Chapter 1

Introduction

The term "Hematological malignancies" (HM) defines a diverse group of neoplastic diseases originated in the hematopoietic and lymphoid tissues; with a particular cytogenetic profile and clinical presentation depending on each case and cell lineage (Taylor et al., 2017). HM constitute a group of cancers that arise from malignant transformation of peripheral blood, lymphatic system, and other bone marrow-derived cells. These diseases include the acute and chronic leukemia, Hodgkin's disease (now termed Hodgkin lymphoma), the non-Hodgkin lymphomas (NHL), and multiple myeloma. The heterogeneity seen in this collection of cancers reflects the complexity of the normal hematopoietic and immune systems (Flowers et al., 2007).

Individually, hematological malignancies occur less commonly than some solid tumors; however, collectively, leukemia, lymphoma, and myeloma accounted for an estimated 118,310 new cancer cases in 2006 (about 9% of cancer cases diagnosed in the U.S.) and 53,920 cancer deaths, placing this group 4th among cancers in each category (Jemal et al., 2006). In contrast to the WHO estimates, a multi-centered hospital-based data presents a different picture about the leukemia that constituted approximately two thirds of (64.3%) all HM cases, while NHL accounted for 16.9%, followed by MM (10.5%) and HL (3.9%). A similar pattern of leukemias (Age-standardized incidence rate or ASR per 100,000 is 3.3), NHL (ASR per 100,000 is 3), and multiple myeloma is observed in India. According to WHO, the commonest type of HM was NHL (ASR is 1.9 per 100,000 persons), which was followed by leukemias (ASR 1.7 per 100,000 persons), HL and multiple myeloma. In Pakistan, NHL is the most prevalent type of HM. In US, NHL is the commonest cancer among HM, which is 1.5 times that of all leukemias. All leukemia cases were over three times higher as compared to NHL cases in Bangladesh. In other Asian countries including Japan, Korea and Singapore, NHL is the most frequent hematological malignancies (Hossain et al., 2014).

Over the past several years, a number of classification systems have been developed to subdivide hematological malignancies in clinically and biologically relevant ways to refine our ability to diagnose and treat these cancers. Over time advances in diagnostic techniques, chemotherapy regimens and other treatment modalities have improved outcomes for patients with hematological malignancies. However, there remain disparities in the extent to which these advances have improved outcomes for all patients (Howlader et al., 2018).

In the 1960s, almost 25% of Global cancer was diagnosed in low-income and lower-middle income countries. In 2020 nearly 55% of Global cancer was found in these countries. By 2030, over 13 million people will die from cancer every year. Almost 9 million (about 70%) of these deaths will be in developing countries (Greer et al., 2013).

The African American population in the United States continues to carry a disproportionate risk of cancer occurrence and has the highest cancer mortality rate of any racial group. From 1996 to 2000, the cancer incidence rates for African Americans were 696.8 cases per 100,000 individuals per year for males and 406.3 cases per 100,000 individuals for females. An estimated 137,910 cases occurred in black individuals in 2005. Cancer death rates are also the highest among the African American community, occurring in 356.2 males per 100,000 per year and 198.6 females per 100,000 per year. Approximately 63,110 cancer deaths were estimated to occur in African Americans in 2005 (Cokkinides et al., 2005).

In 2005, approximately 1.4 million new cancer cases and 570,000 deaths due to cancer were estimated in the United States. African American men and women had 40% and 20% higher death rates from all cancers than white men and women, respectively. Although mortality rates for African American patients with lymphoma and leukemia are similar to white Americans, mortality rates for African Americans with myeloma are approximately twice those observed for white males and females in one study (Cokkinides et al., 2005). More significant are data indicating that, when compared with white patients, black patients have a decreased likelihood of 5-year survival after diagnosis for nearly all cancer sites and at all stages of diagnosis. Although survival rates have improved for some groups of patients with hematological malignancies, in most instances, 5-year survival for black patients remains inferior to the general

population. These differences in outcome appear to be principally related to limitations in access to medical care, later stage at diagnosis, economic factors, and disparities in treatment (Bach et al., 2002).

Bangladesh, at 164 million people, is the ninth most populous country in the world. There are 1.3 to 1.5 million cancer patients in Bangladesh, with about two hundred thousand patients newly diagnosed with cancer each year (Sharmin et al., 2022). Bangladesh shows major advances in relation to the management of infectious diseases as recently highlighted in Lancet, the chronic diseases in particular cancers are less prioritized (Das and Horton, 2013; Hussain and Sullivan, 2013). The status of cancer in this country is largely unknown, as there is no population-based cancer registry nor a national cancer registry of any other kind. According to WHO, Bangladesh is experiencing increasing cancer burden with estimated 122,715 new cancer cases in 2012. The number of new cases is projected to increase approximately by 77% in 2030. These WHO estimates may not reflect the real cancer status, as the estimates were extrapolated based on the incidence and mortality rates from regional data (India) and a single hospital (Ferlay et al., 2013).

Hematological malignancies (HM) are not uncommon in our country. All ages and genders are affected by HM. Hematological malignancies are a group of cancers that arise from a malignant transformation of cells of the bone marrow or lymphatic system (Flowers et al., 2007). There are several classification systems for hematological malignancies. In 2001 WHO classification was the first worldwide consensus classification on hematological tumors. The classification is based on information such as clinical, morphologic, biologic, immunophenotypic and genetic features (Delsol et al., 2001; Arber et al., 2016).

In 2008, as a part of series of classification of tumors ‘blue book’ monographs (4th edition), published a new classification for hematopoietic and lymphoid neoplasm in collaboration with society for Hematopathology and the European Association for Hematopathology. In 2014, a Clinical Advisory Committee (CAC) proposed revisions to the fourth edition of classification. So, In view of recently identified molecular features, improvisation of morphological features and integrated approach, the fourth edition is being updated in 2016 (Sarwar et al., 2016).

Lymphoid neoplasms are classified as mature B cell neoplasm (MBCN), mature T and NK cell neoplasm (MTCN), Post-transplant lymph proliferative disorders, Hodgkin's disease (HL), Histiocytic and dendritic cell neoplasm (HDN), Myeloproliferative neoplasms (MPN), Myelodysplastic/myeloproliferative neoplasms, myelodysplastic syndrome (MDS), acute myeloid leukemia with related neoplasms (AML), blastic plasmacytoid dendritic cell neoplasms, acute leukemia with ambiguous lineage, B and T lymphoblastic lymphoma/ leukemia (ALL) (Arber et al., 2016).

The causes of hematological malignancies remain unclear, but these are believed to be linked with environmental exposure of chemicals (such as pesticides, benzene, smoking etc.), as well as ionizing radiation and infectious agents. The incidence of HM varies with geography, age and race/ethnicity, suggesting different etiological factors may contribute for the development of these malignancies (Lichtman, 2008; Rodriguez-Abreu et al., 2007). Hematopoietic homeostasis is maintained throughout the lifetime of an individual through self-renewal of hematopoietic stem cells (HSCs). Defects in the self-renewal and differentiation lead to hematopoietic insufficiency and development of malignancies. Leukemic stem cells (LSCs), which are considered to originate from hematopoietic stem or progenitor cells, not only adopt the regulatory machinery operating in normal HSCs but establish their own mechanisms against apoptosis and senescence (Ramdass et al., 2013).

Most population-based studies on the incidence of HMs have grouped these diseases into two broad categories Leukaemia and Lymphoma. The four common types of leukaemia are acute myeloid leukaemia (AML), acute lymphoblastic leukaemia (ALL), chronic myeloid leukaemia (CML), chronic lymphocytic leukaemia (CLL). The two main types of lymphoma are: non-Hodgkin lymphoma (NHL) and Hodgkin's lymphoma (HL). Other common haematological malignancies include myelodysplastic syndrome (MDS) and multiple myeloma (MM) (Hossain et al., 2014; Farhad et al., 2021).

