RESEARCH ARTICLE

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Peripheral Blood Neutrophil Nadir and Time to Platelet Recovery during Induction Chemotherapy: Predictors of Clinical Outcomes and Markers for Optimizing Induction Treatment Intensity in Acute Lymphoblastic Leukemia

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Abstract

Objective: To study the kinetics of blood count nadir and time to recovery and find its association with clinical outcomes in a cohort of Acute Lymphoblastic Leukemia (ALL). **Methods:** Data from 243 cases treated between January 2018 to December 2020 was retrospectively analysed. Along with baseline data, serial measures of peripheral blood counts, nadir, and time to partial and complete recovery of counts during course of induction chemotherapy were recorded. Post-induction Complete Remission (CR) status, Event-Free Survival (EFS) , and Overall Survival (OS) were recorded as clinical outcomes for analysis. **Results:** Median age was 15 (range,1-62) years. Immunophenotype was B-ALL in 71% (n=172), and T-ALL in 27% (n=66). Good steroid response (D8) was seen in 89%(n=216), CR in 79% (n=192), and induction mortality in 12% (n=29). Median neutrophil nadir was 0.06(0-0.49) *10^9/L and median day to nadir was D17. Median time to partial and complete platelet recovery was D18 and D25. Late neutrophil nadir (>D15) was independent predictor of refractory disease post-induction [OR=5.43 (95%CI 1.06-27.75)]. Late partial platelet recovery (>D22) was independent predictor of poorer EFS and OS [HR = 1.63 (95%CI 1.07-2.47)] and [HR = 1.5 (95% CI 1-2.4)] respectively. **Conclusion:** We found that a longer time to neutrophil nadir independently predicts refractory disease post-induction and late partial platelet recovery is an independent factor for poorer EFS and OS. Thus, Blood count kinetics as independent predictors of induction outcomes can provide a simple, easy-to-use tool for balancing toxicity-efficacy during induction therapy for ALL.

Keywords: Acute Lymphoblastic Leukemia- nadir- time to partial and complete recovery- neutrophil- platelets

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Introduction

Acute Lymphoblastic Leukemia (ALL) is a form of lymphoid malignancy that is more prevalent in children than in adults [1]. Treatment outcomes of both paediatric and adult ALL are better in developed countries when compared to low-middle income countries (LMICs) [2–12]. Possible factors leading to inferior outcomes in LMICs are socio-economic and resource constraints, scarcity of specialized treatment facilities, and suboptimal adherence to treatment protocols. Other possible factors include compromised treatment intensity due to frequent interruptions because of toxicities, patient default, and other factors [13]. Moreover, there is an increased incidence of treatment-related mortality due to neutropenic infections mostly in the induction phase [9,10].

The remission induction phase is the most intensive phase of chemotherapy where maintaining treatment intensity may be crucial to achieve the best outcomes. However, due to the unpredictability of blood count kinetics and recovery caused by a weekly drug schedule, it becomes difficult to anticipate periods of cytopenia that can lead to serious infections and other toxicities. This results in frequent and empirical adjustments in dosage and schedule of chemotherapy especially for myelosuppressive drugs to minimize the risk of cytopenia and infection. However, arbitrary treatment adjustments can lead to compromised efficacy [13,14].

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Understanding the fluctuations in blood counts during the induction phase can help in better prediction of toxicities and in adapting the dose intensity of weekly chemotherapy drug(s) in patients with/or at risk of serious infection. Blood counts also serve as indicators of the restoration of normal haematopoiesis, and remission status in a case of leukemia. Early bone marrow regeneration leads to a shorter time of peripheral blood count recovery which can be readily monitored and indicates the effectiveness of treatment. However, there is scant literature on kinetics of blood counts during the induction phase of ALL, and its association with treatment outcomes. The aim of the present study was to evaluate kinetics of blood count nadir, time to recovery and explore its association with clinical outcomes, of post-induction remission and survival, in a cohort of newly diagnosed ALL.

Materials and Methods

Patient Recruitment

This retrospective, observational study included 243 newly diagnosed ALL patients registered between January 2018 to December 2020 in a tertiary care centre in Southern India. During this study period, a total of 313 patients were registered of which following patients were excluded from analysis: (i) patients who didn't receive induction therapy (ii) patients who had already received ≥7 days of induction treatment outside our centre, (iii) patients with incomplete laboratory data, and (iv) cases of lymphoblastic lymphoma (LBL).

The patients were characterised into standard risk (SR) and high risk (HR) groups based on the National Cancer Institute (NCI) risk group criteria [15] Baseline demographic features, clinical characteristics, disease parameters, treatment details and timelines, toxicities, and outcomes were collected from medical case records. Due to the retrospective design of the study and the use of anonymized patient data, requirements for written informed consent were waived after the Institute Ethics Committee approval (letter no. JIP/IEC/2020/200). This research adhered to the ethical standards in line with the Declaration of Helsinki.

