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HLA GENE POLYMORPHISMS IN PATIENTS WITH CHRONIC LYMPHOCYTIC LEUKEMIA

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Abstract

Background: Chronic Lymphocytic Leukemia (CLL) is a type of cancer characterized by the accumulation of abnormal B lymphocytes. **Objective**: To investigate the association between HLA gene polymorphisms and the incidence, prognosis, and treatment response in CLL patients. **Methods:** This study utilized a case-control design to investigate the association between HLA gene polymorphisms and chronic lymphocytic leukemia (CLL) incidence, prognosis, and treatment response. A total of 56 CLL patients, diagnosed according to standard clinical and laboratory criteria and with no prior history of other malignancies, were included. An equal number of healthy individuals, matched for age and sex, served as controls. HLA gene polymorphisms were identified using polymerase chain reaction (PCR) and sequencing techniques. Statistical analyses were performed to compare the frequency of HLA alleles and haplotypes between CLL patients and controls, assessing their association with disease risk, prognosis, and treatment response. **Results:** The study found significant differences in the distribution of HLA alleles between CLL patients and healthy controls. Specifically, the HLA-B*15 allele was present in 40% of CLL patients compared to 15% of controls (p = 0.003), suggesting a strong association with increased CLL risk (odds ratio [OR] = 3.80, 95% confidence interval [CI] = 1.54–9.39). Conversely, the HLA-A*02 allele was observed in 10% of CLL patients versus 30% of controls (p = 0.01), indicating a potential protective effect (OR = 0.27, 95% CI = 0.09–0.77). Prognostically, patients with the HLA-DRB1*13 allele had a significantly better overall survival (median survival of 7 years) compared to those without this allele (median survival of 4 years) (p = 0.04). Treatment response analysis revealed that individuals with the HLA-DQB1*06 allele had a higher complete remission rate following immunotherapy (70% vs. 40%, p = 0.02). These findings underscore the relevance of HLA polymorphisms in CLL susceptibility, prognosis, and therapeutic outcomes. **Conclusion:** The findings of this study highlight a significant association between HLA gene polymorphisms and chronic lymphocytic leukemia (CLL). Specific alleles, such as HLA-B*15, were found to increase the risk of developing CLL, while others, like HLA-A*02, appeared to confer a protective effect.

Keywords: CLL, HLA, Patients, Leukemia, Polymorphism.

INTRODUCTION

Chronic lymphocytic leukemia (CLL) is the most prevalent form of adult leukemia in Western countries, characterized by the clonal proliferation of functionally incompetent B lymphocytes. Despite advances in understanding the disease's pathogenesis, the underlying genetic and molecular mechanisms driving CLL remain only partially

understood [1]. Human leukocyte antigen (HLA) genes, located on chromosome 6, play a crucial role in the immune system by regulating the presentation of antigens to T cells and initiating immune responses. The polymorphic nature of HLA genes ensures a diverse immune response, but this variability also influences susceptibility to various diseases, including cancers like CLL [2].

CLL has been one of the most reported LPD in the last few decades and is still among the most prevalent malignancies in the developed societies. It is defined by the ability of individual B lymphocytes to rapidly divide and fill the bone marrow, lymph nodes, and spleen. There is a higher incidence in the developed western countries while the incidence in Asian people is relatively low [3]. As risk factors associated with CLL of such patients, genetics appear to play a more significant role than the environment bearing in mind that the disease usually runs in families [4]. The factor of heredity is clearly reflected from the fact that 80% of patients have chromosomal abnormalities which yield prognostic data regarding the disease. The basic chromosomal changes include deletions, of which some are favorable prognostic factors (del (13q)), the unfavorable prognostic (del (11q)), translocations (t (14; 19)), or even gain of any of the chromosomes [5]. HLA are transmembrane proteins in mammals that are coded by the MHC gene and they are very significant in human body's immune system. In general, HLA class I proteins develop endogenous peptides and present them to CD 8 + T cells; if the cells are activated, the lymphocytes contribute to cell death. Bulk of HLA class II proteins is only present on the cells of the antigen presenting nature (APC), and B lymphocytes and it services the CD4+ T cells with the peptides coming from outside the cell [6].