Acute leukemias including AML and ALL are the most prevalent HM affecting Bangladeshi population (Hossain et al., 2014). Leukemia is a malignant neoplasm of the hematopoietic stem cells characterized by diffuse replacement of the bone marrow and/or peripheral blood by neoplastic cells and was identified as a separate malignancy

in 1889. Leukemia is part of a broader group of neoplasms which affect the blood, bone marrow and lymphoid system, known as tumors of the hematopoietic and lymphoid tissues (Vardiman et al., 2009; Paudyal et al., 2018).

Examination of the peripheral blood smear is an inexpensive but powerful diagnostic tool in both children and adults suffering from Leukemia. It provides rapid, reliable access to information about a variety of hematologic disorders. The role of the blood smear in the diagnosis of leukemia and lymphoma is to suggest a likely diagnosis or range of diagnoses, to indicate which additional tests should be performed and to provide a morphologic context without which immune-phenotyping and other sophisticated investigations cannot be interpreted. Peripheral blood analysis by complete blood count and thin smear analysis are first steps to detect most hematologic malignancies which have emerged as a major cause of morbidity and mortality (Bain, 2005).

The diagnosis involves a multi parameter approach including morphologic examination and phenotypic or genotypic studies. The smear offers a window into the functional status of the bone marrow, the factory producing all blood elements. Review of the smear is an important adjunct to other clinical data. In some cases, the peripheral smear alone is sufficient to establish a diagnosis (Bain, 2005; Abdulsalam, 2010).

Overall incidence of hematological malignancies in our country appears to be rising but it is very difficult to describe their epidemiological behavior in a consistent and uniform way. There is no unique reporting system or registry for hematological cancers in our country. In a developing country like Bangladesh, proper number of patients should be known to allocate our limited resource in an equitable way. Bangladesh shows significant capacity in the management of infectious diseases, but chronic diseases in particular hematological malignancies are less prioritized.

1.2 Rationale

Bangladesh is a developing country which despite being scarce in composition, are strong enough to withstand any life-threatening illnesses, including AIDS, infectious diseases, hepatitis, and others. The main barrier today is cancer, with a wide range of symptoms and revealed as a conglomerate of over 100 different diseases accompanied by unchecked cell growth and proliferation that eventually results in death. HM are more common in the developed countries as compared to the developing countries. Peripheral blood smear is an inexpensive but powerful diagnostic tool in both children and adults suffering from HM which involves morphological examination of blood elements. The morphological study of the blood elements provides a peek into the functional status of the bone marrow. Sometimes, study of peripheral blood smear alone is sufficient to establish the diagnosis. Hematological malignancy is 1 of the top 10 malignant diseases with regards to cancer patient morbidity and mortality. Despite very less scientific information, the National Institute of Cancer Research and Hospital bulletin provides a short but overall idea about the current scenario in Bangladesh. Due to the insufficient diagnosis facilities, ignorance, costly treatment, only one-third of the cancer patients have the capability of primary care, treatment, and regular follow-up facilities. Although hematopoietic stem cell transplantation, chemotherapy and targeted therapy have made great progress in recent years, patients with hematological malignancies still have adverse clinical outcomes, particularly elderly patients**.** In case of Bangladesh, there is no published report on HM prevalence and treatment. An understanding of the epidemiological aspects of HM would surely contribute to identify the risk factors in our environmental background and would provide epidemiological basis for devising the cancer care management and preventive strategies of these malignancies. Therefore, it is necessary to explore for an optimal prediction model to evaluate the clinical outcome, which is important for devising a therapeutic strategy for hematological diseases. This study was carried out to assess the patterns of common Hematological Malignancies in Chittagong which might give a detailed insight about the patterns and distribution of hematological malignancies in Chittagong and assist in identifying commonest clinical features that lead to the diagnosis and also state and evaluate the diagnostic tools and its cost effectiveness and availability.

1.3 Research Question

What are the patterns of common hematological malignancies in Chattogram region?

1.4 Objectives

14.1 General Objective:

To see the patterns of common hematological malignancies in Chattogram region

1.4.2 Specific Objectives:

The specific objectives of the study are following:

- To identify socio-demographic distribution of hematological malignancies.
- To identify relative frequency of different types of hematological malignancies.
- To evaluate and identify patterns of clinical symptoms that leads to diagnosis of hematological malignancies.
- To identify commonest investigation tools needed for diagnosis and its availability and cost effectiveness.
- To compare treatment modalities and the availability of treatment options and their cost effectiveness.

Chapter 2

Literature review

2.1 Epidemiology of Cancer

Cancer is predicted to be an increasingly important cause of morbidity and mortality in the next few decades, in all regions of the world. The forecasted changes in population demographics in the next two decades mean that even if current global cancer rates remain unchanged, the estimated incidence of 12.7 million new cancer cases in 2008 will rise to 21.4 million by 2030 (Hussain and Sullivan, 2013).

Hematologic malignancies make up approximately 10% of all cancer types in the USA, and the management of patients suffering from hematologic malignancies has dramatically changed over the last 20 years (Alteri R et al., 2020). In the multinational CONCORD program, which estimates survival from cancer in 1.9 million adults from 101 population-based cancer registries in 31 countries on 5 continents, only 2 of the participating countries were located in Latin America (Curado and de Souza, 2014). Moreover, according to the WHO, only 8% of Latin American populations are covered by cancer registries (Tietsche et al., 2019).

Hematological malignancies are of diverse incidence, prognosis, and etiology. Hematological malignancies originate from the cells of bone marrow and lymphatic system (Farhad et al., 2021). It is a malignant neoplasm of the hematopoietic stem cells characterized by diffuse replacement of the bone marrow and/or peripheral blood by neoplastic cells (Paudyal et al., 2018). Death rates have reduced across the various malignancy types, and rapidly fatal diseases, such as chronic myeloid leukemia, have become curable thanks to therapies like Imatinib Mesylate, occasionally termed an oral ‘magic bullet’(Hochhaus et al., 2017).

There are three major groups: leukemia, lymphoma, and plasma cell neoplasms. In general, in Western countries the overall incidence of hematological malignancies appears to be rising but it is very difficult to describe in a consistent and uniform way

their epidemiological behavior, especially in Europe. The number of European Union (EU) Member States has increased and this expansion enlarged the Union to incorporate a diversity of peoples with a much larger degree of heterogeneity in lifestyle habits and disease risk, as well as in cancer incidence and mortality (either overall or site-specific). Moreover, there is not a unique European reporting system or registry for hematological cancers, so the estimation of the exact number of patients is difficult. In addition, the comparison of the incidences reported by various European registries may be biased because their populations can be predominantly either urban or rural, or because there are differences in the methodology and accuracy of registration. The number of new patients in Europe diagnosed with hematological malignancies in 2005 can be estimated in 230 000 patients. These new cases of leukemia, Hodgkin's and non-Hodgkin's lymphoma (NHL) and myeloma account for 8% of all the new cancer patients diagnosed in Europe and the estimated deaths from these tumors account for 7% of the cancer-related deaths in 2005 (Tietsche et al., 2019).

2.2 Pathophysiology of HM

White blood cells help the body fight off diseases from bacteria, viruses, and other antigens that are trying to harm it. Red blood cells carry oxygen to the body, white blood cells offer immune protection, and platelets help with clotting. All of these processes are grossly disrupted by hematological malignancies, or blood cancers. Stem cells from the bone marrow become blood cells. They then become progenitor cells, which then become either myeloid progenitor cells or lymphoid progenitor cells. Myeloid cells are precursors for red blood cells, white blood cells (i.e. granulocytes), and platelets. Lymphoid cells are precursors for white blood cells (i.e. T and B lymphocytes). Hematological malignancies affect not only the stem and blood cells, but also lymph nodes and other components of the lymphatic system. Essentially, the stem cells are malignant, which make the progenitor cells malignant as well. The causes are highly genetic. Cancer risk assessments and genetic counseling can be provided to those who have a familial predisposition, in addition to preventative measures and long-term follow up (Taylor et al., 2017).