Treatment Protocol and Response assessment

Treatment protocol for ALL generally consists of multiagent chemotherapy in different schedules and combinations and is typically divided into three phases: induction, consolidation, and maintenance. Backbone of induction phase of treatment for ALL across various protocols consists of steroid, vincristine, asparaginase, anthracycline and prophylactic intrathecal methotrexate. As per department policy, Berlin, Frankfurt, Muenster, 95 protocol (BFM-95) [16] or Multi-center Protocol-841 (MCP-841) [17] was used for cases up to 25 years of age, and German Multicenter Study Group for Adult ALL (GMALL) [18] protocol for adults above 40 years. Starting in 2019, patients up to 18 years of age were treated according to the Indian Collaborative Childhood Leukemia (ICiCLe) protocol [19]. Different induction protocols with drugs and dosage schedules used in the study is defined in detail in Supplementary Table 1. During treatment course, available data on grade 3/4 toxicity was recorded. Standard supportive care as per department policy was given to patients for the management of febrile neutropenia and other toxicities.

Efficacy of therapy was assessed by Day 8 steroid response, Bone Marrow examination (morphological) on Day 35 ±5 days, and Minimal Residual Disease (MRD) assessment by flow cytometry either post-induction or consolidation. Prednisolone good responders (PGR) and Prednisolone poor responders (PPR) at Day 8 were defined by the presence of absolute blast count of <1000/µl or >1000/µl in the peripheral blood, respectively after 7 days of prednisolone prophase. Early response to steroids is a strong predictor of overall treatment success. CR (complete remission) was defined as the absence of blasts in the peripheral blood; <5% blasts in bone marrow at the end of induction phase (D35 ±5 days). MRD positivity was defined as >0.001 leukemic cells in the marrow assessed by flowcytometry and performed only from 2019 onwards, hence was not available for all patients. Induction mortality was taken as death from any cause from start of induction till Day 42.

Definitions for kinetics of Peripheral Blood counts: Nadir and Recovery

Patient records were reviewed, and the following data were collected; the date of commencing induction chemotherapy, serial measures of blood count up to Day 42 from start of induction, including total leucocyte counts (TLC), absolute lymphocyte count (ALC), absolute neutrophil counts (ANC) and platelet counts at various time points during the induction period. As a department policy, growth factors were not used during course of induction chemotherapy. The nadir counts, day to nadir, duration of the periods to partial and complete recovery of the three cell lines were calculated for each case based on recorded dates. The cut-offs for partial and complete recovery were defined as >3.5*10⁹/L and >4.5*10⁹/L for TLC; >1*10⁹/L and >1.5*10⁹/L for ANC; >50*10⁹/L and >100*10⁹/L for platelets respectively and complete recovery for ALC as>1*10⁹/L from previous studies reported in the literature [20–22]. For nadir counts and day to nadir, patients who reached an ANC of ≤0.5*10⁹/L and/or platelets of ≤50*10⁹/L were included in the analysis. Patients whose ANC was ≥0.5*10⁹/L and/or platelets of ≥50*10⁹/L throughout the course of induction were excluded from analysis of kinetics of nadir. For analysis of peripheral blood counts recovery, cut-off for partial and complete recovery are mentioned in Supplementary Table 2. Graphs were used to present the haematopoietic regeneration during induction chemotherapy achieved by the whole cohort. Patients who died were excluded from the association analysis.

Definitions of Survival Outcomes

Event-free Survival (EFS) was defined as the duration from start of induction-phase chemotherapy to occurrence of any event (refractory, relapse, or death from any cause) or to the final follow-up. Overall survival (OS) was defined as the duration from start of induction chemotherapy to death from any cause/ final follow-up. Data for survival

analysis were censored on December 31, 2021.

Statistical Analysis

Descriptive and inferential statistics was used to summarize the baseline data. Association of baseline parameters and the blood count kinetics were done with induction outcome (Remission vs Not in Remission) and survival outcomes. Variables were examined with chi-square test, Mann-Whitney test and Odd's Ratio. Receiver Operator Curve (ROC) was explored for all the blood counts and optimal cut-off were chosen for association with post-induction outcomes. The EFS and OS were estimated using the Kaplan Meier method and the impact of baseline and other variables was assessed using the log-rank test. Further, multivariate analysis was performed using the Logistic regression for induction outcome and Cox regression method for survival outcome. Variables with P-value <0.05 in the univariate analysis were included in the multivariate Logistic Regression model and Cox proportional hazards model. All tests were 2-sided and P-value <0.05 was considered to indicate significance. IBM SPSS Statistics for Windows, Version 20.0, IBM Corp, Armonk, New York, United States software was used for all statistical analyses.

