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Article

The Utilization of The Large Unstained Cell (LUC) Parameter in Lymphoid, Haematopoietic and Related Tissue's Malignant Neoplasms

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Abstract: Large unstained cells (LUC) is a differential count parameter reported by routine hematology analysis, and LUC percentages (LUC %) reflect active lymphocytes and peroxidase-negative cells. We aimed to evaluate the LUC % parameter in routine practice towards malignant neoplasms, stated or presumed to be primary, of lymphoid, haematopoietic, and related tissue. LUC analysis was performed with Siemens ADVIA® 2120 Hematology System. Data were obtained from Ankara Bilkent City Hospital's laboratory information system. A statistical difference in the LUC % data in the case of LUC % <4.5 and LUC % ≥4.5 among preliminary diagnoses was observed ($p < 0.001$). According to the Kruskal-Wallis test, a statistical difference was observed between preliminary diagnosis and LUC % values ($p < 0.001$). The One-way ANOVA test with Bonferroni correction was performed for post hoc multiple comparisons of the preliminary diagnosis among LUC%. LUC % was higher in Hodgkin Lymphoma patients than Myeloid leukaemia patients ($p = 0.002$). LUC % was higher in the Lymphoid leukaemia patients than in the patients with Hodgkin lymphoma ($p < 0.001$), Other and unspecified types of non-Hodgkin lymphoma ($p < 0.001$), Multiple myeloma and malignant plasma cell neoplasms ($p < 0.001$). LUC % was higher in patients with leukemia unspecified cell type than Hodgkin lymphoma ($p < 0.001$), Follicular lymphoma ($p < 0.001$), Non-follicular lymphoma ($p < 0.001$), Mature T/NK-cell lymphomas ($p < 0.001$), Other and unspecified types of non-Hodgkin lymphoma ($p < 0.001$), Malignant immunoproliferative diseases ($p < 0.001$), Multiple myeloma and malignant plasma cell neoplasms ($p < 0.001$), Lymphoid leukaemia ($p < 0.001$), Myeloid leukaemia ($p < 0.001$), Other leukaemias of specified cell type patients ($p < 0.001$). Prospective studies may be useful in assessing LUC%.

Keywords: large unstained cells (LUC); siemens ADVIA 2120; complete blood cell count (CBC); ICD-10; neoplasms

Introduction:

Laboratory tests, along with anamnesis and physical examination, are included in the diagnosis of hematological diseases. Complete blood cell count (CBC) is a commonly performed blood test. It presents hemoglobin, the count of platelets, the indices of the red blood cell, the count of white blood cells (WBC), and its differential, which provides information about the different subset percentages of WBC and their absolute numbers [1]. The assessment of hemoglobin, the indices of red blood cells, and hematocrit maintain knowledge concerning the patient's oxygen-carrying capacity while evaluating the WBC count ensures that information about the immune system is available. CBC has a role in diagnosing anemia, immune deficiencies, infection, acute hemorrhagic conditions, and some

types of cancer [2]. Hematological malignancies represent a heterogeneous group of tumors and constitute 6.5% of whole cancer types and 7.2% of cancer-related deaths according to the World Health Organization [3].

Large unstained cells (LUC), and LUC percentages (LUC %) are displayed on some automatic cell counters [4]. In a letter to the editor, it was stated that of the twenty-two doctors in the study, nineteen had no idea about the LUC abbreviation meaning, and seventeen did not know how to interpret high LUC results [5]. LUC is one of the differential count parameters reported during routine hematology analyses, and LUC % externalizes and peroxidase-negative cells and active lymphocytes [4]. LUC presents virocytes, hematopoietic stem cells, large lymphocytes, abnormal cells, and blasts [6]. An increment of LUC % may be observed in some infectious and hematological diseases [7].