2.3 Types of Hematological malignancies

A progenitor cell either becomes a myeloid or lymphoid progenitor cell. The various types of hematological malignancies can be grouped according to which pathway they travel.

Myeloid, or myelogenous hematological malignancies typically affect the elderly and include:

- Acute myelogenous leukemia - quickly progressing and rare blood cancer that produces too many white blood cells.
- Chronic myelogenous leukemia - slowly progressing and rare blood cancer that produces too many white blood cells.
- Myelodysplastic syndromes - rare bone marrow cancers that inhibits the production of mature blood cells (i.e. red, some white, and platelets).

Lymphoid, or lymphoblastic hematological malignancies affect children and adults and include:

- Acute lymphoblastic leukemia - quickly progressing cancer that produces too many T and B lymphocytes; the most common childhood leukemia.
- Chronic lymphocytic leukemia - the most common adulthood leukemia.
- Lymphomas (Hodgkin and non-Hodgkin) - cancer of the lymphatic system affecting T and B lymphocytes. Includes Hodgkin lymphoma (one of the most curable forms of cancer) and non-Hodgkin lymphoma.

Myelomas - rare cancer of plasma cells that causes cancer cells to crowd out healthy blood cells in the bone marrow and peripheral blood usually affects the middle age and elderly patients. These include

- Multiple Myeloma- monoclonal gammopathy is found in plasma protein electrophoresis
- Light chain disease-presence of kappa, lamda small chain in electrophoresis

Consequences of these disrupting cancers include less oxygen to body tissues, decreased function of the immune system, removing excess fluid from the body via lymph, and decreased ability of the blood to clot. No matter which type of blood cancer a patient has, the effects are similar and can include bone pain, weight loss, fatigue, night sweats, fevers, bruising easily, infections, and pruritus (itchy skin) (De Lange et al., 2020).

2.4 Leukemia

Worldwide, over 250 000 people are diagnosed with leukemia each year, accounting for 2.5% of all cancers. An estimated 75,700 new patients of leukemia will be diagnosed in Europe in 2005. All age groups can be affected; leukemias are the most common pediatric tumor (35% of cancers in children aged 0–14 years). Most cases, however, occur in older adults; more than half after 65 years of age. In the United States SEER registries, from 1998 to 2002, the median age at diagnosis for leukemia was 67 years. Approximately 11% were diagnosed under age 20; 11% between 20 and 44; 10% between 45 and 54; 14% between 55 and 64; 21% between 65 and 74; 23% between 75 and 84; and 10% after 85 years of age (Rodriguez-Abreu et al., 2007).

Leukemias are usually divided into four major categories, with different clinical features and prognosis: acute lymphocytic leukemia, chronic lymphocytic leukemia, acute myelogenous leukemia, and chronic myelogenous leukemia. The incidence rates for all types of leukemia are slightly higher among males than among females. There are also geographical and ethnic variations in leukemia rates. In the United States, the incidence is higher in Caucasians than in Afro-Americans and Hispanics. The American Indians/Alaskan natives have the lowest incidence rates. Trends in overall incidence of leukemia have generally been stable or slowly increasing. A substantial reduction in death rates from acute lymphoblastic leukemia (ALL), particularly in childhood, however, have been observed since the 1970s, thanks to advances in treatment and subsequent improvement in survival (Rodriguez-Abreu et al., 2007).

2.4.1 Acute Myelogenous Leukemia (AML)

AML is mainly an adult's disease with a median age at presentation of 64 years. It accounts for 30% of all leukemias in adults, and 18 000 new patients are diagnosed in Europe each year, representing 0.6% of all cancers. The annual incidence rate in Europe ranges from two per 100 000/year to four per 100 000/year. In the past decade, the trend in overall incidence of AML has generally been stable or slowly increasing in most European countries. Incidence in England and Wales, however, has risen by 70% in both sexes since 1971 (Rodriguez-Abreu et al., 2007)**.**

Acute myeloid leukemia was most frequent (28.3%) with a median age of 35 years, followed by chronic myeloid leukemia with (18.2%; median age 40 years), non-

Hodgkin lymphoma (16.9%; median age 48 years), acute lymphoblastic leukemia (14.1%; median age 27 years), multiple myeloma (10.5%; median age 55 years), myelodysplastic syndromes (4.5%; median age 57 years) and Hodgkin's lymphoma (3.9%; median age 36 years) (Hossain et al., 2014).

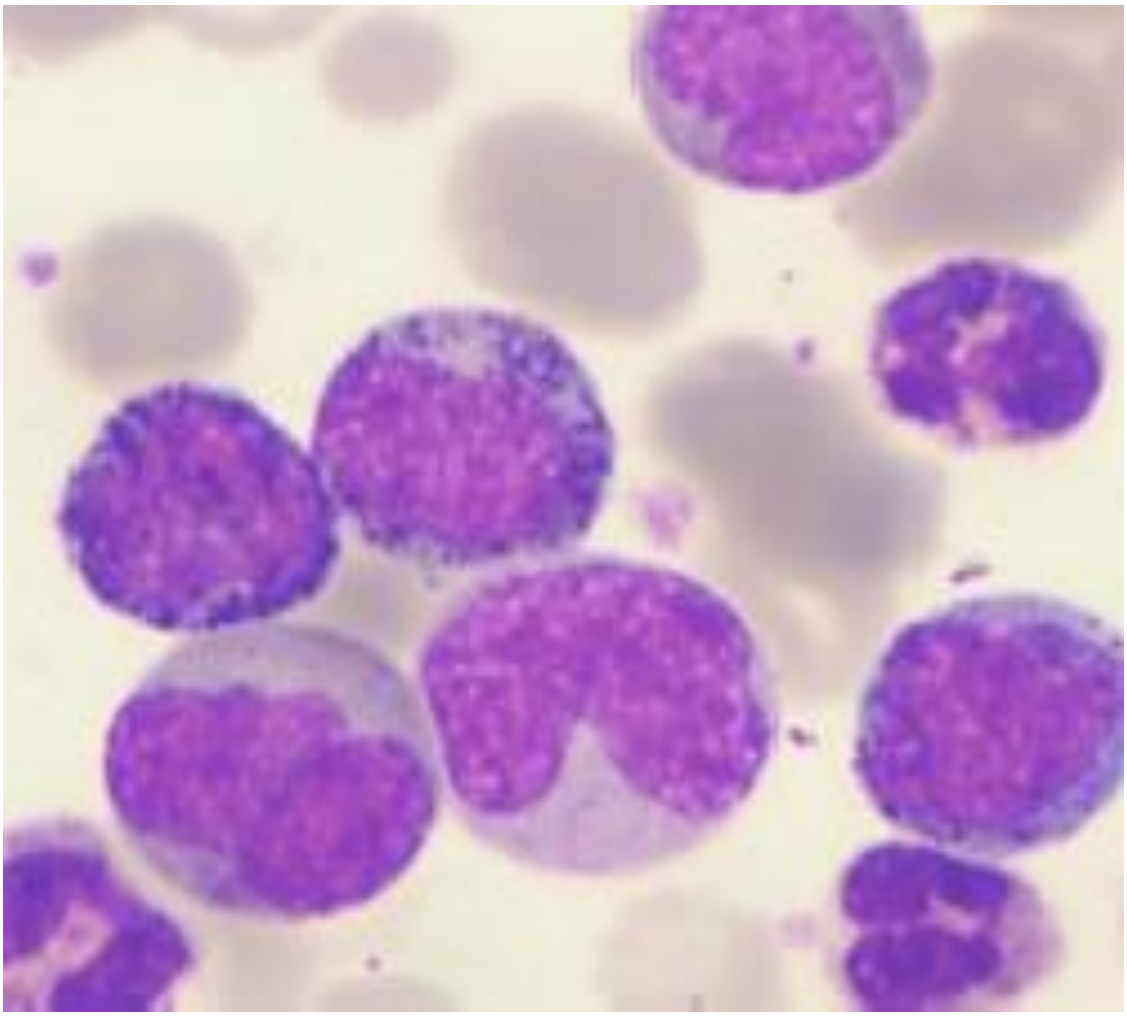

Figure 1: Acute Myeloid Leukemia revealing myeloblasts having enlarged nuclei, opened up chromatin, irregular nuclear membrane and 2-3 prominent nucleoli (Paudyal et al., 2018)