Results

Baseline characteristics

During the three-year study period, 243 patients met the inclusion criteria. The median age was 15 years (range, 1-62). The immunophenotype was B-ALL in 172 (70.8%), T-ALL in 66 (27.2%), and MPAL in 5 (2%) patients. According to the NCI risk group criteria, patients were classified as High Risk [69% (n=168)], and Standard Risk [31% (n=75)]. At baseline, median TLC was 13.4 *10⁹/L (range, 0.11-797.1), median platelet was 35*10⁹/L (range, 3-612), and median peripheral blood blasts were 70% (range, 0-98). Distributions of baseline characteristics for these patients are presented in Table 1.

Treatment course and Outcomes

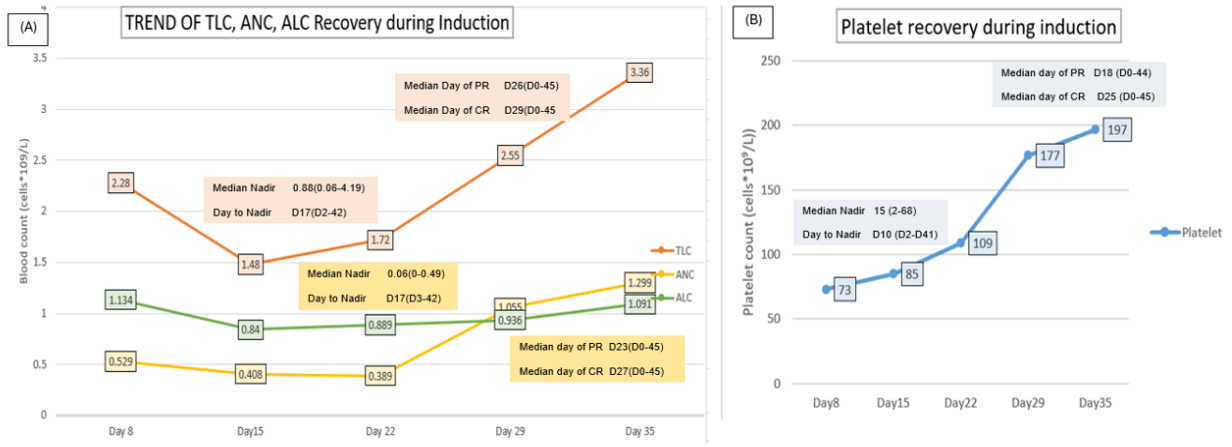
Majority of the patients were treated with BFM-95 protocol (43%), followed by ICiCLe (25%), MCP841 (19%), and GMALL (13%). The major toxicities were tumour lysis syndrome, deep vein thrombosis and peripheral neuropathy. On day 8, 89% (n=192) of patients were good responders to prednisolone. At the end of induction, 79% (n=192) patients had achieved complete remission (CR), 9% (n=22) had refractory disease and 12% (n=29) had died. Infections and neutropenic sepsis were the most common cause of death in our study. MRD data was available in 85 patients, and it was positive in 19 patients (22%). Treatment related characteristics and outcomes are summarized in Table 1.

Kinetics of Peripheral Blood counts during Induction course

Figure 1 shows the changes in weekly peripheral blood counts (on day 8,15, 22, 29 and 35), nadir, time to nadir, time to partial and complete recovery during induction course for TLC, ANC, ALC and platelets. The median ANC nadir was 0.06*10⁹/L (range, 0-0.49) and the median day to nadir was Day 17. The median time to partial and complete recovery for ANC was Day 23 and Day 27 respectively. The median platelet nadir was 15*10⁹/L (range, 2-69) and the median day to nadir was

Table 1. Baseline Characteristics, Ttreatment Course, and Induction Outcomes of Newly Diagnosed ALL.

Feature	Category	Distribution N=243 N (%)
Baseline Characteristics		
Age (years) Median age (range) 15 (1-62 y)	<15 y	116 (47.7)
	15-39 y	100 (41.2)
	40+ y	27 (11.1)
Sex	Male	157 (64.6)
	Female	86 (35.4)
Diagnosis subtype	B-ALL	172 (70.8)
	T-ALL	66 (27.2)
	MPAL	5 (2.1)
Extramedullary involvement (N=22)	CNS	19 (86.4)
	Testis	3 (13.6)
NCI Risk Group	SR	75 (30.8)
	HR	168 (69.2)
TLC (cells*10⁹/L) Median TLC 13.4 (0.11-797.1)	<50*10⁹/L	175(72)
	>50*10⁹/L	68 (28)
Platelets (cells*10⁹/L) Median (range) 35 (3-612*10⁹/L)	<20*10⁹/L	84 (34.6)
	21–50*10⁹/L	73 (30)
	>51*10⁹/L	86 (35.4)
Hemoglobin (g/dL)	Median	6.9 (1.8-15.40)
PS Blasts Median blast% (range) 70% (0-98%)	0-20%	72 (29.6)
	21-50%	27 (11.1)
	51-100%	144 (59.3)
Philadelphia Chromosome (N=217)	Normal	192 (88.5)
	Mutated	25 (11.5)
Treatment Characteristics		
Protocol Used	MCP841	46 (18.9)
	BFM95	104 (42.8)
	GMALL	32 (13.2)
	ICICLE	61 (25.1)
Intensity (all drugs)	100 % (maintained)	140 (55)
	<100%(not maintained)	103 (44)
Major Toxicities N=72	Tumour Lysis Syndrome	21 (8)
	Deep Vein Thrombosis	17 (6)
	Peripheral Neuropathy	15 (6)
	Paralytic ileus	9 (3)
	Pancreatitis	10 (3)
Fever in induction	Present	131 (49)
Induction Outcomes		
Day 8 Steroid response	Poor prednisone response	27 (11.1)
	Good prednisone response	216 (88.9)
Post Induction Outcomes	Remission	192 (79)
	Death in induction	29 (11.9)
	Refractory Disease	22 (9.1)
MRD N=85	Positive	19 (22.4)
	Negative	66 (77.6)