Nosology is systematically sorting diseases. International Classification of Diseases (ICD) is among the most preferred nosologies. The ICD system was created to track diseases in the population accurately [8]. International Statistical Classification of Diseases and Related Health Problems 10th Revision (ICD-10) Version:2019 contains 22 chapters. The neoplasms take place in Chapter II. The diseases under the title of malignant neoplasms, stated or presumed to be primary, of lymphoid, haematopoietic, and related tissue are as follows: Hodgkin lymphoma, Follicular lymphoma, Non-follicular lymphoma, Mature T/NK-cell lymphomas, Other and unspecified types of non-Hodgkin lymphoma, Other specified types of T/NK-cell lymphoma, Malignant immunoproliferative diseases, Multiple myeloma and malignant plasma cell neoplasms, Lymphoid leukaemia, Myeloid leukaemia, Monocytic leukaemia, Other leukemias of specified cell type, Leukaemia of unspecified cell type, Other and unspecified malignant neoplasms of lymphoid, haematopoietic and related tissue [9].

The diagnoses placed in the laboratory information system of the Ankara Bilkent City Hospital are in line with ICD-10. The purpose of the present study was the assessment of the LUC % parameter in routine practice towards the codes of malignant neoplasms, stated or presumed to be primary, of lymphoid, haematopoietic, and related tissue to provide new data on LUC % in the literature.

Material and Methods:

In Ankara Bilkent City Hospital, CBC tests and WBC differentials are performed on the Siemens ADVIA® 2120 Hematology System (Siemens Healthineers, Erlangen, Germany) devices. Calibration and internal quality controls are performed daily on the device. After being deemed appropriate by biochemistry experts, analysis begins on the device. External quality controls are performed once a month.

The ADVIA® 2120 autoanalyzer analyzes whole blood samples which are obtained in Ethylenediaminetetraacetic acid tubes. The device uses flow cytometry as its basis for measurements. The device uses light scattering to ensure CBC, WBC differentials, reticulocyte counts, WBC lysis, myeloperoxidase, and oxazine 750 staining. It does not require cyanide to measure hemoglobin colorimetrically. WBCs are sorted into neutrophils, eosinophils, basophils, granulocyte monocytes, lymphocytes, and LUC [10].

The codes of ICD-10 consisting of the malignant neoplasms, stated or presumed to be primary, of lymphoid, haematopoietic, and related tissue between 01/02/2019 and 29/02/2020, were acquired from the laboratory information system of the hospital. In this retrospective study, only each patient's first result and preliminary diagnosis were included.

The LUC% data were evaluated according to gender and age intervals: pediatric group (0–18 years), adult group (19–64 years), and geriatric group (65 years and over).

Statistical Analyses

Categorical numerical variables were stated as percentages, and the descriptive statistics data were stated as median and interquartile ranges (IQR). Kruskal Wallis, chi-square tests, and Post Hoc ANOVA tests were carried out to compare data among the groups. A p-value <0.05 was accepted as

statistically expressive. All the statistical analyses were performed with IBM SPSS Statistics for Windows, Version 27.0 (IBM Corp., Armonk, NY, USA).

Results:

1710 LUC % data were rolled in the present study. Quantitative variables distribution is presented in Table 1. Quantitative data among genders is presented in Table 2. Quantitative data among age intervals is presented in Table 3. LUC % data among preliminary diagnoses is presented in Table 4.

Figure 1 presents LUC % values among preliminary diagnoses with error bars. The Median and IQR levels of LUC % among preliminary diagnoses are presented in Table 5.

Table 1. Quantitative variables distribution.