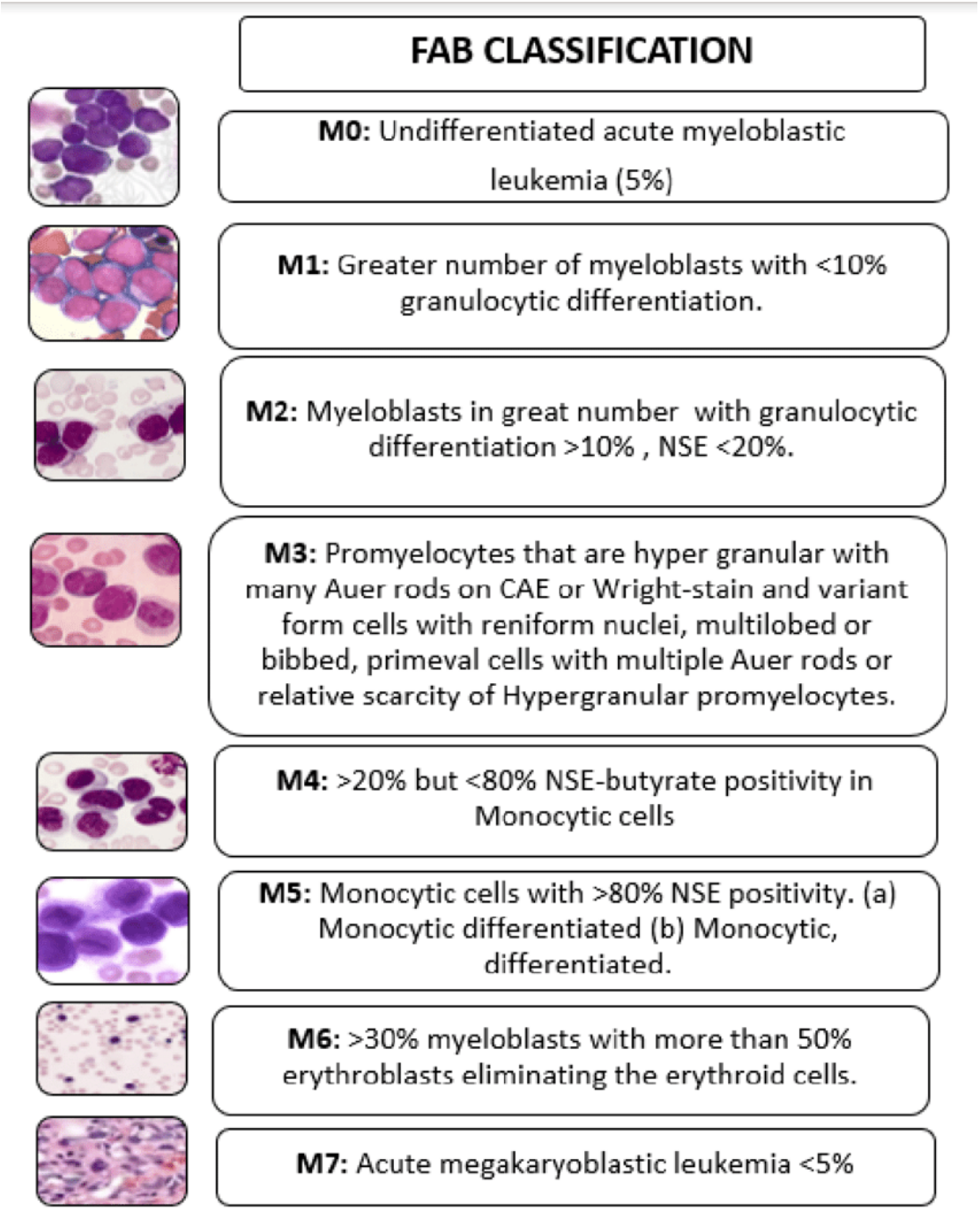

Figure 2: FAB (French American British) Classification of Acute Myeloid Leukemia (Hoffbrand's Essential Hematology, Ed 8th)

2.4.2 Chronic Myelogenous Leukemia (CML)

The least common was chronic lymphocytic leukemia (3.7%; median age 60 years). Below the age of 20 years, acute lymphoblastic leukemia was predominant (37.3%), followed by acute myeloid leukemia (34%). Chronic lymphocytic leukemia and multiple myeloma had mostly occurred among older patients, aged 50-over (Hossain et al., 2014).

Most cases of CML occur in adults with a median age at presentation around age 60. CML comprises only 2-3% of all the leukemias diagnosed in patients <20 years of age but the incidence increases with age slowly until the mid-40s, then more rapidly from about one per 1000 000/year in children <10 years to two per 100 000 in people in the fifth decade to one per 10 000 at age 80. The disease is more common in males. There is no clear evidence of geographical or ethnic background that predisposes to CML; however, in the United States the incidence is slightly higher in Caucasians than in Blacks or Hispanics (Rodriguez-Abreu et al., 2007).

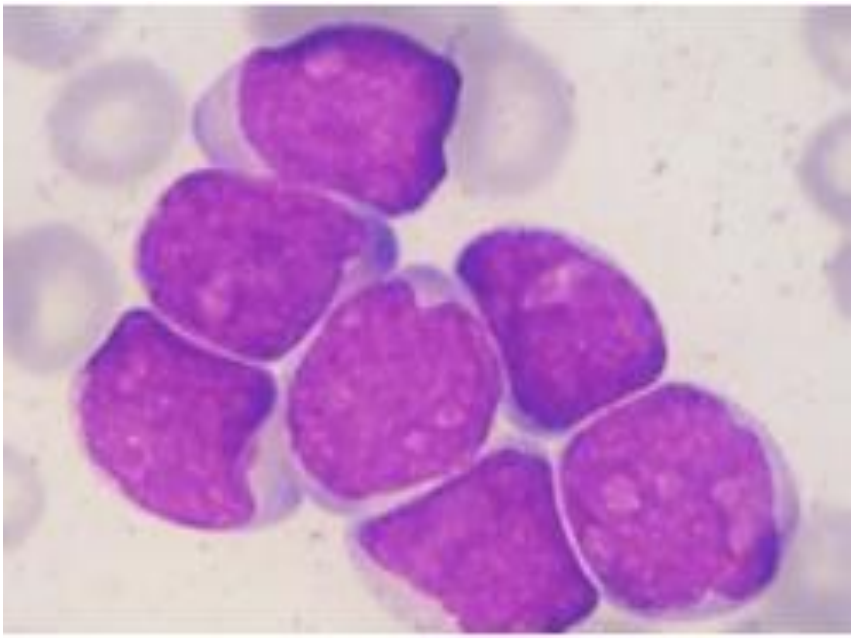

Figure 3: Chronic Myeloid Leukemia revealing leukocytosis and left shift of WBC along with presece of blasts and basophilia (Paudyal et al., 2018)

2.4.3 Acute Lymphoblastic Leukemia (ALL)

Acute Lymphoblastic Leukemia, is uncommon in adults, where it represents 15% of leukemias, but is the most common form of leukemia in people <20, accounting for over 80% of all leukemia patients and for 30% of all cancers in children. The incidence rate of ALL among 1- to 4-year-old children is >10 times greater than in the young adults aged 20–24. About 10 000 new cases are diagnosed in adults in Europe each year, with incidence rates between two and four per 100 000/year, roughly similar to the rates in other developed continents. ALL is slightly more common in men than in women (Rodriguez-Abreu et al., 2007).