(C)

DAY (min-max)	TLC (cells*10⁹/L)	ANC (cells*10⁹/L)	ALC (cells*10⁹/L)	Platelets (cells*10⁹/L)
Day 8	2.28(0.05-351)	0.529(0-14.15)	1.134(0-12.14)	73(2-550)
Day15	1.48(0.12-77.1)	0.408(0-14.63)	0.84(0.016-76.24)	85(4-614)
Day 22	1.72(0.11-16.86)	0.389(0-12.12)	0.889(0-6.40)	109(8-640)
Day 29	2.55(0.06-26.35)	1.055(0-22.66)	0.936(0.05-9.37)	177(10-1092)
Day 35	3.36(0.49-23.76)	1.299(0-17.85)	1.091(0.13-5.86)	197(130-1160)

Figure 1. Trend of Peripheral Blood Nadir Count and Recovery during Induction Therapy of Patients with ALL (N=243). (A) Shows the trends of TLC, ANC, and ALC recovery during induction. (B) Shows the trend of platelet recovery during induction. (C) Shows the median (range) values for each cell count on days 8,15, 22, 29 and 35. *Total Leukocyte Count, Absolute Lymphocyte Count, and Absolute Neutrophil Count in cells*109/L.

Day 10. The median time to partial and complete recovery for platelets was Day 18 and Day 25 respectively.

Predictors of Refractory disease post-induction

Amongst the baseline patient characteristics, higher odds of refractory disease [OR= 9(95%CI 2.5-31.8); p<0.001)] was observed in patients with age 15-39 years as compared to paediatric patients (age<14 years). Similarly, patients with poor steroid response at D8 had higher odds of refractory disease, [OR=9.8(95%CI 3.6-26.4; p<0.001)]

Table 2. Multivariate Analysis for Factors Predictive of Refractory Disease Post-Induction

*Parameters	n	CR	No CR	Chi-square p-value	Univariate analysis	Multivariate analysis
Total	214	N (%)	N (%)		O. R (95% CI)	O. R (95% CI)
		192 (90)	22 (10)			
Age (yrs)						
0-14	111	108 (97)	3 (3)			
15-39	85	68 (80)	17 (20)	<0.001	9 (2.54-31.86)	7.34 (1.55-34.77)
40-70	18	16 (89)	2 (11)		4.5 (0.69-29)	1.15 (0.04-31.9)
Protocol						
MCP 841	44	43 (98)	1 (2)			
BFM 95	92	77 (84)	15 (16)		8.37 (1.06-65.6)	2.15 (0.22-20.5)
GMALL	19	15 (79)	4 (21)	0.007	11.5 (1.2-110.3)	22.4 (0.8-624)
ICICLE	59	57 (97)	2 (3)		1.5 (0.13-17.18)	0.87 (0.06-11.77)
D8 response						
Good PR	189	177(94)	12 (6)			
Poor PR	25	15 (60)	10 (40)	<0.001	9.83 (3.65-26.5)	8.11 (2.24-29.39)
Early ANC nadir (by D15)	76	72 (95)	4 (5)			
vs late ANC nadir(>D15)	97	81 (84)	16 (16)	0.03	3.57(1.16-10.9)	5.25 (1.04-26.53)
Platelet Partial Recovery						
Early (D22)	130	122 (94)	8 (6)			
Late (>D22)	77	64 (83)	13 (17)	0.013	2.89(1.17-7.11)	1.26 (0.33-4.82)

Univariate analysis for baseline factors predictive of Refractory disease post-induction is shown in supplementary table 1. Factors found significant in univariate analysis were taken for multivariate analysis. * Abbreviations: CR, complete remission; OR, odds ratio; CI, confidence interval; PR, prednisolone response; ANC, Absolute Netrophil Count