	N	%
Gender		
Female	853	49.9
Male	857	50.1
Age (years)		
0-18	238	13.9
19-64	854	49.9
65 and over	618	36.1
LUC % Data		
LUC % <4.5	1459	85.3
LUC % ≥4.5	251	14.7
Preliminary Diagnosis		
Hodgkin lymphoma	201	11.8
Follicular lymphoma	46	2.7
Non-follicular lymphoma	116	6.8
Mature T/NK-cell lymphomas	36	2.1
Other and unspecified types of non-Hodgkin lymphoma	251	14.7
Malignant immunoproliferative diseases	9	0.5
Multiple myeloma and malignant plasma cell neoplasms	226	13.2
Lymphoid leukaemia	507	29.6
Myeloid leukaemia	279	16.3
Other leukaemias of specified cell type	19	1.1
Leukaemia of unspecified cell type	17	1
Other and unspecified malignant neoplasms of lymphoid, haematopoietic and related tissue	3	0.2

LUC: Large Unstained Cells.

Table 2. Quantitative Data Among Genders.

		Female		Male		p*
		N	%	N	%	
Age (years)						
0-18		105	12.3	133	15.5	<0.001
18-64		400	46.9	454	53.0	
	65 and over	348	40.8	270	31.5	
LUC % Data						
LUC % <4.5		712	83.47	747	87.2	0.031
LUC % ≥4.5		141	16.53	110	12.8	
Preliminary Diagnosis						
Hodgkin lymphoma		84	9.8	117	13.7	<0.001

Follicular lymphoma	28	3.3	18	2.1
Non-follicular lymphoma	63	7.4	53	6.2
Mature T/NK-cell lymphomas	25	2.9	11	1.3
Other and unspecified types of non-Hodgkin lymphoma	107	12.5	144	16.8
Malignant immunoproliferative diseases	3	0.4	6	0.7
Multiple myeloma and malignant plasma cell neoplasms	117	13.7	109	12.7
Lymphoid leukaemia	253	29.7	254	29.6
Myeloid leukaemia	152	17.8	127	14.8
Other leukaemias of specified cell type	15	1.8	4	0.5
Leukaemia of unspecified cell type	4	0.5	13	1.5
Other and unspecified malignant neoplasms of lymphoid, haematopoietic and related tissue	2	0.2	1	0.1

p*: Chi-squared test. LUC: Large Unstained Cells.

Table 3. The Quantitative Data Among Age Intervals.

	Age intervals						p*
	0-18		18-64		65 and over		
	N	%	N	%	N	%	
LUC % Data							
LUC % <4.5	205	86.1	766	89.7	488	79.0	p<0.001
LUC % ≥4.5	33	13.9	88	10.3	130	21.0	
Preliminary Diagnosis							
Hodgkin lymphoma	24	10.1	151	17.7	26	4.2	
Follicular lymphoma	4	1.7	21	2.5	21	3.4	
Non-follicular lymphoma	13	5.5	56	6.6	47	7.6	
Mature T/NK-cell lymphomas	1	0.4	24	2.8	11	1.8	
Other and unspecified types of non-Hodgkin lymphoma	10	4.2	128	15.0	113	18.3	
Malignant immunoproliferative diseases	0	0	4	0.5	5	0.8	
Multiple myeloma and malignant plasma cell neoplasms	0	0	84	9.8	142	23.0	p<0.001
Lymphoid leukaemia	175	73.5	200	23.4	132	21.4	
Myeloid leukaemia	7	2.9	175	20.5	97	15.7	
Other leukaemias of specified cell type	0	0	6	0.7	13	2.1	
Leukaemia of unspecified cell type	3	1.3	5	0.6	9	1.5	
Other and unspecified malignant neoplasms of lymphoid, haematopoietic and related tissue	1	0.4	0	0	2	0.3	

p*: Chi-squared test (Likelihood Ratio). LUC: Large Unstained Cells.

Table 4. The LUC % Data Among Preliminary Diagnosis.