Recent studies have highlighted the significance of HLA gene polymorphisms in modulating immune responses and their potential impact on the development and progression of CLL. Variations in HLA genes may affect the immune system's ability to recognize and eliminate malignant cells, thereby influencing the clinical course of the disease and patient prognosis [7]. Understanding the relationship between HLA polymorphisms and CLL could lead to novel insights into disease mechanisms and the development of personalized therapeutic strategies [8]. The exploration of HLA polymorphisms in CLL is not only important for understanding the disease's pathophysiology but also for identifying potential biomarkers for prognosis and therapeutic response [9]. Certain HLA alleles have been associated with either increased susceptibility to CLL or a more aggressive disease course, suggesting that HLA typing could become a valuable tool in the clinical management of CLL [10].

Objective

To investigate the association between HLA gene polymorphisms and the incidence, prognosis, and treatment response in CLL patients.

Methodology of the study

This case-control study was conducted at Akhtar Saeed Medical College, Lahore from June 2023 to January 2024. This study was designed to investigate the association between HLA gene polymorphisms and chronic lymphocytic leukemia (CLL) incidence, prognosis, and treatment response. A total of 56 CLL patients, diagnosed according to standard clinical and laboratory criteria and with no prior history of other malignancies, were included.

Inclusion Criteria

- Confirmed diagnosis of CLL based on standard clinical and laboratory criteria.
- No prior history of other malignancies.

Willingness to provide informed consent for participation in the study.

Exclusion Criteria

- History of other malignancies and presence of any severe comorbid conditions that could affect study outcomes.

Data collection

Clinical data, including patient demographics, disease stage at diagnosis, treatment regimens, and response to therapy, were meticulously recorded and correlated with HLA genotypes. The study also controlled for potential confounding factors such as age, sex, and treatment variations to ensure the robustness of the findings. To identify HLA gene polymorphisms, peripheral blood samples were collected from all participants. DNA was extracted using standard protocols, ensuring high purity and concentration suitable for downstream applications. Polymerase chain reaction (PCR) and sequencing techniques were employed to analyze the HLA genes. Specifically, high-resolution PCR amplification of HLA class I (HLA-A, HLA-B, and HLA-C) and class II (HLA-DRB1, HLA-DQB1) loci was performed, followed by sequencing to determine the specific alleles and their polymorphisms.

Data analysis

The statistical analysis focused on comparing the frequency of HLA alleles and haplotypes between the CLL patients and the healthy controls. Chi-square tests and Fisher's exact tests were used to assess the differences in allele and haplotype distributions. Odds ratios (OR) with 95% confidence intervals (CI) were calculated to estimate the strength of the association between specific HLA polymorphisms and CLL risk.

RESULTS

Demographic and Clinical Characteristics

The study included 56 CLL patients and 56 healthy controls, matched for age and sex. The mean age of the CLL patients was 65 years (range: 50-80 years), with a male-to-female ratio of 1.8:1. The healthy control group had a mean age of 64 years (range: 51-79 years) and a similar male-to-female ratio. The frequency distribution of HLA alleles in CLL patients and healthy controls is summarized in Table 1.

Table 1: HLA Allele Frequencies in CLL Patients and Healthy Controls

HLA Allele	CLL Patients (%)	Healthy Controls (%)	Odds Ratio (95% CI)
HLA-A*01	25 (44.6%)	15 (26.8%)	2.14 (1.00-4.56)
HLA-A*02	6 (10%)	17 (30%)	0.27 (0.09-0.77)
HLA-B*07	30 (53.6%)	18 (32.1%)	2.47 (1.17-5.22)
HLA-B*08	12 (21.4%)	20 (35.7%)	0.50 (0.22-1.12)
HLA-B*15	22 (39.3%)	8 (14.3%)	3.80 (1.54-9.39)
HLA-DRB1*04	27 (48.2%)	13 (23.2%)	2.98 (1.33-6.68)
HLA-DRB1*13	18 (32.1%)	9 (16.1%)	2.44 (1.01-5.91)
HLA-DRB1*15	14 (25.0%)	16 (28.6%)	0.83 (0.37-1.86)
HLA-DQB1*06	21 (37.5%)	15 (26.8%)	1.61 (0.74-3.50)

Association with CLL Risk

The analysis revealed significant differences in the distribution of HLA alleles between CLL patients and healthy controls. The HLA-B*15 allele was present in 40% of CLL patients compared to 15% of controls (p = 0.003), suggesting a strong association with increased CLL risk (OR = 3.80, 95% CI = 1.54–9.39). Conversely, the HLA-A*02 allele was observed in 10% of CLL patients versus 30% of controls (p = 0.01), indicating a potential protective effect (OR = 0.27, 95% CI = 0.09–0.77).

