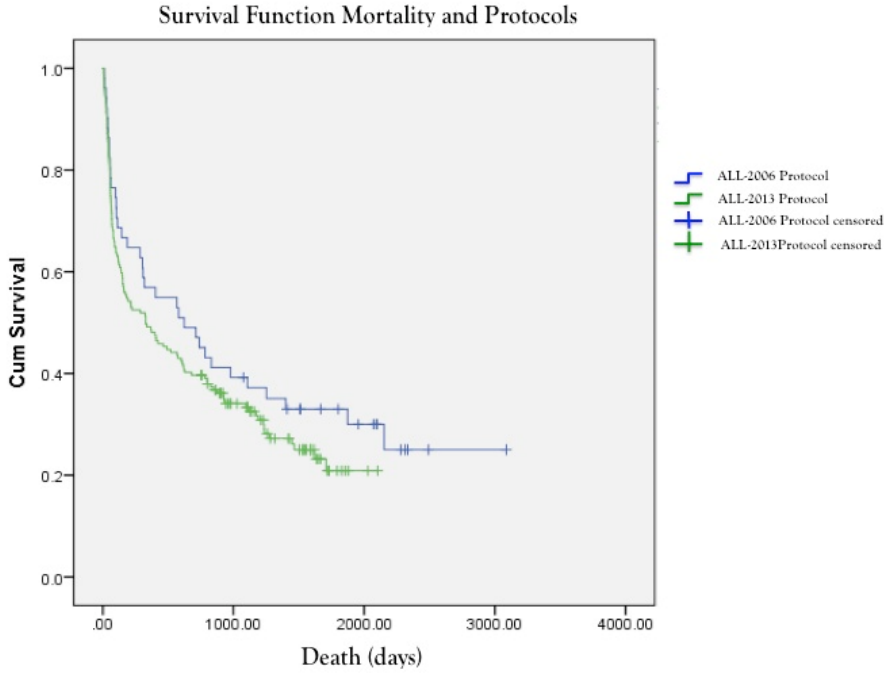

Figure 1. Mortality by *ALL-2006* and *ALL-2013 Protocols*

Table 6 shows that the ALL-2013 group had lower remission rate (33%) than the ALL-2006 group (39.2%), but the difference was not significant (P=0.963). Remission was reached in a mean of 848.66 (63.29) days in the ALL-2006 group, and 907.56 (35.41) days in the ALL-2013 group.

Relapse rate in the ALL-2006 group was higher than in the ALL-2013 group, but not significantly different (P=0.687) (**Table 7**). The mean time to relapse was 1,521.73 (146.08) days in the ALL-2006 group and 1,493.53 (84.29) days in the ALL-2013 group.

Univariate analysis revealed that age at diagnosis of <1 year and >10 years, hyperleukocytosis, and high-risk status were significantly correlated with increased risk of mortality (P<0.05). But that was not the case when Cox multivariate regression analysis of ALL-2006 group with regards to hyperleukocytosis, age <1 year and >10 year, T-Cell, L3, and high risk status, as only ALL protocol and risk status was significantly associated with increased mortality. The ALL-2006 group had 0.3 times lower risk of mortality compared to the ALL-2013 group (HR=0.34; 95%CI 0.121 to 0.925; P=0.035). High-risk patients had 3 times higher risk of mortality than standard risk patients (HR=3.407; 95%CI 1.897 to 4.894; P<0.001).

Univariate analysis revealed no significant factors for relapse, but when Cox multivariate regression applied to the ALL-2006 group with regards to hyperleukocytosis, age <1 year and >10 year, T-cell, L3, and high risk status, revealed that high risk status patients were significantly more likely (2.964 times higher) to have relapse compared to standard risk patients (HR=2.964; 95%CI 1.22 to 7.190; P=0.016).

Cox multivariate regression analysis of ALL-2013 group with regards to the age at diagnosis, leukocyte count, risk status, morphology, immunophenotyping, and chemotherapy protocol revealed that remission was

Table 6. Remission in the ALL-2006 and ALL-2013 protocol groups

Group	Remission n (%)	No remission n (%)	Time To remission (SD), days	P value
ALL-2006 (n=51)	20 (39.2)	31 (60.8)	848.66 (63.29)	0.936
ALL-2013 (n=179)	59 (33.0)	120 (67.0)	907.56 (35.41)	

Table 7. Relapse in the ALL-2006 and ALL-2013 protocol groups

Group	Relapse n (%)	No relapse n (%)	Time to relapse (SD), days	P value
ALL-2006 (n=51)	15 (29.4)	36 (70.6)	1,521.73 (146.08)	0.687
ALL-2013 (n=179)	32 (17.9)	147 (82.1)	1,493.54 (84.30)	

reached about 2.19 times higher in patients without hyperleukocytosis (HR=2.19; 95%CI 1.04 to 4.62; P=0.038).