Preliminary Diagnosis	LUC % Data				p*
	LUC % <4.5		LUC % ≥4.5		
	N	%	N	%	
Hodgkin lymphoma	198	13.6	3	1.2	p<0.001
Follicular lymphoma	45	3.1	1	0.4	
Non-follicular lymphoma	105	7.2	11	4.4	
Mature T/NK-cell lymphomas	36	2.5	0	0.0	
Other and unspecified types of non-Hodgkin lymphoma	230	15.8	21	8.4	
Malignant immunoproliferative diseases	9	0.6	0	0.0	
Multiple myeloma and malignant plasma cell neoplasms	210	14.4	16	6.4	

Lymphoid leukaemia	366	25.1	141	56.2
Myeloid leukaemia	241	16.5	38	15.1
Other leukaemias of specified cell type	10	0.7	9	3.6
Leukaemia of unspecified cell type	7	0.5	10	4.0
Other and unspecified malignant neoplasms of lymphoid, haematopoietic and related tissue	2	0.1	1	0.4

p*: Chi-squared test, LUC: Large Unstained Cells, N: Number of the population.

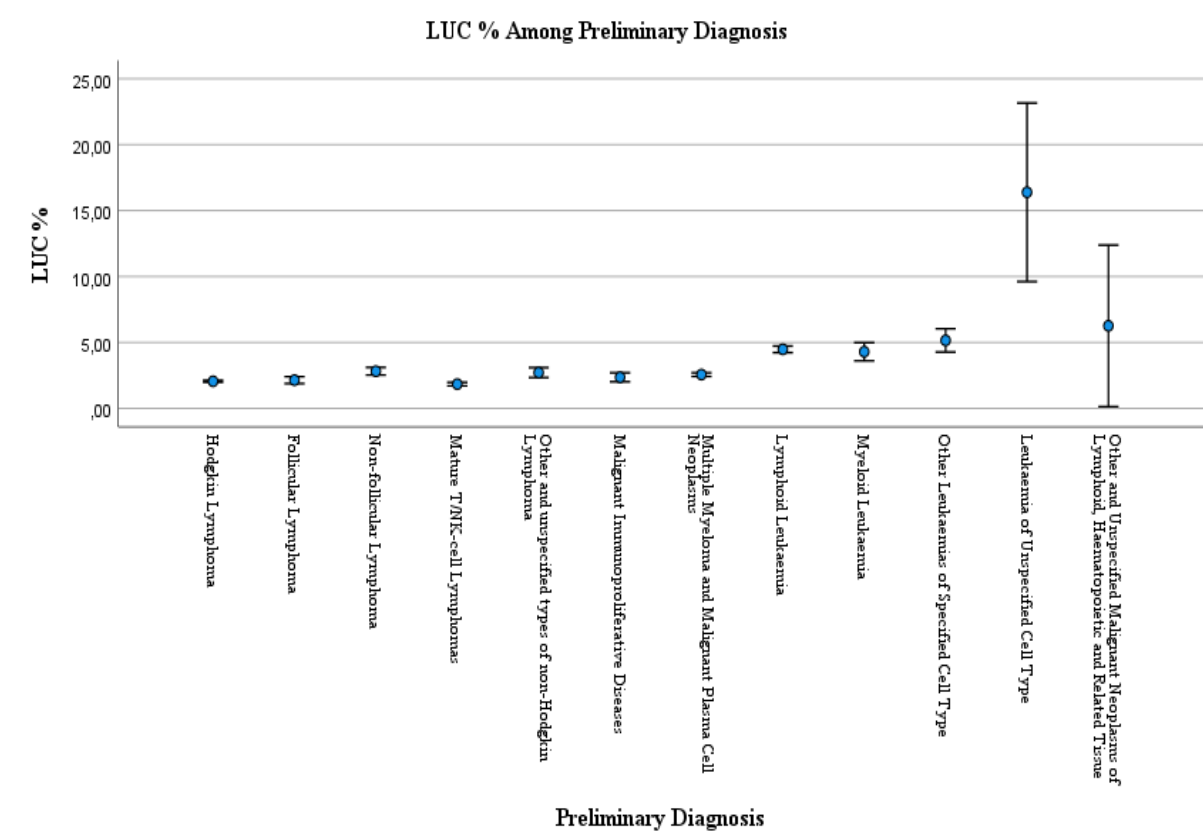


Figure 1. LUC % Among Preliminary Diagnosis.