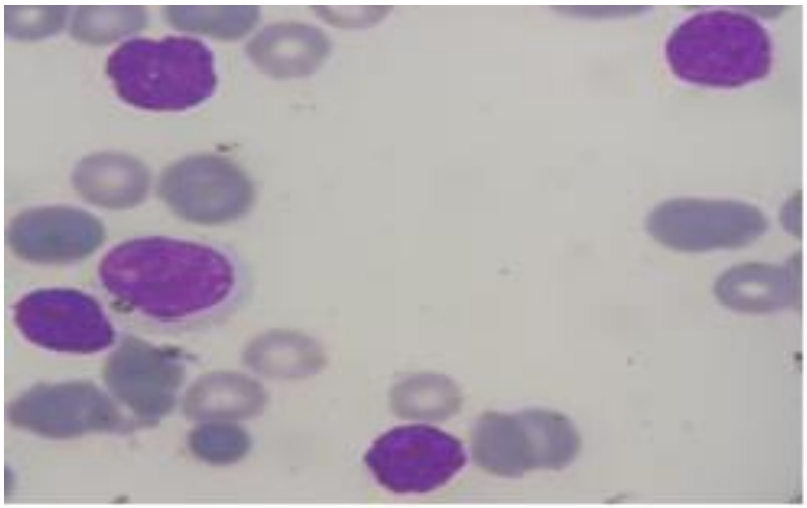

Figure 4: Acute Lymphoblastic Leukemia revealing lymphoblasts with condensed chromatin, inconspicuous to single nucleoli, irregular nuclear membrane and scant amount of cytoplasm (Paudyal et al., 2018)

2.4.4 Chronic Lymphocytic leukemia (CLL)

CLL is a disease of the elderly, with nearly no patients before age 30, 90% of cases occurring after age 50 and a median age of presentation of 70 years. CLL is the most common leukemia in adults in many Western countries, but it is rare in Asia and possibly also in Americans of Asian descent. In the United States, in the 1998–2002 time period, the age-adjusted incidence rate was 3.6 per 100 000/year and CLL was the most common leukemia type among white adult males The incidence of CLL in Japan

is at least 4–5 times lower than that in Western countries. The basis for this ethnic and geographic variation is unknown. In a recent epidemiologic study, neither birthplace nor socioeconomic state accounted for this difference suggesting a role for genetic or other environmental factors (Rodriguez-Abreu et al., 2007).

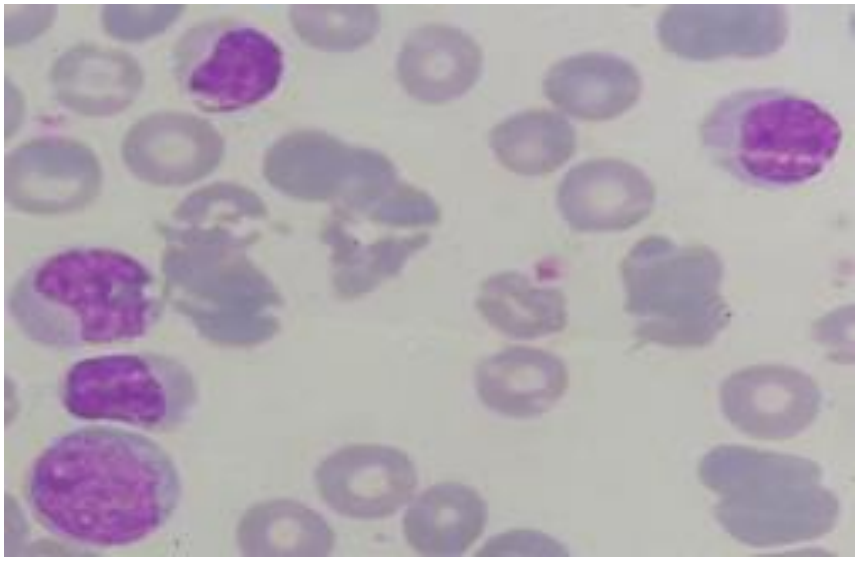

Figure 5: CLL/PLL revealing mature appearing lymphocytes and few larger cells having central prominent nucleoli and scant amount of basophilic cytoplasm (Paudyal et al., 2018).

2.5 Lymphomas

Malignant lymphomas constitute a heterogeneous group of neoplasms deriving from cells of the immune system (either B or T/natural killer (NK) lymphocytes) and primarily arising from lymphoid organs and tissues but also in organs normally devoid of lymphocytes. The term extra nodal lymphoma usually refers to the latter group that comprises about one third of the patients. Malignant lymphomas comprise Hodgkin's lymphoma (HL) and NHLs, which consist of >30 separate disease entities with different morphology, immunologic and genetic profile, and clinical behavior. This variety, as well as the repeated changes over time in lymphoma classification systems, have made it difficult to study their epidemiology. About 120 000 Europeans were diagnosed with lymphoma in 2005, at least three of four of them with NLH, the rest with HL (Rodriguez-Abreu et al., 2007).

2.5.1 Hodgkin's lymphoma

The incidence of HL is about three per 100 000 in Western Europe and the United States, consistently lower than that of NHL and has remained stable over the last 25 years, with possibly a slight decrease of the incidence in males. Lower incidence rates have been reported for Asia, especially Japan and China, suggesting genetic resistance to disease development, possibly associated with human leukocyte antigen type, as well as environmental influences in the etiology of HL. Indeed, recent population-based studies from the United States and several Asian countries, showed a quite low incidence in all Asian subgroups. HL incidence rates were approximately two times higher in the United States Asians than in native Asians and in both groups, rates were lower for Japanese and Chinese than for Filipinos and Asian Indians. Hodgkin's disease can occur in both children and adults. It is more common, however, in two age groups: early adulthood (age 15–40, usually 25–30) and late adulthood (after age 55). About 10% of cases are diagnosed in young boys before 15 years of age but the disease is very rare before 5 years of age (Rodriguez-Abreu et al., 2007).

2.5.2 Non-Hodgkin's lymphomas

The overall incidence of NHL has steadily risen in most developed areas of the World between the 1970s and the late-1990s. While other cancers have increased 25% in the past 25 years, NHL has increased >80%; in the population over the age of 65, the rate of NHL has tripled. Some of the rise may be related to Acquired Immunodeficiency Syndrome (AIDS) and some may be the results of better diagnosis but the causes of this long-term increase are largely unknown, though, age-related immunodeficiency is likely involved. In the early 2000s, NHL in Western countries has become in general the sixth most common cancer in males (after prostate, lung, colon and rectum, bladder and melanoma) and the fifth most common cancer in females (after breast, lung, colon and rectum, uterine corpus). It represents the third most common neoplasm group in children (after ALL and central nervous system tumors). The incidence of NHL is usually slightly higher in men than in women and this difference is more marked in younger than older individuals. The most common histological subtype, diffuse large B-cell lymphoma, accounts for 40% of NHL and occurs more frequently among males than females at middle age and among whites than blacks at older ages. Follicular lymphomas account for 20% of NHL, are more common in whites and occur almost equally in men and women. Distribution of the most common subtypes of NHL (diffuse

large B cell and follicular) appears to differ by geographic region, suggesting differences in etiologic or host factors. The difference is particularly striking for follicular NHL, which is most common in North America and Western Europe, and for Burkitt's lymphoma, which is endemic in equatorial Africa, but constitutes only 1%–2% of lymphomas in the United States and Western Europe. In Western countries, NHL is more commonly of B-cell origin; a higher frequency of T-cell diseases is seen in the Far East. In the majority of NHL patients, the disease arises in lymph nodes, but primary extra nodal disease accounts for 30% of new lymphoma patients and often present as localized disease. The most frequent primary extra nodal sites are the stomach, small intestine, skin, and brain. Incidence rates increased 3.0%– 6.9% per year for extra nodal cases, compared to 1.7%–2.5% per year for nodal cases (Rodriguez-Abreu et al., 2007).