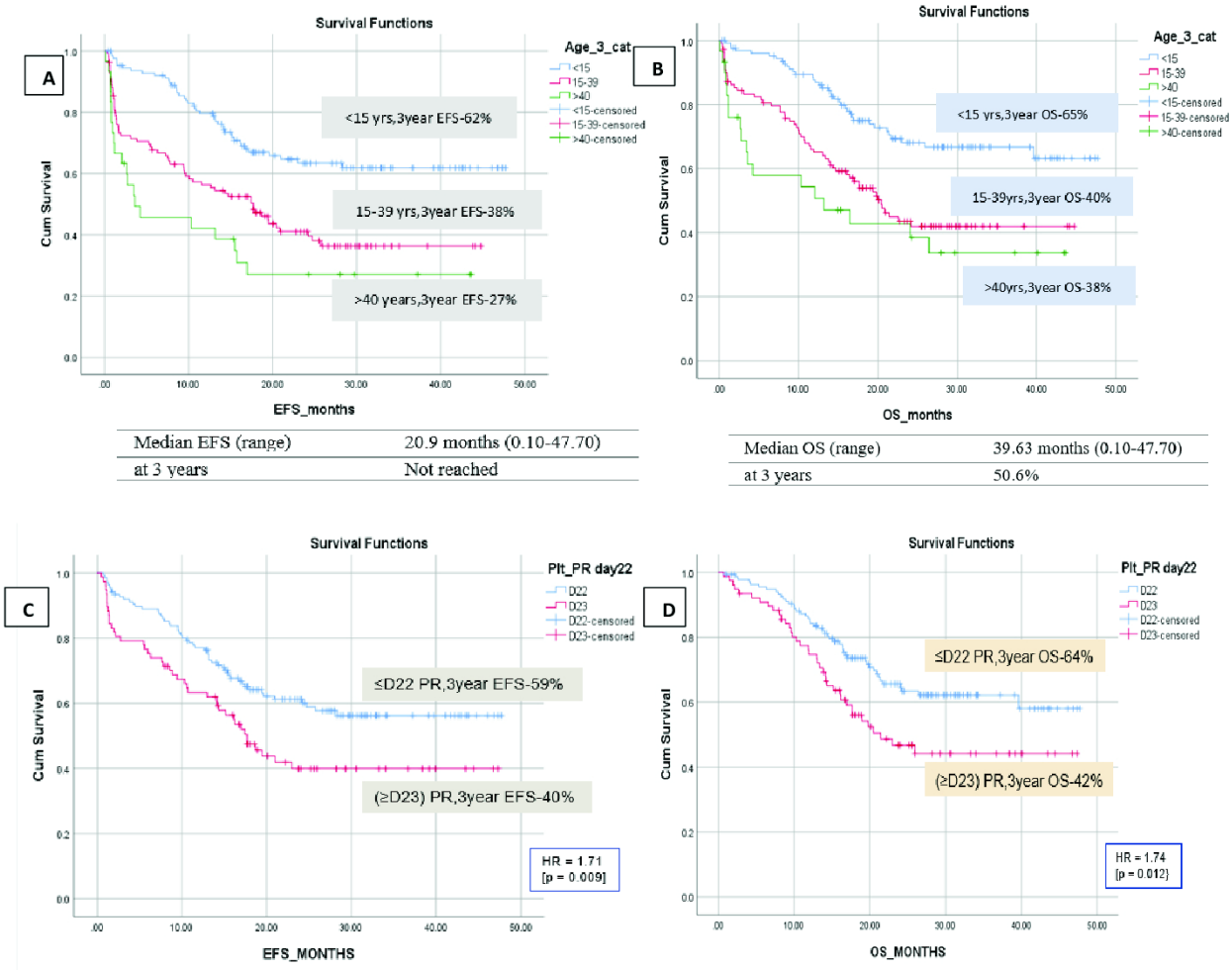


Figure 2. Kaplan-Meier Survival Curve (A) Event-free survival, (B) Overall survival for the entire cohort showing the difference in survival outcomes between age groups (pediatric, adolescent and young adults, and adults); (C) Event-free survival and (D) Overall survival showing the difference in survival outcome with relation to time to partial recovery of platelet counts (early vs late).

vs good steroids responders. Other baseline characteristics were not significantly associated with outcomes.

