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DOI: 10.4103/ijh.ijh_31_22

Clinical significance of serum sCD23 and B-cell maturation antigen levels in patients with chronic lymphocytic leukemia

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Abstract:

BACKGROUND: Chronic lymphocytic leukemia (CLL) is a malignancy of mature appearing clonal B lymphocytes where there is a progressive accumulation of leukemic cells in peripheral blood, bone marrow, and secondary lymphoid tissues as a consequence of defective apoptosis and survival signals derived from the microenvironment. The soluble CD23 (sCD23) is a 25 kDa fragment that can be found in serum, plasma, and urine in patients with CLL. It is a B-cell growth factor. B-cell maturation antigen (BCMA) is a member of the tumor necrosis factor superfamily, it enhances the survival and proliferation of mature B cells and plasma cells through signal transduction of the B-cell activating factor and a proliferation-inducing ligand.

AIMS: The aims of this study were to assess the serum levels of sCD23 and BCMA in newly diagnosed CLL patients and to correlate them with clinical Binet staging and other hematological and clinical parameters.

PATIENTS, MATERIALS AND METHODS: This study was conducted on 54 newly diagnosed CLL patients and 27 healthy controls. Diagnosis of CLL patients was based on lymphocyte count of >5 × 10^9/L and immunophenotyping. The serum levels of sCD23 and BCMA were measured in both groups using an enzyme-linked immunosorbent assay.

RESULTS: Serum levels of sCD23 and BCMA were significantly higher in CLL patients in comparison with control group ($P < 0.001$ for both). There was a significant direct association between serum levels of sCD23 and BCMA with the clinical Binet stage of the disease ($P < 0.001$ for both). sCD23 showed significant correlation with hemoglobin (Hb) level ($P < 0.001$), total white blood cell (WBC) count ($P = 0.001$), lymphocyte count ($P < 0.001$), platelet count ($P = 0.017$), B-symptoms ($P = 0.001$), and splenomegaly ($P = 0.019$), whereas BCMA has significant correlations with Hb level, total WBC count, lymphocyte count ($P < 0.001$ for each one), B-symptoms ($P < 0.001$), lymphadenopathy ($P = 0.001$), splenomegaly ($P = 0.024$), and hepatomegaly ($P = 0.04$).

CONCLUSIONS: The levels of serum sCD23 and serum BCMA increase with advancing Binet stages of the disease indicating their possible usefulness as good and reliable parameters for prognostic evaluation in CLL patients. The significant correlation of serum sCD23 and serum BCMA with hematological parameters and clinical features render them as reliable tumor burden markers in CLL patients.

Keywords:

B-cell maturation antigen, Binet staging, chronic lymphocytic leukemia, sCD23

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Submission: 19-06-2022
Revised: 15-07-2022
Accepted: 22-07-2022
Published: 25-10-2022

Introduction

Chronic lymphocytic leukemia (CLL) is a heterogeneous disease biologically and clinically.[1] The diagnosis of CLL needs the

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How to cite this article: Ameen TM, Al-Rubaie HA. Clinical significance of serum sCD23 and B-cell maturation antigen levels in patients with chronic lymphocytic leukemia. Iraqi J Hematol 2022;11:134-8.

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presence of at least 5×10^9/L circulating B lymphocytes, with clonality demonstrated by flow cytometry according to the international workshop on CLL criteria.[2]

Soluble factors, whether originated from the neoplastic cells (autocrine) or the surrounding cells in the microenvironment (paracrine), were assumed to play a substantial role in the growth mechanism of the neoplastic clone.[3]

The CD23 antigen, a low-affinity receptor for Immunoglobulin E (IgE), is a 45 kDa Type I1 membrane glycoprotein. It is found on the surface of Ig M bearing B cells, eosinophils, macrophages, and some T and NK cells and is cleaved into soluble fragments (soluble CD23, sCD23) of various sizes displaying pleiotropic biological activities. The sCD23 is a 25 kDa fragment that can be found in serum, plasma, and urine in patients with CLL. It has been shown to be the B-cell growth factor. In patients with CLL serum levels of sCD23 are highly elevated (3- to 500-fold) in comparison with its levels in healthy controls. Moreover, among CLL patients, higher levels of serum sCD23 correlate with more advanced disease stage, rapid, and shorter doubling time of lymphocytes, diffuse bone marrow infiltration, and poorer prognosis in terms of expected survival time.[4,5] Therefore, sCD23 reflects both the bulk of the disease and its kinetics providing a valuable prognostic information.