2.6 Plasma cell neoplasms

Multiple myeloma (MM) is a plasma cell malignancy that accounts for <1% of all cancers and for 10% of the hematological malignancies. MM primarily affects older individuals with the median age of onset of 65–70 years. The incidence of MM has been increasing over the past several decades as the elderly population has increased. Incidence rates are similar in Europe and in the United States. In the EU, the estimated incidence of MM is 5.7 per 100 000/year. In the United States, the age-adjusted incidence rate in the 1998–2002 SEER registries were 5.5 per 100 000 men and women per year. The incidence in African Americans is markedly higher than in whites. The disease is more common in men for all ethnic groups with a male/female ratio of 1.5 : 1 (Rodriguez-Abreu et al., 2007).

2.7 Etiologic factors

The etiology for HM is at present largely unknown. Several risk factors, however, have been shown by epidemiological studies to be associated with the development of these diseases (Rodriguez-Abreu et al., 2007).

2.7.1 Leukemia

The exact cause of most cases of leukemia is not known. Studies, however, have found a number of conditions that can be associated with a higher risk of leukemia.

a. Ionizing radiation

Several studies of populations exposed to ionizing radiation as a result of military (either participants at nuclear weapons tests or atomic bomb survivors) or occupational circumstances (nuclear workers and people exposed to nuclear power plant accidents) have provided adequate evidence of the association between exposure to a certain level of radiation and development of acute leukemias and chronic myeloid leukemia. Nevertheless, different cell types may have different responses to radiation. The epidemiologic evidence of a link between ionizing radiation and CLL remains weak. Medical irradiation can be another source of exposure. If very high doses are given to a limited tissue volume, cell killing will, however, predominate over cell transformation and secondary leukemia is a relatively rare event after radiotherapy for cancer. The doses associated with diagnostic radiation procedures are generally very small and not linked to an increased leukemia risk (Rodriguez-Abreu et al., 2007).

b. Electromagnetic fields

In the recent past, a number of reports have indicated that strong electromagnetic fields may be a risk factor for leukemia, although other investigations have failed to confirm these findings. To date, most publications indicate that leukemia seems not related to the exposure to electromagnetic fields (Rodriguez-Abreu et al., 2007).

c. Chemicals

Professional exposure to benzene, formaldehyde, and dioxins is associated with greater risk of leukemia. Organic solvents, agricultural pesticides, and herbicides have been also associated with higher risk. Cigarette smoking is a known lifestyle-related risk factor for leukemia. Potential leukemia-causing carcinogens in tobacco smoke include benzene, polonium-210, and polycyclic aromatic hydrocarbons. It is estimated that 20% of AMLs can be associated with cigarette smoking (Rodriguez-Abreu et al., 2007).

d. Medical therapy and medical conditions

A previous cancer treatment with both chemotherapy and radiation therapy increases the risk of ‘secondary’ leukemias. Among anticancer drugs, alkylating agents such as melphalan, busulphan, procarbazine, chlorambucil, and cyclophosphamide and topoisomerase II inhibitors such as mitoxantrone, etoposide, and teniposide have been most commonly associated with the risk of development of secondary myelodysplasia

and AML. The risk is higher for combined modality therapy and is well documented for Hodgkin's disease, NHLs, breast, ovarian, and testicular cancers. Leukemias and lymphomas have been observed in recipients of organ transplants. Aplastic anemia and myelodysplastic syndrome are associated with an increased risk of leukemia (Rodriguez-Abreu et al., 2007).

g. Genetic disorders

Certain immunological conditions and genetic disorders characterized by chromosomal alterations such as Bloom's syndrome and ataxia telangiectasia appear to predispose to leukemia. Klinefelter's syndrome and Down's syndrome are also associated with a greater risk of leukemia (Rodriguez-Abreu et al., 2007).

h. Infections

The oncogenic retroviruses Human T-cell leukemia virus (HTLV) type-1 and type-2 have been identified as being related to the development of rare types of leukemia and lymphoma. HTLV-1 is endemic in certain areas of Japan, the Caribbean islands areas, Central and South America, and central Africa, and is associated with the development of adult T-cell leukemia or lymphoma (ATLL), which accounts for about half of the lymphoid malignancies in the endemic areas. The virus is transmitted mainly from mother to child, especially by breastfeeding. Sexual transmission and blood transfusion are minor routes of infection and cell-free blood products are not infectious. Over one million people in Japan are infected with the virus, and 10–20 million worldwide, but <1000 new patients with ATLL are diagnosed each year. The lifetime risk of developing ATLL is 0.5%–7%, with the highest risk associated with neonatal infection. Low incidence and long latency strongly indicate the accumulation of other genetic mutations is needed for induction of ATLL (Rodriguez-Abreu et al., 2007).

2.7.2 Lymphoma

Largely lymphomas are categorized into two typesnamely Hodgkin's Lymphoma and Non-Hodgkin's Lymphoma.

2.7.2.1 Hodgkin's lymphoma (HL)

HL is more common in men than in women and the main risk factors are a family history and a previous Epstein–Barr virus (EBV) infection. Brothers and sisters of HL patients have a higher-than-average chance of developing this disease, non-familial

clusters of HL have been observed too, but the evidence for common etiological factors behind such clusters is weak. Various findings are suggestive of EBV being a factor contributing to oncogenesis in HL. Indeed, the risk of developing the disease is increased up to four-fold in patients with a previous history of infectious mononucleosis, a disease of adolescence caused by EBV. Moreover, HL patients have increased EBV antibody titers at the time of diagnosis and several years before the onset of lymphoma. More definitive evidence for a pathogenic association is provided by the finding that EBV can be detected in 50% of classic HL patients in developed countries. The extent to which occupational factors can increase the risk for HL has not been established; organic solvents, phenoxy herbicides, and wood dust may be involved but the epidemiological evidence is limited and controversial (Rodriguez-Abreu et al., 2007).