Overall, the three-year survival of ALL in our setting was 33%, with a three-year survival rate of 41.7% in the ALL-2006 group and 30.72% in the ALL-2013 group (P=0.209). We were able to count only up to a three-year survival rate because not all patients in the ALL-2013 group had reached more than 3 years since the first diagnosis.

Discussion

This retrospective cohort study with survival analysis and Kaplan-Meier approach was conducted in ALL patients diagnosed from 2011 to 2018. Findings validated that age at diagnosis, leukocyte counts, and risk status were significantly correlated to mortality and remission. Subjects aged <1 year and >10 years had significantly higher mortality rate than those aged 1-10 years at time of diagnosis, with a mean survival time of 370.97 (80.48) days (P<0.001). A similar study in Surabaya, East Java, stated that patients aged 2 - 5 years had 2 times higher survival rate than those aged <2 years and >10 years.[3] Age at diagnosis was a significant prognostic factor, despite differences in immunophenotype and morphology. Almost 80% of patients under 1 year with ALL had a mixed-lineage leukemia gene accompanied by higher leukocyte counts and risk for CNS leukemia, which leads to lower survival rate under 1 year group. Lymphoblasts in older children are usually pluripotent lymphoid precursor cells, which progressively proliferate and are more resistant to chemotherapy.[4-7]

Mortality was also higher in patients with hyperleukocytosis (81.4%) compared patients without hyperleukocytosis (67.5%) (P<0.05), with mean survival time 622.64 (100.69) days in those with hyperleukocytosis. Similarly, a study in Dr. Sardjito Hospital, Yogyakarta, Central Java, reported that hyperleukocytosis increased the risk of mortality by 2 times.[8] Complications from hyperleukocytosis such as hyperviscosity, and tumor lysis syndrome, may lead to bleeding, respiratory distress, and thromboembolism, thus increasing the risk of mortality.[6,9,10]

We found that 84% of the high-risk status patients died, which was significantly higher than mortality in the standard-risk patients (57%). The mean survival times were 583.40 (70.75) days and 1,407.96 (139.81) days, respectively. Similar studies at Dr. Soetomo Hospital, Surabaya, East Java, and Dr. Sardjito Hospital, Yogyakarta, Central Java, also found that high risk status increased the risk of death about twice as high as standard risk.[3,11] *The National Cancer Institute* (NCI) classifies the prognosis of ALL into high risk and standard risk. Patients are considered to be high risk with leukocyte counts >50,000 mm^3, age <1 year or >10 years, mediastinal mass, CNS leukemia, or remission failure in the first week of induction phase.[4,12]

While 85.7% of patients with mature B-cell and T-cell died in our study, the difference in those with B-cell precursor was not significantly different (P>0.05), which was in contrast to a previous study which found lower mortality in B-cell compared to T-cell (HR 0.57; 95%CI 0.48 to 0.65).[13] Specifically, ALL was categorized by lineage and maturation of specific antigen, but unfortunately, 109 patients (47.4%) in our study did not have immunophenotyping done because it was unaffordable and not covered by *National Health Insurance*. Most deaths of both protocols occurred during induction phase (49%). Bleeding was the major cause of death

in both ALL-2006 (37%) and ALL-2013 (38,8%). In contrast, other studies in Surabaya (East Java) and in Vietnam reported that cause of death was infection related, with 76% and 43.2% respectively.[3,14]

Cox multivariate regression revealed that high risk status had significantly increased risk of death by 3 times (HR=3.0; 95%CI 1.9 to 4.9), and the ALL-2006 group had significantly lower risk by 0.3 times compared to the ALL-2013 group (HR=0.3; 95%CI 0.12 to 0.9). Similarly, a study in Yogyakarta (Central Java) found that even though leukocyte counts and age were statistically significant in univariate analysis, they were not significant in multivariate analysis.[15]

Seventy-three patients (39.5%) aged 1 to 10 years had remission compared to those aged <1 year or >10 years (P<0.001). Patients without hyperleukocytosis had significantly higher chance to reach remission by 2 times (P<0.05). A Brasilia study found that 73% of patients aged 1 to 9 years and leukocyte count less than 50,000/mm3 had remission.[16] A clinical review reported that children aged 1 year to less than 10 years without hyperleukocytosis had a higher chance of remission.[17]

In our study, standard risk patients had significantly increased chance of remission (51.4%) by 2.5 times, compared to high risk patients (P<0.001). Similarly, studies in Surabaya (East Java) and Jakarta found higher remission in the standard risk than in the high risk patients, with 81% and 87.1% achieving remission, respectively.[3,18] Our study had lower percentage of remission, probably because we analyzed remission at the end of maintenance phase for complete remission.