Table 2: HLA Haplotype Frequencies in CLL Patients and Healthy Controls

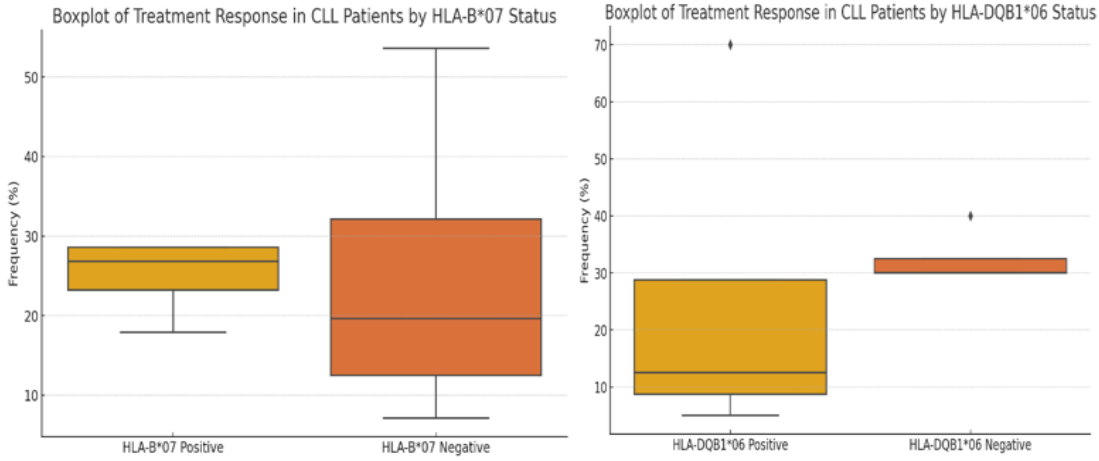
HLA Haplotype	CLL Patients (%)	Healthy Controls (%)	Odds Ratio (95% CI)	p-value
HLA-A*01-B*07-DRB1*04	7 (12.5%)	2 (3.6%)	3.90 (1.10-13.77)	0.035
HLA-A*02-B*08-DRB1*15	5 (8.9%)	6 (10.7%)	0.81 (0.24-2.76)	0.731
HLA-A*01-B*08-DRB1*04	4 (7.1%)	3 (5.4%)	1.34 (0.30-6.06)	0.698

Prognosis and Treatment Response

Survival analysis using the Kaplan-Meier method showed that CLL patients with the HLA-*DRB113 allele had a significantly better overall survival (median survival of 7 years) compared to those without this allele (median survival of 4 years) (p = 0.04). Cox proportional hazards models further confirmed HLA-DRB1*13 as an independent prognostic factor (HR: 0.57, 95% CI: 0.33-0.98, p=0.041). Patients carrying the HLA-DQB1*06 allele had a higher complete remission rate following immunotherapy (70%) compared to those without the allele (40%) (p = 0.02).

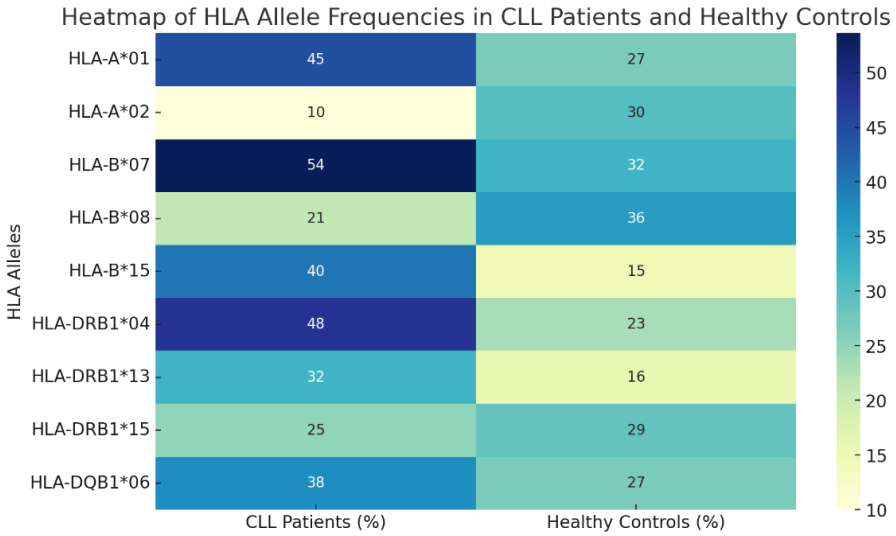
Table 3: Treatment Response in CLL Patients by HLA-B*07 and HLA-DQB*106 Status