2.7.2.2 Non-Hodgkin's lymphoma (NHL)

There are a few well established and a number of postulated risk factors that can be implicated. Immunodeficiency, including both congenital and acquired conditions is the most powerful etiologic factors. NHL is the most frequent malignancy associated with ataxia-telangiectasia or the Wiskott–Aldrich syndrome in young persons, as well as with X-linked lymphoproliferative syndrome or combined immunodeficiency in children. EBV seems to be a cofactor in the NHL development associated with congenital immunodeficiency. These disorders, however, are quite uncommon and their contribution to the NHL increase is limited Rodriguez-Abreu et al., 2007).

a. Immuno suppressive drugs

Patients treated with immunosuppressive drugs are at much greater risk for NHL. Polyclonal B-cell proliferation is often seen in organ transplant recipients (either solid organ or allogeneic bone marrow/PBSC) and it can evolve into a monoclonal disease but can sometimes be reversible if immunosuppressive medication is discontinued. Loss of immune control of persistent EBV infection seems to be part of the process (Rodriguez-Abreu et al., 2007).

b. Autoimmune Disease

An excess risk of developing NHL has also been reported among patients with a variety of autoimmune diseases, including rheumatoid arthritis, Sjogren's syndrome, systemic

lupus erythematosus, and celiac disease. Patients with rheumatoid arthritis or polymyositis treated with methotrexate (MTX) develop EBV-positive lymphomas more frequently than patients treated with other agents that should be equally or more immunosuppressive. It has been suggested that this predisposition may be due to the ability of MTX to induce EBV replication while at the same time producing immunosuppression. Transplant-related NHL as well as those associated with autoimmunity disorders are also relatively rare and cannot explain the rise of NHL in the general population (Rodriguez-Abreu et al., 2007).

c. Associated with HIV

NHL represents one of the most common malignancies associated with AIDS. The risk of NHL in persons infected with human immunodeficiency virus (HIV) is >100 times higher than that in the general population. These lymphomas are typically of B-cell origin with high-grade histology (diffuse large cell or Burkitt's lymphoma) and frequently occur in extra nodal sites, such as the brain. HIV-related NHL accounts only for a relatively small fraction of NHL. Moreover, with the arrival of HAART, after the mid-1990s, the incidence of HIV-associated NHL decreased among HAART users and their survival improved. In a Swiss cohort study, incidence rates and risk of HL, however, have apparently slightly increased in recent years both in men and women with HIV. Since random variation cannot be ruled out, the increase in HL risk among HAART users requires confirmation in other studies with longer post-HAART follow up (Rodriguez-Abreu et al., 2007).

d. Viruses other than HIV

Viruses other than HIV can contribute to pathogenesis of NHL. Some viruses are implicated in the pathogenesis of NHL, probably because of their ability to induce B- or T-cell stimulation and proliferation. EBV infection unlike HTLV-I, is a highly prevalent infection in the adult population, has been associated with a heterogeneous group of lymphomas, including Burkitt's lymphoma (especially the endemic form in Africa), Hodgkin's disease, NK, and T malignancies with cytotoxic phenotypes, and lymphomas in the immunocompromised patient (congenital immunodeficiency, organ transplantation, AIDS). The role of EBV in contributing to lymphomagenesis is well established in the EBV lymphoproliferative diseases that arise in immunosuppressed individuals. It is, however, less well defined in other EBV-associated lymphomas.

Usually, these malignancies respond poorly to standard chemoradiotherapy and immunotherapeutic or pharmacologic strategies targeting EBV are being explored. Hepatitis C virus (HCV) has been associated with mixed cryoglobulinemia type II and with B-cell lymphomas, especially the splenic B-cell marginal zone lymphoma and the lymphoplasmacytic lymphoma, but also with diffuse large cell lymphomas. The significance of the epidemiologic association between HCV and NHL shows a clear geographic variability, being the association with NHL more evident in the areas (e.g. Italy) with elevated endemic rate of HCV infection. A strong support for the etiological role of HCV in splenic marginal zone lymphoma came from the demonstration of lymphoma regression after HCV treatment. Human herpes virus 8 (HHV 8) is associated with primary effusion lymphoma in patients with HIV infection and in patients with multicentric Castleman disease. Other viruses (including HHV 6, simian virus 40, HTLV-II and CMV) have been inconsistently reported as associated with lymphomas (Rodriguez-Abreu et al., 2007).

e. Bacterial Agents

There are some Bacterial agents. A significant association has been reported in epidemiological studies between Helicobacter pylori infection and gastric lymphomas with either extra nodal marginal zone B-cell lymphomas or diffuse large B-cell lymphomas; low grade or high grade. Moreover, the presence of the B-cell clone that will become predominant in the transformation to MALT lymphoma has been demonstrated in the chronic H. pylori gastritis that preceded the lymphoma and several clinical studies have reported regressions of gastric MALT lymphoma in more than half of the treated patients after antibiotic eradication of H. pylori. The association of H. pylori with gastric MALT lymphoma has led to the hypothesis that the microorganism may provide the antigenic stimulus for sustaining the growth of the lymphoma in the stomach. Besides H. pylori, other infectious agents are being associated to particular extra nodal marginal zone B-cell lymphomas. Borrelia *burgdorferi* may be implicated in the pathogenesis of at least a subset of cutaneous marginal zone B-cell lymphomas. The microorganism can be found in skin lymphomas and a lymphoma complete remission can be achieved with adequate antibiotics therapy. Recently, the presence of Chlamydia *psittaci* has been associated with lymphomas of the ocular adnexa and it has been showed that antibiotic therapy aimed at C. *psittaci* can be followed by histological regression of these NHL (Rodriguez-Abreu et al., 2007).

f. Geographical Variability

A wide geographical variability, however, has been found for the epidemiologic association between NHL C. *psittaci* and B. *burgdorferi*. Since the 1970s it was already known that early-stage immunoproliferative small intestine disease, also known as alpha chain disease or Mediterranean lymphoma, may regress after antibiotic therapies eliminating unknown organisms, but only in 2004 this lymphoma has been linked to a specific pathogen, namely Campylobacter *jejuni*. All these data, together with the pattern of somatic hypermutation and ongoing mutations of the immunoglobulin genes, strongly associate the origin of extranodal marginal zone lymphomas from a background of chronic antigenic stimulation associated with infectious conditions (and/or autoimmune conditions) (Rodriguez-Abreu et al., 2007).

g. Exposure to Pesticides and Chemicals

Exposure to agricultural pesticides and other chemicals has an association with NHL. The professional use of herbicide, insecticides, and fertilizers has been reported by several studies with high variability of risk estimates among different studies. Occupational exposure to solvents has also been associated with an increased risk. Epidemiological evidence indicates that exposure to the class of chemicals called organochlorines, which includes DDT and other pesticides, polychlorinated biphenols, and dioxins, may result in increased risk of NHL. The association between NHL and pesticide exposures is perhaps the best studied topic. An increased risk of NHL has been observed repeatedly, but not consistently. Results from a number of epidemiologic studies indicate that the excess risk of NHL is related to the use of phenoxyacetic acid herbicides, organophosphate insecticides, triazine herbicides, and fertilizers. Risk estimates, however, vary widely among studies, and in some studies no risk excess was detected; therefore, the role of agricultural and residential pesticides in the etiology of NHL needs further evaluation (Rodriguez-Abreu et al., 2007).

h. Ultra violet (UV) exposure

UV exposure is another important etiological factor. Higher risk for developing squamous cell skin cancer and melanoma was reported among patients with NHL and CLL and conversely, patients with squamous cell skin cancer showed an excess risk for NHL and CLL. Therefore, it has been hypothesized that increased exposure to solar UV may have contributed to the incidence rise of NHL in many countries. Most recent

studies, however, showed that sun exposure is associated with a decreased risk of NHL, therefore, the association between skin cancer and malignant lymphomas is unlikely to be mediated by UV exposure (Rodriguez-Abreu et al., 2007).

i. Dietary and lifestyle risk factors

The risk of NHL has been linked to increased consumption of animal protein, fat, and meat but the data on diet are not conclusive. Some epidemiologic studies have evaluated the role of alcohol consumption in the etiology of NHL, but the findings have been inconsistent. A meta-analysis of nine case–control studies from the United States, United Kingdom, Sweden, and Italy with a pooled study population of >15 000 individuals, however, found decreased odds ratios among consumers of alcohol. Further studies are needed to determine whether confounding lifestyle factors or immunomodulatory effects of alcohol explain this finding. Cigarette smoking appears to have no or only a weak association with increased risk of NHL (Rodriguez-Abreu et al., 2007).

j. Hair coloring products

Hair coloring products are widely used and contain compounds that are mutagenic and carcinogenic in animals. Several studies have indicated that exposure to hair dyes, particularly long-term use of dark permanent dyes produced before the 1980s, is associated to a moderately increased risk of lymphoma and chronic lymphocytic leukemia (Rodriguez-Abreu et al., 2007).

k. Blood transfusions

Allogeneic blood transfusion has been indicated as a risk factor for NHL, possibly because they can expose the recipients to viruses or other immunomodulating antigens, but the results from epidemiologic studies have been inconsistent. While cohort studies supported the hypothesis, several case–control studies subsequently carried out failed to confirm the findings (Rodriguez-Abreu et al., 2007).

l. Genetic susceptibility

Genetic susceptibility is also an important etiology in this regard. Familial clustering of NHL has been described; it accounts only for a small proportion of NHL and may be due to inherited immune function abnormalities but also to shared exposure to environmental factors. Polymorphisms in genes regulating the inflammatory and the immune response or regulating the antioxidative mechanisms may influence the risk of lymphoma, but there are no conclusive data (Rodriguez-Abreu et al., 2007).