Table 5. Median and Interquartile Values of LUC% Among Preliminary Diagnosis.

Preliminary Diagnosis	LUC %	
	Median	IQR
Hodgkin lymphoma	2	[1.5-2.5]
Follicular lymphoma	2.1	[1.3-2.6]
Non-follicular lymphoma	2.2	[1.7-2.9]
Mature T/NK-cell lymphomas	1.7	[1.4-2.1]
Other and unspecified types of non-Hodgkin lymphoma	1.9	[1.5-2.6]
Malignant immunoproliferative diseases	2.1	[1.9-2.75]
Multiple myeloma and malignant plasma cell neoplasms	2.2	[1.6-3.1]
Lymphoid leukaemia	2.7	[1.8-5]
Myeloid leukaemia	1.9	[1.5-2.8]
Other leukaemias of specified cell type	4.3	[2.6-6.4]
Leukaemia of unspecified cell type	7.5	[2.2-16.7]
Other and unspecified malignant neoplasms of lymphoid, haematopoietic and related tissue	2.6	[2.3-]

LUC: Large Unstained Cells, IQR: Interquartile Ranges.

A flag occurs in Siemens ADVIA 2120 device when LUC% data $\text{LUC\%} \geq 4.5$. In the present study, a statistical difference in the LUC % data in the case of $\text{LUC \%} < 4.5$ and $\text{LUC \%} \geq 4.5$ among preliminary diagnoses was observed ($p < 0.001$).

According to the Kruskal-Wallis test, a statistical difference was observed between preliminary diagnosis and LUC % values ($p < 0.001$). The one-way ANOVA test was performed with Bonferroni correction for post hoc multiple comparisons of the preliminary diagnosis among LUC%. A statistical difference in LUC% was observed between Hodgkin lymphoma and Myeloid leukaemia ($p = 0.002$).

Statistical differences of LUC % between Lymphoid leukaemia and the other diagnoses were as follows: Hodgkin lymphoma ($p < 0.001$), Other and unspecified types of non-Hodgkin lymphoma ($p < 0.001$), Multiple myeloma and malignant plasma cell neoplasms ($p < 0.001$). Statistical differences of LUC% between leukemia unspecified cell type and the other diagnoses were as follows: Hodgkin lymphoma ($p < 0.001$), Follicular lymphoma ($p < 0.001$), Non-follicular lymphoma ($p < 0.001$), Mature T/NK-cell lymphomas ($p < 0.001$), Other and unspecified types of non-Hodgkin lymphoma ($p < 0.001$), Malignant immunoproliferative diseases ($p < 0.001$), Multiple myeloma and malignant plasma cell neoplasms ($p < 0.001$), Lymphoid leukaemia ($p < 0.001$), Myeloid leukaemia ($p < 0.001$), Other leukaemias of specified cell type ($p < 0.001$).

Discussion:

As far as the authors know, the LUC % among malignant neoplasms, stated or presumed to be primary, of lymphoid, haematopoietic, and related tissue was evaluated retrospectively for the first time with a biochemistry laboratory perspective.

LUC parameters were evaluated in various hematological conditions. According to Merter et al., LUC may play a role in the timing of CD34 counting by flow cytometry in peripheral blood and also LUC may be used to predict the mobilization success [6].

In a study involving 118 acute leukemia patients, LUC values were found as follows; M1 leukemias had high proportions of LUC, M2 and M3 leukemias had low LUC values, M4 leukemias had large and M5 leukemias had very large numbers of LUC [11]. It has been suggested that LUC % could be useful for improving chronic B-cell leukemia diagnostic reproducibility [12].

It has been stated that adding the LUC parameter to the reflex review parameters in acute leukemia patients concluded in all acute leukemia patients' detection [13].

