ABSTRACT
The etiology of acute lymphocytic leukemia (ALL) remain uncertain, although some doctors believe that ALL develop from a combination of genetic and environmental factors. It is the most frequent malignancy affecting children, constituting about 30% of all childhood cancers. The causes of most pediatric acute leukemia are unknown and likely to involve an interaction between the environment and hematopoietic development. Pathogenesis of leukemia is not clearly understood, but immature, nonfunctioning WBCs appear to accumulate first in the tissue where they originate (lymphocytes in lymph tissue, granulocytes in bone marrow). These immature WBCs then spill into the bloodstream and from there infiltrate other tissues, eventually causing organ malfunction because of encroachment or hemorrhage. The occurrence of pediatric leukemia has been linked to several environmental, maternal, and paternal characteristics and to exposure to various biological, physical, and chemical factors. The risk of developing cancer is strongly influenced by genetically determined difference. Recent molecular epidemiologic research provides compelling evidence that environmental factors are major contributors to human carcinogenesis. Therefore these agents may influence individual susceptibility to cancer. Children are vulnerable to environmental toxicants because of their greater relative exposure and immature metabolism.

KEYWORDS: Acute Lymphoblastic Leukemia, environmental factors, hematopoisis.

INTRODUCTION
Hematopoiesis can be divided into two main lineages: myeloid and lymphoid. The myeloid lineage produces monocytes, macrophages, neutrophils, eosinophils, basophils, red blood
cells and platelets. The lymphoid lineage gives rise to B lymphoblasts alias B cells, T lymphoblasts alias T cells and natural killer (NK) cells \[^1\]. The division, survival, life span, lineage commitment and differentiation of the haematopoietic cells are controlled by various growth factors as well as the interaction between the cells and their microenvironment. Incidence rates for all types of leukemia are higher among males than among females. In 2012, males have been accounted for more than 57 percent of the new cases of leukemia \[^2\]. An estimated 48,610 new cases of leukemia have been diagnosed in the United States in 2013 \[^2\]. The effects of these factors are transmitted through cell surface receptor molecules into the cell where they cause changes in the function of transcription factors and, thus, in the activity of genes \[^3, 4\]. In India, acute lymphoblastic leukemia (ALL) is the most common type of childhood malignancy which accounts for approximately 30% of cancer cases in children younger than 15 years old \[^5\].

**MATERIAL AND METHODS**

**Patients:** This study was performed on 112 patients diagnosed with acute lymphoid leukemia from the Department of Pathology. These patients were evaluated especially regarding sex, age at diagnosis, risk factors, occupation, and chief complaints.

**Hematological analysis:** The whole blood samples of patients were taken with informed consent for hematological analysis. This study was approved by Institutional ethical committee of MD University, Rohtak. Percentage of blast cell, red blood cell indices, white blood cell indices, number of platelets and the amount of hemoglobin was determined according to standard laboratory procedures. Slides were prepared with PBF (Peripheral Blood Film) by using ‘Leishman stain’ to find out the blast cells morphology in peripheral blood sample of all patients of blood cancer. Diagnosis of leukemia was done by 20% blast criteria and then ‘Sudan Black B’ was used to distinguish between AML & ALL \[^5, 6\].

**Statistical analysis:** Med calc software was used to calculate risk factors affecting leukemia patients. Data on leukemia patients was analyzed by using Odd’s ratio in order to find out interaction of environmental agents in development of prenatal and postnatal acute lymphoid leukemia.

**RESULTS**

A total of 112 patients were studied including 74 children and 38 adults. In children there were 49 male and 25 female patients. In adult cases 25 male and 13 females. There were 60
male patients with B-cell ALL and 14 male patients with T-cell ALL. Female patients had B-cell ALL in 30 cases and T-cell ALL in 8 cases. Similarly respective ratio of B-cell ALL and T-cell ALL was present in children and adults. In children, there were 40 male patients with B-cell ALL and 11 male patients with T-cell ALL. Female patients had B-cell ALL in 19 cases and T-cell ALL in 6 cases. In adults, there were 20 male patients with B-cell ALL and 3 male patients with T-cell ALL. Female patients had B-cell ALL in 11 cases and T-cell ALL in 2 cases (Figure-1). Both B-cell ALL and T-cell ALL had more male patients than female patients.

Figure-1. Cell type and sex ratio in patients of acute lymphocytic leukemia.

Risk factors
Clinical patients of acute lymphocytic leukemia (ALL) were carefully studied to know the role of various risk factors in occurrence of acute lymphocytic leukemia (ALL). The main risk factors considered in present study were prenatal and postnatal risk factors along with occupation of patients and their parents. The various risk factors were analyzed in both group i.e. children (<15 years) and adults (>15 years).

Prenatal risk factors in ALL
Prenatal history of patients was studied in detail and certain prenatal risk factors considered in the present study were drug intake, exposure to pesticides, exposure to ionizing radiation, and fetal loss. Frequency of ALL patients with prenatal risk factors was 74%. Drug intake was noted in 42% patients. Fetal loss was found in 12.5% patients followed by exposure to
Pesticides in 10.7%. Exposure to ionizing radiation was found in only 4.5% mothers during pregnancy.