B-cell maturation antigen (BCMA) is a member of the tumor necrosis factor superfamily, it is also known as tumor necrosis factor receptor superfamily member 17 (TNFRSF17), it enhances the survival and proliferation of mature B cells and plasma cells through signal transduction of the B cell activating factor (BAFF/BLys) and a proliferation-inducing ligand (APRIL).[6] BCMA is expressed on the cell surface of mature and malignant B lymphocytes. Previous studies demonstrated that patients with multiple myeloma have increased levels of serum BCMA compared with individuals with monoclonal gammopathy of undetermined significance and healthy controls.[7] Serum BCMA levels are higher in patients with CLL compared with healthy individuals and changes in BCMA levels correlate with patients' clinical status.[8] This study aims to assess the serum levels of sCD23 and BCMA in newly diagnosed CLL patients and to correlate them with clinical Binet stages and with other hematological and clinical parameters.

Patients, Materials, and Methods

This prospective study was conducted on 54 newly diagnosed adult CLL patients attending the Hematology outpatient clinic in Medical City Complex, Baghdad. Data were collected for each patient including name, age, sex, the presence of B symptoms, lymphadenopathy (LAP), splenomegaly, and hepatomegaly. A clinical Binet staging system was used. All patients had absolute lymphocyte count (ALC) >5×10^9/L with CLL score >3. Twenty-seven healthy age-and sex-matched individuals were involved in this study as a control group to determine the median levels of sCD23 and BCMA.

The measurement of serum sCD23 was done using sandwich-enzyme-linked immunosorbent assay (ELISA) kit polyclonal anti-sCD23 antibody (Human FcεRII/CD23, Receptor II for the Fc Region of Ig E, E = EL-H0034, Elabscience Biotechnology Co., China). The measurement of serum BCMA was done using a sandwich ELISA kit polyclonal anti-BCMA antibody (Human TNFRSF17 member 17, E6650Hu, BT LAB, China). This study was approved by the review ethical committee of Iraqi council for medical specialization. All patients were given their written informed consent prior to this study.

Statistical analysis

Data were collected, summarized, analyzed, and presented using statistical package for social sciences (SPSS) version 16 (SPSS, IBM, Chicago, USA) and Microsoft Office Excel 2007. Nominal variables were expressed as frequency (number) and percentage. The continuous variables were presented as mean, standard deviations (SD), range, median, and interquartile range (IQR) accordingly. Mann–Whitney *U*-test was used to evaluate the difference in mean of numeric variables between any two groups because variables were not normally distributed. Kruskal–Wallis test was used to evaluate the difference in mean of numeric variables among more than two groups, followed by *post hoc* Dunn's multiple comparison test, capital letters were used so that different letters indicate significant difference; whereas, similar letters indicate no significant difference. Pearson's Chi-square was used to assess the association between the categorical data. Spearman's rho test was used to predict correlation between the parameters of the patients. The level of significance was considered at $P \leq 0.05$. The level of high significance was considered at $P \leq 0.01$.

Results

The current study included 31 male and 23 female patients, with male-to-female ratio of 1.3:1. The mean (± SD) age was 59.42 ± 11.75 years and a range of 23–80 years and a median of 61 years.

Out of the 54 patients, 61.1% (33) were in Stage A, 13% (7) in Stage B, and 25.9% (14) in stage C.

At presentation, 13 patients (24.1%) were diagnosed incidentally, while 41 (75.9%) patients were symptomatic. B-symptoms, LAP, and splenomegaly were the most

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common presenting features, seen in 30 (55.6%), 29 (53.7%), and 23 (42.6%) patients, respectively, whereas hepatomegaly was reported in 14 (25.9%) patients.

The mean concentration of hemoglobin (Hb) was 11.57 ± 2.66 g/dL (range 6.4–17.2), the median (IQR, range) of total white blood cell (WBC) count, ALC, and platelet count were 51.2 × 10^9/L (78.6, 16–418.9), 39.2 × 10^9/L (54.48, 6.9–276), 185.5 × 10^9/L (90.75, 4–491), respectively.