A LUC % value above 10 % may indicate an accelerated stage of chronic granulocytic leukemia [14]. In a study, that included 148 chronic B-lymphocytic leukemia patients with no treatment before, an association among LUC values and survival was found. It was stated that the increase in LUC numbers coincided with the worsening of the clinical condition [15].

In a study including 48 untreated and 30 treated patients whose diseases directly or indirectly affected hematopoiesis, the LUC value was found to be high in patients with refractory anemia with excess blasts or in transformation, and dys-myelopoietic syndrome while the LUC value was found to be low in patients with lymphoma [16]. The LUC rate is increased in myelodysplastic syndrome [17]. Investigating the cell suspensions stated that the LUC value was higher in non-Hodgkin's lymphoma patients and lower in patients with non-neoplastic lymphadenopathies [18].

In the present study, a statistical difference in the LUC % data in the case of $\text{LUC \%} < 4.5$ and $\text{LUC \%} \geq 4.5$ among preliminary diagnoses was observed ($p < 0.001$). LUC % was higher in patients with Hodgkin Lymphoma than in Myeloid leukaemia patients ($p = 0.002$).

LUC % was higher in the Lymphoid leukaemia patients than in the patients with Hodgkin lymphoma ($p < 0.001$), Other and unspecified types of non-Hodgkin lymphoma ($p < 0.001$), Multiple myeloma and malignant plasma cell neoplasms ($p < 0.001$).

LUC % was higher in patients with leukemia unspecified cell type than Hodgkin lymphoma ($p < 0.001$), Follicular lymphoma ($p < 0.001$), Non-follicular lymphoma ($p < 0.001$), Mature T/NK-cell lymphomas ($p < 0.001$), Other and unspecified types of non-Hodgkin lymphoma ($p < 0.001$), Malignant immunoproliferative diseases ($p < 0.001$), Multiple myeloma and malignant plasma cell neoplasms

($p < 0.001$), Lymphoid leukaemia ($p < 0.001$), Myeloid leukaemia ($p < 0.001$), Other leukaemias of specified cell type patients ($p < 0.001$).

Strengths and Limitations:

The strengths of the present study were as follows: Our study contains 13 months of data from one of the largest hospitals in Türkiye. Only the first result and preliminary diagnosis of each patient were included. LUC % values were analyzed according to gender, age, and preliminary diagnosis, and the results were presented.

Not only in hematologic diseases but also changes in LUC were observed in some different diseases. LUC is a valuable parameter that can help assess immune activation levels in HIV infection [19]. It has been noted that the LUC% value changes significantly in HIV-infected individuals and correlates with markers of immune activation and disease progression [20]. Shin et al. suggested that the % LUC parameter could be used to differentiate varicella caused by varicella zoster from Kaposi's varicelliform eruption and disseminated herpes zoster [21]. It has been stated that WBC, LUC, neutrophils, lymphocytes, and platelets bind to dengue virus-infected endothelial cells more than control group endothelial cells, and this increase in binding may be the cause of the neutropenia and thrombocytopenia that occur in dengue hemorrhagic fever [22]. In Nixon et al.'s study, as a result of 9000 whole blood analyses, 62 patients had high LUC values and were followed up for signs of viral infection. It was stated that 40 patients had viral infections, 26 of which were caused by the Epstein-Barr virus [23]. Changes in LUC may be due to medication. In a study conducted by Bononi et al., it was stated that the LUC value could be one of the biological indicators of leukocyte recovery after chemotherapy in the hematological response to the rHu-G-CSF medication [24].

In light of the literature, our study's limitation is that it did not include information about the patient's medication and additional diseases.

Conclusions:

In our study that evaluated the LUC% value retrospectively from a laboratory perspective some differences were observed in LUC % in lymphoid, haematopoietic, and related tissue's malignant neoplasms. Prospective studies including patient and control groups may be useful in assessing LUC%. The present study underscores the importance of LUC% in line with ICD-10 and may provide ideas for new research.

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