In relation to neutrophil kinetics, patients with late ANC nadir (>D15) had higher odds of refractory disease

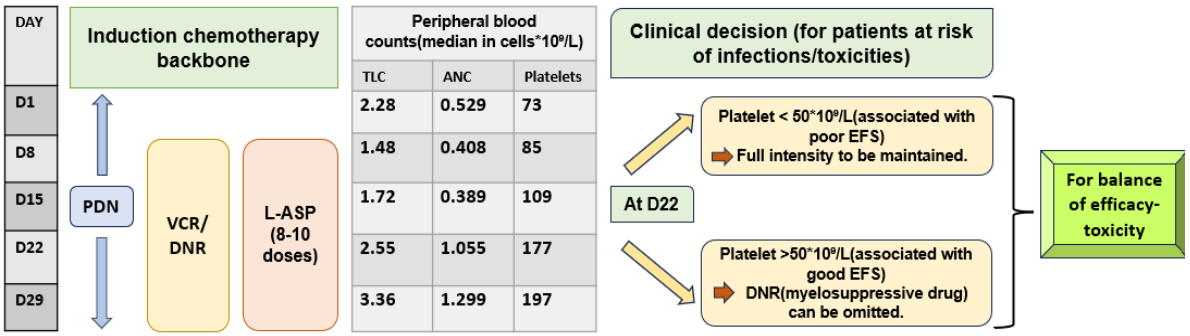


Figure 3. Application of Blood Count Kinetics in Clinical Decision Making for Weekly drug Schedule during ALL Induction is Explained with an Example. (A) Induction chemotherapy backbone of various protocols used. (B) Median Peripheral Blood (PB) counts observed in the study. (C) Utilization of PB count for clinical decision making. In patients, on intermediate or high-risk induction regimen, who have or are at risk of serious infections/toxicities (with or without neutrophil of < $0.5*10^9$/L beyond d15), D22 platelet count can guide the decision on treatment intensity and dose modifications of weekly schedule of DNR on D22 and D29. If the D22 platelet count is < $50*10^9$/L which is a predictor of poor EFS, full intensity of chemotherapy should be preferably maintained. For patients with D22 platelet count > $50*10^9$/L, a marker of good EFS, chemotherapy can be de-escalated to minimize risk of toxicity and infection related late induction mortality. However, the final decision has to be taken by considering the overall clinical status of the patient. For patients who are stable and/or with recovering neutrophils, treatment intensity should be maintained. Abbreviations: PDN, prednisolone; VCR, vincristine; DNR, daunorubicin; L-ASP: L-asparaginase; TLC: total leucocyte count; ANC: absolute neutrophil count.

Table 3. Factors Predicting Event Free Survival (EFS) and Overall Survival (OS) (Cox Regression Analysis for the Entire Cohort).

Parameters		N= 243	Median EFS (months)	3-year EFS(%)	EFS univariate HR (95%CI)	EFS multivariate HR (95%CI)	Median OS (months)	3-year OS (%)	OS univariate HR (95%CI)	OS multivariate HR (95%CI)
Age (years)	0-14	116	Not reached	61.8			Not reached	63.2		
	15-39	100	14.7(8.1-21.2)	36.4	2.19 (1.49-3.23)	2.01(1.30-3.09)	19	42	2.16(1.42-3.29)	1.9 (1.2-3.0)
	40-70	27	10.3(0-27.2)	27	3.36 (1.59-5.68)	2.14(0.98-4.66)	10.3	33.7	2.60(1.44-4.68)	2.2(1-4.7)
					1.22 (0.85-1.75)					
Sex	Male	157	22.9	49.2			26.4	50		
	Female	86	18.6(13.4-24)	43.9			19.7(12.1-28.6)	43.8	1.35(0.91-1.99)	
					1.59 (1.11-2.29)					
ECOG	0-1	171	24.2	50.9			26.4	49.5		
	>2	72	15.1(6.6-23.6)	38.9		1.22(0.79-1.92)	17.7(10-30)	44	1.49(0.99-2.23)	1.1(0.7-1.8)
Diagnosis	B-ALL	172	21	47.8			52.1	48.8		
	T-ALL	66	18.6(12.8-24.4)	46.9	1.24 (0.84-1.84)		21.1	45.9	1.135(0.71-1.81)	
	Negative	192	21	47.1			26.4	48		
Ph+ALL	Positive	25	10.3(6.5-19.4)	25.2	1.82 (1.07-3.09)	0.74(0.37-1.47)	12.9(6.4-19.4)	37.2	1.80(1-3.1)	
TLC	<50k	175	28.3	51			39.6	53.8		
	>50k	68	13.1(9.1-17.2)	30.3	1.71 (1.18-2.48)	1.51(0.99-2.31)	19.4(12.6-26.2)	26.4	2.105(1.35-3.26)	1.8(1.1-2.8)
Blasts	0-20%	64	Not reached	67.3			Not reached	72.7		
	21-50%	24	22.9	22	1.53(0.7-3.2)		24(17.6-30.4)	44.3	1.9(0.9-4.1)	
	>51 %	126	19.4(13-26)	19.4	2.0(1.2-3.4)		26.4(10.2-42.6)	44.1	2.2(1.2-3.9)	
D8 Res	PPR	216	22.9(0.1-47.7)	47.7			10.7	31		
	GPR	27	4.4(0-11.3)	25.9	2.36 (1.46-3.80)	2.70(1.58-4.63)	26.4	50	2.537(1.47-4.37)	2.3(1.3-4.1)
MRD Status of IA	Negative	66	Not reached	65.6			Not reached	65.2		
	Positive	19	16.9	49.3	1.65 (0.75-3.65)		Not reached	59.5	1.25(0.52-3.0)	
Platelet partial recovery	Early (D22)	130		56.2				62.2		
	Late (D23+)	77	17.7(13.7-21.6)	39.9	1.71(1.14-2.55)	1.63(1.07-2.47)	21.37(13-29)	44.1	1.74(1.13-2.67)	1.5 (1-2.4)