Serum sCD23 and BCMA were significantly higher in patients' group in comparison with the control group with $P < 0.001$ for each [Table 1].

There was a significant difference in the median level of sCD23 among different Binet stages with $P < 0.001$ [Table 2]; the level was highest in Stage C followed by Stage B and then Stage A; the difference between Stage A and Stage B was significant ($P = 0.043$) and between Stage B and Stage C as well ($P = 0.016$). A significant difference in the median level of BCMA was found among different Binet stages ($P < 0.001$); the level was highest in Stage C followed by Stage B and then stage A; the differences between Stage A and Stage B and Stage B and Stage C were both significant ($P = 0.046$ and 0.038, respectively).

Spearman's rho correlation between the studied markers and the hematological parameters showed a significant positive correlation between sCD23 with total WBC count, lymphocyte count, and significant negative correlation with Hb, and platelet count. In addition, there were significant positive correlations between BCMA and total WBC count, lymphocyte count, and a negative significant correlation with Hb [Table 3].

Spearman's rho correlation between the studied markers and the clinical features showed significant positive correlations between sCD23 and B symptoms, and splenomegaly whereas BCMA showed significant positive correlation with B symptoms, LAP, splenomegaly, and hepatomegaly [Table 4].

Table 1: Comparison of the serum soluble CD23 and B-cell maturation antigen levels between patients and control groups

Characteristic	Patients group (*n*=54)	Control group (*n*=27)	*P**
sCD23 (pg/mL)			
Median (IQR)	1435.40 (909.83)	337.99 (157.72)	<0.001
Range	220.88-2067.21	95.46-554.27	
BCMA (ng/L)			
Median (IQR)	203.24 (381.20)	121.60 (27.52)	<0.001
Range	80.30-830.25	66.59-162.56	

*Mann-Whitney *U*-test. *n*=Number of cases, IQR=Interquartile range, BCMA=B-cell maturation antigen, sCD23=Soluble CD23

Discussion

The mean age of CLL patients in this study was similar to that of Aljabban and Alalsaidissa[9] and Naji[10] in Iraq and almost similar to that of neighboring countries such as Kuwait[11] and Iran,[12] while the median age tends to be higher in western countries.[13,14] This difference may be related to the geographical and environmental factors mainly due to wars that occurred in this area which lead to decrease in the median age of CLL patients in comparison with western countries. Male patients were more than females which is comparable to that of Al-Rubaie HA *et al*.[15] in Iraq and was almost similar to that of a Canadian study,[16] but slightly lower than that of Aljabban and Alalsaidissa[9] in Iraq and studies from western countries (1.7:1).[17,18] This disparity may be due to the difference in the number of CLL patients in each study but in general, there was a male predominance in all studies.

Studies from western countries revealed that most CLL cases are diagnosed on routine blood investigations in asymptomatic participants.[19] In contrast, only (24.1%) of patients enrolled in this study were asymptomatic at presentation and diagnosed incidentally, which might be attributed to a variation in the biology of the disease.[18] Among the symptomatic patients, B-symptoms and LAP were the most common presenting features followed by splenomegaly and the least hepatomegaly. This result was in agreement with a study done by Naji *et al*.[20] Another comparable study in the Kurdistan region of Iraq had shown that the most common presenting feature was LAP (57.6%) followed by splenomegaly (51.1%) and the least was hepatomegaly (15.3%).[21]

Table 2: Comparison of the levels of soluble CD23 and B-cell maturation antigen according to binet stage of disease

Parameters	Total (*n*=54)	Stage A (*n*=33)	Stage B (*n*=7)	Stage C (*n*=14)	*P**
sCD23 (pg/ml)					
Median (IQR)	1435.40 (909.83)	1186.40 (739.52)^C	1479.40 (809.79)^B	1990.80 (116.78)^A	<0.001
Range	220.88-2067.21	220.88-1931.91	901.00-1995.27	1457.48-2067.21	
BCMA (ng/L)					
Median (IQR)	203.24 (381.20)	157.43 (95.32)^C	419.34 (428.25)^B	541.64 (144.61)^A	<0.001
Range	80.30-830.25	80.30-540.85	85.62-615.15	361.74-830.25	