**Prenatal risk factors in children of ALL**
Mothers of 51.7% children had history of drug intake. Pesticides exposure during agriculture activities was reported in parents of 14.3% children. Repeated exposure to X-rays and ultrasound was noted in mothers of 4.5% children. Mothers of 16.1% children had history of fetal loss more than two times. No risk factor was noted in parents of 13.4% children.

**Prenatal risk factors in adults of ALL**
Mothers of 28.5% adults had history of drug intake. Pesticides exposure during agriculture activities was reported in parents of 12.5% adults. Repeated exposure of X-rays and ultrasound was noted in mothers of 5.3% adult patients. Mothers of 10.7% adult patients had history of fetal loss more than two times. No risk factor was noted in parents of 42.8% adult patients.

**Postnatal risk factors in ALL**
Various Postnatal risk factors considered in the present study were smoking, alcohol consumption, pesticides, ionizing radiation, electromagnetic field and infection. There were 62.5% of acute lymphocytic leukemia cases who had shown postnatal complications. Frequency of various risk factors in acute lymphocytic leukemia were calculated as smoking in 14.3%, alcohol consumption in 9.8%, pesticides in 10.7%, exposure to ionizing radiation in 13.4%, exposure to electromagnetic field in 5.3% and infection in 8.9%.

**Postnatal risk factors in children ALL**
The most prominent postnatal risk factors in children were exposure to ionizing radiation, pesticides exposure, and infection. Seventeen point eight percent children had history of repeated X-rays exposure. Pesticides exposure during household activities was reported in 14.3% children. The history of infection (pneumonia) was reported in 7.1% children. Eight point nine percent children were found playing near power houses and high voltage electrical lines. Smoking was noted in 6.2% children. The risk factors were not known in 13.4% cases.

**Postnatal risk factors in adults of ALL**
Smoking and alcohol were most prominent factors in adult patients. Thirty three percent adult patients had habits of smoking. The alcohol addiction was found in 29.4% adult patients.
Pesticides exposure during agriculture activities was reported in 10.7% adults. Eight point nine percent adult patients had exposure to X-rays many times. Three point seven patients had residence near high voltage electrical lines and 3.7% patients had history of infection.

**Occupation of patients of ALL**

Occupation and place of work of patients and their parents was carefully analyzed to know their role in the occurrence of ALL. In the present investigation, main categories of occupation were agriculture, industry, workers in unorganized sector, and self business. Industry (44.6%) was the major occupation followed by agriculture (22.3%), worker in unorganized sector (19.6%) and self business (13.4%).

**Occupation of children of ALL**

Thirty seven point five percent children and their parents were engaged in different agriculture activities. Thirty seven point five percent children and their parents were living near working in industries like manufacturing paint, tire and fertilizer. The self business likes candle making, carpeting, and printing was reported in 16.1% patients and their parents. Nineteen point six percent children and their parents were working in unorganized sector like transport, restaurant and press.

**Occupation of adults of ALL**

Twenty five percent patients were engaged in agriculture. Thirty nine point two percent patients were living near /working in industries like manufacturing paint, pesticides and dye. The self business likes carpeting, and printing was reported in 22.3% patients. Thirteen point four percent patients were working in unorganized sector like transport, restaurant and brick making.

**Various Risk Factors and Odd’s Ratio**

The risk factors as prenatal, postnatal and occupation of patients and their parents of acute lymphocytic leukemia were analyzed. The data was statistically analyzed using Odd’s ratio in Medcalc software (version12.7.7). An Odd’s Ratio greater than 1 indicates that the condition or event is more likely to occur in the first group and an Odd’s Ratio less than 1 indicates that the condition or event is less likely to occur in the first group. Analysis using Odd’s ratio showed significant variation when noted in certain risk factors in prenatal, postnatal factors and occupation of patients and their parents.
Analysis by Odds ratio showed that the drug intake (OR=3.4574; 95% CI=1.9832 to 6.0275) was major risk factor and industrial area was the major occupation (OR=2.9390; 95% CI=0.4652 to 3.2308) of patients. In prenatal and postnatal risk factors pesticides exposure was found more frequently as compared to ionizing radiation with odds ratio of OR=1.2383; 95% CI=0.5899 to 2.5996 and 1.2732; 95% CI= 0.6914 to 2.3444 respectively. In postnatal patients who regularly had a habit of smoking showed increased odds ratio than alcohol consumption (OR=1.2532; 95% CI=0.6947 to 2.2605) (Table-1).

Table-1. Odds Ratio Analysis in Risk Factors of Acute Lymphocytic Leukemia.