*Factors significant in univariate analysis were taken for multivariate regression.*Abbreviations- EFS, event free survival; OS, overall survival; HR, hazard ratio; CI, confidence interval; ECOG, Eastern Cooperative Oncology Group; TLC, total leucocyte count; MRD, minimal residual disease

[OR=3.57(95%CI 1.16-10.9; p=0.026)]. In relation to platelet kinetics, higher odds of refractory disease were observed in patients with late partial recovery of platelets (>D22) [OR=2.88(95%CI 1.17-7.11; p=0.021)]. In multivariate analysis, factors independently predictive of refractory disease were age 15-39 years [OR=7.12(95%CI 1.54-32.84; p=0.012)], poor day 8 steroid response [OR=8.11(95%CI 2.36-27.81; p=0.001)], and late ANC nadir (>D15) [OR=5.43 (95%CI 1.06-27.75; p=0.042)]. Table 2 shows the baseline parameters and blood count kinetics that were taken for multivariate analysis for prediction of refractory disease post-induction. Supplementary Table 3 shows the univariate results of baseline factors and blood counts for predicting induction outcomes.

Survival Outcomes

The median follow up was 27.57 months. For the entire cohort, median EFS was 24.2 months and three years EFS was 47.2%; median OS was not reached, and three years OS was 50.6%.

Predictors of Survival Outcomes

Univariate analysis of baseline patient characteristics and kinetics of blood counts for predicting EFS and OS is shown in Table 3. Factors predicting poorer EFS and OS were older age, ECOG status (≥2), Ph positivity, higher TLC, poor D8 steroid response and late partial recovery of platelet counts (>D22).

As shown in Table 3, independent predictors of EFS and OS in multivariate analysis were older age, higher TLC, poor d8 steroid response, and late partial platelet recovery(>D22) with HR = 1.63 [p = 0.020] for EFS and HR = 1.5 [p = 0.044] for OS respectively for late platelet recovery. Figure 2 shows the Kaplan-Meier survival curves for EFS and OS with relation to age group and time to partial recovery of platelets (early vs late).

Discussion

Treatment response and survival outcomes in Acute Lymphoblastic Leukemia (ALL) are determined by baseline patient and disease characteristics, and the treatment course [4,10]. In resource limited settings, cytopenias lead to frequent treatment interruptions, and higher mortality rates commonly due to infections and sepsis during the induction phase of ALL. Nevertheless, blood count kinetics can be potential markers of chemotherapy efficacy and bone marrow recovery. In this study, we have evaluated the kinetics of blood count, nadir, and recovery during induction and identified kinetics of neutrophil and platelets as independent predictors of refractory disease and worse survival in ALL. We propose that monitoring of blood count kinetics can help in rational tailoring of treatment to maintain an efficacy-toxicity balance and thereby in reducing treatment related morbidity and mortality during course of induction.

The median age of patients in our study was 15 (range, 1-62) years. Immunophenotype was B-ALL in 71%, followed by T-ALL in 27%. These numbers align with other studies conducted in India that report T-ALL in 21-43% cases [7,23,24], relatively higher than western data (25). During treatment we observed d8 good steroid response in 89% and post-induction complete remission (CR) was attained in 79% patients. The induction mortality in our study was 12%(n=29), which is higher than the rates reported in western multicentre studies (ranging from 1.1% to 2.2%) [26,27] and Indian case series (2% to 7%) [7,10,23,24]. We observed a higher percentage of late induction mortality of 80% (n=23/29) in week 3 or 4 compared to early mortality of 20% (n=6/29) within 2 weeks of start of induction. Infections and neutropenic sepsis were the most common cause of death in our study.

Understanding the nadir and recovery in blood counts during induction can potentially predict the disease control status in the marrow, help in adapting weekly course of induction therapy according to the kinetics of blood counts, and help in anticipation and management of toxicities of treatment. However, there is very limited data on the descriptive trends and weekly fluctuations of blood counts during induction therapy of ALL [21,22,28,29]. In our study, the median time to partial and complete recovery for ANC was Day 23 and Day 27 respectively and for platelets was Day 18 and Day 25 respectively. A study by Grunnan et al [22] in paediatric ALL reported relatively earlier complete recovery of both ANC and platelets at Day 26 and Day 16 respectively. The difference in kinetics of count recovery can be multi-factorial depending on the age, presenting baseline counts, risk group, specific population characteristics, treatment course, infections and toxicities and because of variations in the definition of recovery timeline.