*Kruskal-Wallis test. A, B, and C capital letters were used to indicate the level of significance A>B> C following *post hoc* Dunn's multiple comparison test. *n*=Number of cases, IQR=Interquartile range, BCMA=B-cell maturation antigen, sCD23=Soluble CD23

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Table 3: Spearman's rho correlation of the studied markers to other hematological parameters

Characteristic	Hb		WBC count		Lymphocyte		Platelet	
	r	*P*	*r*	*P*	*r*	*P*	*r*	*P*
sCD23 (pg/mL)	−0.557	<0.001	0.454	0.001	0.462	<0.001	−0.323	0.017
BCMA (ng/L)	−0.509	<0.001	0.458	<0.001	0.466	<0.001	−0.195	0.158

BCMA=B-cell maturation antigen, sCD23=Soluble CD23, WBC=White blood cell, Hb=Hemoglobin, *r*=Correlation coefficient

Table 4: Spearman's rho correlation of the studied markers with clinical features

Marker	B-symptoms		LAP		Splenomegaly		Hepatomegaly	
	r	*P*	*r*	*P*	*r*	*P*	*r*	*P*
sCD23 (pg/mL)	0.449	0.001	0.250	0.068	0.319	0.019	0.237	0.084
BCMA (ng/L)	0.513	<0.001	0.429	0.001	0.306	0.024	0.281	0.040

r=Correlation coefficient, BCMA=B-cell maturation antigen, sCD23=Soluble CD23, LAP: Lymphadenopathy

The median levels of total WBC count and ALC were lower than those in other Iraqi studies done by Al-Rubaie *et al*.[15,22] and a study done by Cavalcanti Júnior *et al*.,[23] these differences may be due to the difference in sample size or improvement in the early detection of the disease. The median level of platelet count is comparable with other Iraqi studies[15,24] and a Brazilian study.[23]

The serological tests that were done to estimate serum markers which play an important role in both diagnosis and evaluation of prognosis in CLL are standard and inexpensive.[25] Serum sCD23 was significantly higher in the patients' group than the control group which is comparable to an Italian study,[26] a Turkish study,[4] and a European study.[27] The correlation between the serum levels of sCD23 and Binet clinical stages of the disease showed statistically significant differences among various stages which are comparable to the Italian study[26] that showed different serum levels of sCD23 in different Binet clinical stages and also divided stage B into two different prognostic subgroups according to these different serum levels. The Turkish study[4] also reported higher levels of serum sCD23 in Stage C.

There was a significant correlation between serum sCD23 and total WBC count, ALC, Hb, and platelets count. The correlation between sCD23 and ALC was observed in two previous studies done by Callea *et al*.,[3] Molica *et al*.[26] Furthermore, serum sCD23 showed a significant correlation with B symptoms and splenomegaly. The significant correlation of sCD23 with clinical and hematological parameters gave this serum biomarker a prognostic importance in CLL patients.

BCMA is expressed at high levels on the surface of malignant cells from multiple myeloma patients and is elevated in the serum of these patients and correlates with disease burden and response to treatment.[6] Plasma and serum show identical levels of BCMA.[7] In this study, the median serum level of BCMA in CLL patients was high in comparison with the control group, comparable to that reported by Udd *et al*.[8]

There was a significant association between the serum levels of BCMA and various stages of Binet system; the level was highest in Stage C, providing an important poor prognostic indicator in patients with advanced CLL.

Serum BCMA showed significant correlations with total WBC count and ALC. A positive correlation of BCMA with total WBC count was reported in Udd *et al*.[8] study. In addition, serum BCMA showed statistically significant positive correlation with B symptoms, LAP, splenomegaly, and hepatomegaly. These results indicate that this serum biomarker could be used as a reliable tumor burden biomarker in CLL patients.

Conclusions

The levels of serum sCD23 and serum BCMA were high in CLL patients and increased with advancing Binet stages of the disease indicating their usefulness as good and reliable parameters for prognostic evaluation in CLL patients. Both markers showed significant correlation with hematological parameters; mainly total WBC and ALC, and also with the clinical features which make them reliable tumor burden markers in CLL patients.

Financial support and sponsorship
Nil.

Conflicts of interest
There are no conflicts of interest.

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