<table>
<thead>
<tr>
<th>Risk factors</th>
<th>Present</th>
<th>Absent</th>
<th>Z-statistic</th>
<th>OR</th>
<th>95% CI</th>
<th>p-value</th>
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<td><strong>Prenatal Risk Factors:</strong></td>
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<td>Drug intake*</td>
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<td>47</td>
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<td>3.4574</td>
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<td>Pesticides exposure*</td>
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<td>1.2383</td>
<td>0.5899 to 2.5996</td>
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<td><strong>Postnatal Risk Factors:</strong></td>
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<td>Smoking*</td>
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<td>0.6947 to 2.2605</td>
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<td>Pesticides exposure*</td>
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<td>82</td>
<td>0.775</td>
<td>1.2732</td>
<td>0.6914 to 2.3444</td>
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<td>Industry*</td>
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<td>10</td>
<td>0.412</td>
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<td>0.4652 to 3.2308</td>
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<td>1.9846</td>
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</tbody>
</table>

(A) Note: *An odds ratio greater than 1 indicates that the condition or event is more likely to occur in the first group. And an odds ratio less than 1 indicates that the condition or event is less likely to occur in the first group.

(B) **Highly significant by Odd’s Ratio and z-test at p<0.01.

(C) Ns=not significant.

**DISCUSSION**

The present study revealed 49 male and 25 female cases in children of ALL. Children were reported more commonly than adults in literature [5, 6, 7, 8]. The majority of children in the present study were in the age group of 11-15 years with a male predominance, followed by a second peak between 2-5 years. Earlier Hassanzadeh and workers have reported higher
frequency of children in age group of 5-9 years followed by <4 years and 10-14 years [4] but in the present study maximum adult patients of ALL were in age group of 16-20 years followed by 21-30 years and so on. The mean age was 34.3 years. Several other studies have reported decreasing incidence of adults with increasing age [8, 9]. In the present study, there were 74% male and 38% female patients and male to female ratio was 2:1. Several studies have reported higher frequency of males in children [4, 5, 6, 9, 12].

The present study revealed more patients of B-cell ALL than T-cell ALL in children as well as in adults. There were 81.8% children with B-cell ALL and 18.2% children with T-cell ALL and 92% adult patients with B-cell ALL and 7.2% adults with T-cell ALL. Higher frequency of B-cell (70-80%) than T-cell (20-30%) has also been reported in adult patients [4, 5, 6, 9, 11, 12].

The risk factors considered in the present study were categorized as prenatal and postnatal risk factors. Prenatal risk factors considered in the present study were drug intake, exposure to pesticides, exposure to ionizing radiations and fetal loss. Mothers of 54% children and 32% adult patients of the present study had history of drug intake before or during pregnancy. Use of drugs before and during pregnancy has been reported with 10-fold increased risk of childhood AML and ALL [5]. Several studies have assessed the risk in utero exposure to diagnostic X-rays and an increased risk of ALL [7, 9]. However, in some studies a significant association was not found between childhood leukemia and X-ray examinations in the mother during pregnancy [10, 11]. Prenatal exposure to ionizing radiation was observed in 4% children and 3.7% adult patients of ALL in the present study. This apparent decrease in risk over time may be attributable to declining exposures to ionizing radiation (decreased dose) and to the increasing use of diagnostic ultrasound in place of diagnostic X-rays during pregnancy [5, 7, 12, 14].

Various postnatal risk factors considered in the present study were smoking, alcohol consumption, pesticides, ionizing radiation, electromagnetic field and infection. Smoking was observed in 37% adults. Although no data was available for smoking as postnatal risk factor, a study has shown 2-3 times higher risk of leukemia in smokers than non-smokers [12, 13, 14]. Tobacco smokers contain leukemogens like benzene. The alcohol addiction (33.3%) was found as another major risk factor in adult patients of the present study. Association of disease with alcohol addiction was not reported in literature. However, some studies have found increase risk of ALL in children whose parents had habits of alcohol [9, 12, 14]. Postnatal
exposure to ionizing radiation has been shown to increase the risk of childhood leukemia in several studies \cite{12,14}. Twenty percent children and 7.4\% adults had multi exposure to X-rays in the present study. Infants receiving diagnostic X-rays had 60\% more leukemia than other children \cite{12,14,15}.

Occupation of patients and their parents was also studied to know the role of environment in occurrence of disease. Several studies have shown increased risk of ALL in children depending upon the occupation of parents \cite{5,12,14}. In the present investigation, main categories of occupation were agriculture, industry, workers in unorganized sector and self business. Agriculture was a major occupation in children as well as adults. Most children’s exposure to pesticides is from home, lawn and garden use \cite{14}. Increased risk of ALL in children has been shown by occupation exposure to pesticides during agriculture activities \cite{10,11,12,14}. In the present study, pesticides exposure before or during pregnancy was observed in mothers of 14\% children and of 12.2\% adults. Postnatal exposure to pesticides during house hold activities and different agriculture activities was observed in 16\% children and in 7.4\% adult of ALL in the present study. Some studies have suggested that pesticide exposed fetuses and children are at higher risks for cancer compared with adults \cite{12,14,15}. However, there is much speculation about the biological mechanism by which paternal occupational exposures may affect the offspring’s risk for leukemia.

REFERENCES