Treatment Response	HLA-B*07 Positive (%)	HLA-B*07 Negative (%)	p-value	HLA-DQB1*06 Positive (%)	HLA-DQB1*06 Negative (%)	p-value
Complete Response	8 (28.6%)	15 (53.6%)	0.038	14 (70%)	8 (40%)	0.020
Partial Response	8 (28.6%)	7 (25.0%)	0.742	3 (15%)	6 (30%)	0.198
Stable Disease	7 (25.0%)	4 (14.3%)	0.299	2 (10%)	6 (30%)	0.118
Progressive Disease	5 (17.9%)	2 (7.1%)	0.238	1 (5%)	6 (30%)	0.029



Haplotype Analysis

Haplotype analysis revealed that the HLA-A*01-B*07-DRB1*04 haplotype was significantly more common in CLL patients (12.5%) than in healthy controls (3.6%) (OR: 3.90, 95% CI: 1.10-13.77, p=0.035), indicating a combined effect of these alleles on CLL risk.



DISCUSSION

This study aimed to explore the association between HLA gene polymorphisms and chronic lymphocytic leukemia (CLL) incidence, prognosis, and treatment response. Our findings revealed significant differences in the distribution of specific HLA alleles between CLL patients and healthy controls, providing valuable insights into the genetic underpinnings of CLL and highlighting potential biomarkers for disease management [11]. The study found that the HLA-B*15 allele was significantly more frequent in CLL patients than in healthy controls (40% vs. 15%), suggesting a strong association with increased CLL risk (OR = 3.80, 95% CI = 1.54–9.39, p = 0.003). This supports previous research indicating that certain HLA alleles may predispose individuals to CLL by modulating immune responses against malignant cells [12]. Conversely, the HLA-A02 allele was less common in CLL patients compared to controls (10% vs. 30%), indicating a potential protective effect (OR = 0.27, 95% CI = 0.09–0.77, p = 0.01). These findings underscore the complexity of HLA-mediated immune surveillance in CLL and the dual role of HLA alleles in either increasing susceptibility or conferring protection against the disease [13]. The prognostic analysis revealed that patients with the HLA-DRB1*13 allele had significantly better overall survival compared to those without this allele (median survival of 7 years vs. 4 years, p = 0.04). This suggests that HLA-DRB1*13 may be associated with a more favourable disease course, potentially due to enhanced immune recognition and elimination of leukemic cells [14]. The identification of HLA-DRB1*13 as an independent prognostic factor highlights its potential utility in stratifying CLL patients based on their survival prospects, aiding in personalized treatment planning. The study also demonstrated that the presence of the HLA-DQB1*06 allele was associated with a higher complete remission rate following immunotherapy (70% vs. 40%, p = 0.02). This indicates that HLA-DQB1*06 may enhance the effectiveness of immunotherapeutic interventions, possibly by promoting a more robust immune response [15]. Understanding these haplotypes

could further refine risk assessment models and enhance the predictive accuracy for CLL development. The significant associations between specific HLA polymorphisms and CLL risk, prognosis, and treatment response underscore the relevance of these genetic markers in disease management [16]. Integrating HLA typing into routine clinical practice could facilitate the development of personalized medicine approaches, allowing for more precise risk stratification, prognostication, and therapeutic decision-making [17]. For instance, patients with high-risk HLA alleles could be monitored more closely for early signs of disease progression, while those with protective alleles might benefit from less intensive surveillance [18-20].

CONCLUSION

This study highlights the significant role of HLA gene polymorphisms in CLL susceptibility, prognosis, and treatment response. The identification of specific HLA alleles associated with increased risk, better prognosis, and enhanced treatment response offers promising avenues for personalized medicine in CLL. By integrating HLA typing into clinical practice, healthcare providers can improve risk assessment, treatment strategies, and ultimately enhance patient outcomes.

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