

**Research Article****Role of IL10RA gene in Genetic polymorphisms with the association  
of blood transfusion in leukemia patients****Farah Asghar<sup>1</sup>, Muhammad Shaharyar Afzal<sup>2</sup>  
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**ABSTRACT**

**Aims and objectives:** Before 19<sup>th</sup> century it was thought that all the blood was same and this misunderstanding was lead to the fetal transfusion of blood. The main objective of this study is to find the correlation of genetic polymorphisms in *IL10RA* which modify the association of blood transfusion and induce apoptosis of human leukemia. **Material and methods:** This study was conducted according to the rules and regulations of hospital authority and approved by ethical committee of hospital. There was no violence of rules and regulations of authority. All histologically confirmed cases of leukemia diagnosed from 2015 to 2016 in a hospital. **Results:** Our results suggested that there is no positive correlation between *IL10RA* and leukemia disease. **Conclusion:** Our results suggest that genetic polymorphism in *IL10RA* gene modifies the association between blood transfusion and risk of leukemia B-cells.

**Key words:** Bone marrow, Genotyping, Acute lymphoblastic leukemia, Transplantation**INTRODUCTION**

Before 19<sup>th</sup> century it was thought that all the blood was same and this misunderstanding was lead to the fetal transfusion of blood. Even human blood is not the same. People belong to the different blood groups depending upon the antigens present in the blood. Almost 25 different types of blood groups were present but most common are ABO and Rh systems<sup>1</sup>. Blood transfusion mainly allogeneic blood transfusion can induce immune suppression and has been suggested as a risk factor for leukemia<sup>2</sup>.

Molecular genotyping methods were introduced to the transfusion medicine community over a decade ago. Epidemiological studies linking blood transfusion to the risk of leukemia but they provided inconsistent results<sup>3</sup>. B-lymphocytes are characterized by the expression of CD19 surface antigen, which is present on the progenitor cells of bone and persists during all stages of B-cell maturation<sup>4</sup>.

T-helper cells play an important role in the regulation of key pathways of immune system. Imbalanced regulation and expression of Th1 and Th2 lymphocyte cytokines have been linked to the

development of different blood diseases<sup>5</sup>. Single nucleotide polymorphisms in *TNF* and *IL10* have been reported to be associated with the risk of leukemia<sup>6</sup>. Damaged autologous erythrocytes during blood transfusion has been shown to augment the cytokines *TNF-α* and *IL-10* production of the mononuclear phagocyte system in humans<sup>7</sup>.

**Aims of the study**

The main objective of this study is to find the correlation of genetic polymorphisms in *IL10RA* which modify the association of blood transfusion and induce apoptosis of human leukemia

**MATERIAL AND METHODS**

**Ethical approval**

This study was conducted according to the rules and regulations of hospital authority and approved by ethical committee of hospital. There was no violence of rules and regulations of authority. All histologically confirmed cases of leukemia diagnosed from 2015 to 2016 in a hospital. Enrollment criteria include the age between 20 to 50 years. Pathology slides from all patients were obtained from the original pathology departments and reviewed by two independent pathologists.

**Table 01:**Primer Sequences for PCR

Gene	Forward Primer	Reverse Primer	Ref
Abcg5	5'-TTGCGATACACAGCGATGCT-3'	5-TGACTGCCTCTACCTTCTTGTGT-3'	(Song, et al., 2010)
Abcg8	5'-CCGTCGTCAGATTTCCAATGA-3'	5'-GGCTTCCGACCCATGAATG-3'	-do-
Ldlr	5'-GCTCCATAGGCTATCTGCTCTTCA-3'	5'-CTGCGGTCCAGGGTCATC-3'	-do-
CYP7A1	5'-CCATGATGCAAAACCTCCAAT-3'	5'-ACCCAGACAGCGCTCTTTGA-3'	-do-
MIR33a (Probe sequence)		5'- GUGCAUUGUAGUUGCAUUG-3'	(Li, et al., 2013)

**Table 02:** Associations between *IL10RA* Polymorphisms, Blood Transfusion, and risk of leukemia

SNPs	Overall						B-cell lymphoma			
	Blood transfusion						Blood transfusion			
	No			Yes			No		Yes	
	Control	Case	OR	Control	Case	OR	Cases	OR	Cases	OR
<i>IL10RA</i>										
GG	121	103	1.0	33	47	1.9	76	1.0	33	1.0
AG/AA	376	251	1.0	86	42	0.6	206	1.0	35	1.0
P-interaction	<b>0.003</b>						<b>0.001</b>			

We also collected the patient’s history and compared this data to those patients who have not the history of blood transfusion. The patients who have the history of blood transfusion are suffering from high risk of

**Genotyping**

A genotyping experiment is an end point experiment which is used to determine the genotype of samples in ShoukatKhanum Laboratory center. In this experiment we can easily differentiate between two alleles of SNP. First of all I was collected the sample for this experiment with the help of medical staff of hospital. The concordance rate of all the samples was also measured. The specific type of fluorescent dye was used for PCR. Taq Man® genotype plate was used for the best results. For genotype analysis forward and reverse primers were used.