Univariate analysis revealed no significant factors for relapse, similar to a previous study by the *Nordic Society of Pediatric Hematology and Oncology* (NOPHO). They found that age at diagnosis, leukocyte count, morphology, immunophenotyping, and risk status were not associated with relapse, but cytogenic findings were.19 However, Cox multivariate regression revealed that high risk status significantly increased the risk of relapse by 3 times (P<0.05). Similarly, a Latin American study found that high risk status increased the risk of relapse by 2.47 times (95%CI 1.64-3.24; P<0.001).[20]

Marrow was the most common site of relapse (89.4%); intermediate relapse was the most common relapse time (53.2%). Higher relapse at maintenance phase may have been due to chemotherapy drug availability such as 6-mercaptopuine, because sometimes there were difficulties to get 6-MP during maintenance phase, which probably cause poor medication adherences. The 6-MP is an important component of ALL therapy regimens in maintenance phase and a predictor of relapse.[21]

A remarkable finding in our study was that both ALL-2006 and ALL-2013 protocols had high mortality rates (70.6% and 72.1%, respectively; P>0.05). Dr. Sardjito Hospital, Yogyakarta, reported that *ALL-2006 Protocol* had 0.95 times lower risk of death than the *ALL-2013 Protocol*, but the difference was also not significant (P>0.05).[11] Interestingly, the mean survival time of the ALL-2013 group was shorter, at 764.23 (63.49) days compared to 1,182.15 (176.89) days in the ALL-2006 group.

Remission was not significantly different between protocol groups, with 39.2% remission for ALL-2006 and 33% for ALL-2013 (P>0.05). A previous study at Dharmais Hospital, Jakarta, also found no significant association between the two protocols (32% for ALL-2006 and 29% for ALL-2013; P>0.05) in terms of remission.[2] Relapse was higher in our ALL-2006 group [29.4%; with mean time to relapse of 1,521.73 (146.08) days] compared to the ALL-2013 group [17.9%; mean time to relapse 1,493.53 (84.29) days], but there was no significant difference (P>0.05).

One of the differences between the *ALL-2006* and *ALL-2013 Protocols* is the L-asparaginase (L-asp) dose at induction phase. The ALL-2013 protocol uses cumulatively higher doses. A study compared ALL treatments with and without L-asp, and showed higher remission in patients who had L-Asp (93%).[22] L-asparaginase is an anti-neoplastic enzyme that hydrolyzes L-asparagine, causing decreased levels of L-asparagine, thus depriving tumor cells of nutrition and impeding cancer cell proliferation. This enzyme can also disturb protein synthesis, leading to coagulation dysfunction manifested as bleeding and disseminated intravascular coagulation (DIC).[22,23]

We found that highest mortality in the ALL-2013 group (51.9%) occurred in induction phase, which was higher but not significantly different from the ALL-2006 group in the same chemotherapy phase (38.9%). The major cause of death in induction phase of the ALL-2013 group was bleeding, which was assumed to have been caused by higher doses of L-Asp.

Overall, the three-year survival rate of ALL patients was 33%, similar to other studies in Indonesia. Dharmais Hospital, Jakarta, and Dr. Sardjito Hospital, Yogyakarta, had survival rates of 38.1% and 56.1 (3.9)%, respectively.[11,24] The three-year survival rates of the two protocols were 41.7% (ALL-2006) 30.72% (ALL-2013). However, some developed countries reported long-term survival rates of 90%.25 Brasilia reached a 5-year survival rate of 72%.[15] This difference might have been due to the availability of supportive therapies, such as blood components for massive bleeding, isolation rooms for patients with infection, and hematopoietic stem cell transplantation (HSCT). Results from the NOPHO study showed that high risk patients who underwent HSCT had higher mean survival, from 25.0 (SD 6.0)% to 46.7 (SD 5.1)%.[19]

A limitation of this study was its retrospective design and not all the subjects had immunophenotyping results, but we ensured beside immunophenotyping results that only patients with complete data were included and all dates of important outcomes had been documented. In addition, we could only count the three-year survival rate, because not all ALL-2013 patients had reached more than 3 years after diagnosis. Unfortunately, none of our subjects had cytogenic examination to determine high hyperdiploidy for a better prognosis.

In conclusion, age at diagnosis <1 year and >10 years, hyperleukocytosis, and high-risk group are significantly correlated with higher mortality. Age at diagnosis 1 to 10 years, no hyperleukocytosis and standard-risk group are significantly correlated with higher remission. No variable is significantly correlated with relapse. However, from Cox-multivariate regression we find high-risk group had 3 times higher risk of mortality than standard-risk, leukocyte count less than 50.000/mm^3 has 2,19 times higher probability for remission, high-risk group has 2,96 times higher risk of relapse. The 3-year survival rate of ALL-2006 protocols is higher than that of ALL-2013 protocols but is not statiscally significant.

Conflict of Interest

None declared.

Funding Acknowledgement

The authors received no specific grants from any funding agency in the public, commercial, or not-for-profit sectors.

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