We found late ANC nadir (>D15) as an independent factor predicting refractory disease along with baseline factors of age, type of protocol, and d8 response. There is no available data describing nadir of blood counts during induction therapy in literature. Also, there are very limited studies that have evaluated blood count kinetics as predictors of remission. In a study conducted by Nasution et al, trend of early ANC recovery (>500/mm3) by 3rd week of induction was correlated with higher chances of remission (with a r=0.687 and p-value of 0.001).[21]. Thus, our study provides nadir in neutrophil count as an easy to use, non-invasive early marker of bone marrow disease status which can serve as a potential surrogate for D14 bone marrow assessment. Bone marrow exam at D14 is described in literature for early assessment of disease status and consequent escalation of treatment for non-responders [30], but it's an invasive procedure with challenges in result interpretation and hence not widely practiced.

In our study, late partial platelet recovery (>D22) was found to be independent predictor of both a poorer EFS and OS. Comparable results were found in a study by Faderl et al in adult ALL, where shorter time to platelet recovery was associated with better DFS and OS [28]. In another study by Wang et al, a cut-off of average of daily platelet amount increase (Ap) > 3.9×10^9/L (including both time to platelet recovery and pre-treatment platelet count) was independently associated with a longer EFS (RR = 3.468; 95%CI: 1.037–11.597, P = 0.043) in childhood ALL [29]. Thus, platelet recovery patterns during induction also offer

an early, simple, and cost-effective independent biomarker of long-term survival in ALL.

In summary we have shown that shorter time to ANC nadir is independently predictive of CR post-induction, and early platelet recovery is an independent factor for longer EFS and OS. In a resource limited setting, where infection related mortality is high mostly during the latter half of induction (we observed induction mortality of 12%, of which 80% was beyond d15), blood count kinetics of early ANC nadir and platelet recovery can provide a useful and reliable tool to clinician. It can help to de-intensify the therapy, especially in week 3 or 4 of induction in patients with, or at risk of serious infection without the concern for compromising efficacy. Thus, toxicity-adapted dosing of the weekly (especially myelosuppressive) drugs based on ANC and platelet kinetics can help in reducing late toxicities and infection related mortality during induction chemotherapy. However, intensity of consolidation treatment beyond induction must follow the protocol specific risk-stratification criteria and MRD status. Application of blood count kinetics in clinical decision making for weekly drug schedule during ALL induction is explained with an example in Figure 3.

The strength of our study is a large cohort of ALL patients of all age groups, and the description of the entire spectrum of blood count kinetics during induction course. Limitations of our study are heterogeneity of patient population and treatment protocol used. However, for this study only course and outcomes of induction were analysed as the backbone of induction treatment is similar across protocols. Other limitations are missing records, incomplete data on treatment intensity and toxicity, unavailability of data on therapeutic transfusions and the retrospective study design. Additionally, MRD data was available for a small number of patients and hence it was not analysed for survival prediction. Nonetheless, our study contributes to the limited data in literature on the kinetics of blood count nadir and recovery and its association with clinical outcomes during the course of ALL induction.

In conclusion, we conclude that nadir and recovery time for ANC and platelet are readily obtainable, simple, inexpensive, easy to interpret and independent markers of clinical outcomes in ALL. Blood count kinetics can help balance efficacy-toxicity during induction chemotherapy for ALL especially in resource-limited settings with high induction mortality rates. Further validation of kinetics of platelet recovery in larger cohort, in association with MRD can provide a composite, reliable and sensitive tool for prediction of survival and adaptation of treatment in ALL.

Author Contribution Statement

Study conceptualization & methodology: Smita Kayal, Ramya Ramesh and Vineet Aggarwal. Data collection and analysis: Smita Kayal, Ramya Ramesh and Vineet Aggarwal, Amit Choudhary, Rajath Govind, N.Sreekumaran Nair. Manuscript writing: Smita Kayal, Ramya Ramesh, Vineet Aggarwal, Debdatta Basu, Prasanth Ganesan, Amit Choudhary. Review and editing: Smita Kayal, Ramya Ramesh, Debdatta Basu, Prasanth Ganesan, N.Sreekumaran Nair, Biswajit Dubashi and Rajat Govind. Final approval of manuscript: by all authors.

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Ethics approval

Institute Ethics approval (JIPMER Ethics committee) was taken before commencement of the study (letter no. JIP/IEC/2020/200).

Consent to participate

Waiver of consent was granted for the retrospective data and analysis.

Availability of data & material

will be made available on request

Conflict of interest

The authors have no competing interests.

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