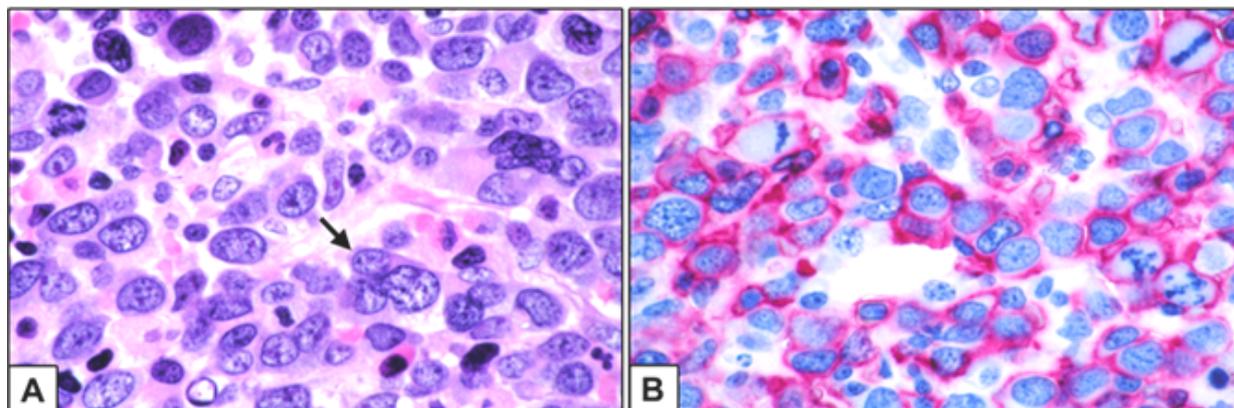
**Statistical analysis**

Unconditional logistic regression was used to find out the odds ratios (ORs) and 95% confidence intervals for relations between blood transfusion, and risk of leukemia. All *P* values presented in the results are two-sided, and all analyses were performed by using SAS software (version 9.2).

**RESULTS**

The association between blood transfusion and leukemia B-cells are clearly presented in the table 01 and 02. Table 01 represents the primer sequence of genes and table 02 shows the Associations between *IL10RA* Polymorphisms, Blood Transfusion, and risk of leukemia.

leukemia if they carried IL10RA GG genotype. And this risk is minimized when they contain IL10RA AG/AA genotype.



**Figure 01:** Histopathological analysis shows the AML bone marrow biopsy. This is the bone marrow biopsy which sometime shows the multi lobated nuclei (arrow). Leukemia cells are myelomocytic in nature and appearance (A; hematoxylin eosin). CD34 negative cells display aberrant cytoplasm expression of nucleophosmin (B- CD34 endothelial cells). (Size: 300×300dpi).

## DISCUSSION

This is the first comprehensive analysis of relation of blood transfusion and risk of leukemia in humans. There is a significant difference were observed in for *IL10RA* and *TNF* for leukemia and the high production of white blood cells. No interactions were observed for blood transfusion and the high production of white blood cells. For the clarification of this statement higher studies will required for further clarification<sup>8</sup>.

The IL-10RA receptor chains have an extracellular domain consisting of 200 amino acids, a transmembrane helix consisting of 20 amino acids, and an intracellular domain consisting of 322 amino acids for IL-10RA<sup>9</sup>. IL10 and TNF were considered to be the key genes for lymphomagenesis. Both the genes code the immunoregulatory cytokines that are considered to be critical mediators of inflammation and apoptosis and also for lymphoid tumors<sup>10</sup>. Different studies related to TNF and IL10 shows that each cell effects on B-cell lymphomagenesis by direct or indirect way<sup>11</sup>.

The long term storage of red blood cells before transfusion has been reported and it can increase the intracellular iron. And they cause the systemic inflammatory response syndrome and it will lead to deleterious consequences<sup>12</sup>. Therefore, it may

be possible that the genetic variation in TNF and IL10RA genes modify the association between blood transfusion and risk of CLL.

IL10RA gene encodes the interleukin 10-receptor alpha chain of the IL10 receptor complex and plays a critical role in the regulation of signal transduction<sup>13</sup>. The signaling of IL10RA causes transcriptional activation of many genes which play an important role in immunological diseases<sup>14</sup>. The level of IL10RA correlates with the strength of IL10 on immune cells<sup>15</sup>.

## Conclusion

Our results suggest that genetic polymorphism in IL10RA gene modifies the association between blood transfusion and risk of leukemia B-cells.

## Conflict of interest

The authors declare that there is no conflict of interest of financial and fiduciary activities from any author.

## Contribution of authors

All the authors contributed equally in this research and for writing this manuscript.

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