2.7.3 Plasma cell neoplasms

Increasing age and a previous monoclonal gammopathy of undetermined significance (MGUS) are the main factors associated with MM. In most cases patients with MM have no known risk factors. Several studies have indicated an association between MM and environmental exposure to chemicals. Slightly higher rates of MM are found among agricultural workers (who are exposed to pesticides and fertilizers), petrochemical and sheet-metal workers, and those professionally exposed to wood dust. The role of viral infections is controversial. In rare cases, MM and MGUS can affect more than one person in a family (Rodriguez-Abreu et al., 2007).

2.8 Diagnosed hematological malignancies in Bangladesh

Hematological malignancies can occur at any age group and men are more commonly affected. Lymphoid neoplasm is the most frequent HM in Bangladesh. NHL is the most common HM in young adults whereas AML is the most common HM in the elderly. Hematological malignancies arising from Precursors T and B cells are common in children and adolescents whereas lymphoid neoplasms arising from the germinal center and memory B cells are common in adults (Sarwar et al.). Hossain et al. showed differences in population distributions as compared to other settings with possibly a lower presence of non-Hodgkin lymphoma. There might be under-reporting of affected women (Hossain et al., 2014).

2.9 Diagnostic tools

Full blood count or FBC testing is a routine test that evaluates three major components found in blood: white blood cells, red blood cells and platelets. There are many reasons for a full blood count test, but common reasons include infection, anemia and suspected haemato-oncological diseases (Hicks et al., 2014).

The first step in the diagnosis involves peripheral blood smear which involves morphological examination of blood elements. The morphological study of the blood elements provides a peek into the functional status of the bone marrow. Sometimes, study of peripheral blood smear alone is sufficient to establish the diagnosis but at other times, it can be later confirmed by genotypic studies of the specimen (Paudyal et al., 2018). Although hematopoietic stem cell transplantation, chemotherapy and targeted therapy have made great progress in recent years, patients with hematological malignancies still have adverse clinical outcomes, particularly elderly patients (Ruppert et al., 2020).

Diagnosis of acute leukemia in Iraq is mainly dependent on the personal experience of the laboratory physician. Local guidelines in this field were never proposed and the international guidelines are very difficult to apply as the only available techniques include morphology of peripheral blood and bone marrow specimens plus very limited immunohistochemistry CD markers and PCR testing for BCR-ABL oncogene (Abdulsalam, 2010).

With Romanowsky stain morphology AML- M2, M3, M4, M5b and M6 can be recognized readily. By adding few special stains such as Sudan black B (SBB) (and not myeloperoxidase as SBB has a little more sensitivity in detecting myeloblasts which is the crucial point), plus a non-specific esterase stain as α-naphthyl acetate esterase it becomes possible to recognize AML-M1 and most cases of AML-M5a. The AML cases that cannot be distinguished by morphology and cytochemistry, specifically M0 and M7, for which the presence of myeloid dysplasia in the former and the cytoplasmic blebs in the latter may give a hint for the probable diagnosis, however there is still the need for more positive diagnostic technique. When the flow cytometry, immunophenotyping is not available then the use of a limited number of CD markers study by immunohistochemistry to identify the lineage of acute leukemia is the option. These include CD33, anti-myeloperoxidase and CD41. Rare types of AML like M5c require high degree of morphology experience, in which malignant cells appearance is reminiscent of tissue histiocytes (Abdulsalam, 2010).

2.10 Prevention

The control and prevention of hematological malignancies will require a better understanding of the origins of the diseases. Avoiding exposure to risk determinants would result in a reduction in cancer risk. The little we know about the risk factors has not yet been translated into consistent attempts to prevent hematological malignancies. Some general consideration, however, can be done (Rodriguez-Abreu et al., 2007).

i. Avoidance of exposure to radiation and benzene will reduce the risk of leukemias.
ii. Banning or restricting the use of organochlorines might on a long-term result in reduction of the NHL incidence.
iii. Prohibition of smoking-about 20% of adult acute myeloid leukemia cases are linked to smoking and here prevention is possible.
iv. HIV infection is clearly preventable and also other infectious agents associated with NHL might be the target of preventive measures.

Finally, more modern and improved cancer therapies will likely result in a lower incidence of second tumors (Rodriguez-Abreu et al., 2007).

Table 1 An Overview of widely used chemotherapeutic drugs in various HM and their mechanism of action (Chabner et al., 2011)

Chemo Therapeutic Drugs	Mechanism of Action
Doxorubicin	Disruption of topoisomerase-II-mediated DNA repair
Bleomycin	Binds to guanosine-cytosine-rich portions of DNA and mediates free radical generated DNS break down
Vinblastine	Binds to microtubular proteins in the mitotic spindle, thereby preventing cell division during metaphase
Dacarbazine	Alkalyzing Agent
Cyclo phosphamide	Alkalyzing Agent
Vincristine	Block cell growth by stopping mitosis by interfering with microtubule polymerization
Prednisolone Dexamethasone	Produces its anti-cancer effects through its anti-inflammatory processes.
Daunorubicin	Inhibit the topoisomerase II enzyme
Cytarabine	Hinders the rotation of the molecule within the DNA. The process of DNA replication ceases, specifically during the S phase of the cell cycle
Arsenic Trioxide (ATO)	Degradation of the fusion protein PML/RARA.
Vesanoid (ATRA)	Firstly, Degradation of PML-RARA protein via caspase-mediated cleavage and proteosome-dependent degradation Secondly, conversion of PML-RARA from a transcription repressor (CoR) to a transcription activator (CoA) under therapeutic concentration of ATRA
Bortezomib	Proteasome inhibitor which reversibly binds to the chymotrypsin-like subunit of the 26S proteasome, resulting in its inhibition and preventing the degradation of various pro-apoptotic factors.
Linamide	Induces tumor cell apoptosis directly and indirectly by inhibition of bone marrow stromal cell support, by anti-angiogenic and anti-osteoclastogenic effects, and by immunomodulatory activity.
L asparginase	Deplete plasma levels of asparagine in leukemic cells by converting L-asparagine to L-aspartic acid and ammonia leading to reduced DNA, RNA and protein synthesis
Methotrexate	Developed as a folic acid analogue, methotrexate inhibits purine and pyrimidine synthesis, which accounts for its efficacy in the therapy of cancer
6-Mercapto purine	Inhibits de novo purine synthesis and acts as an antiproliferative agent by interfering with protein, DNA and RNA synthesis and promoting apoptosis.

Chapter 3

Materials and Methods

3.1 Study design

A retrospective cross sectional study was done on the patients of hematological malignancies enrolled for treatment in the department of hematology, Chattogram Medical College Hospital for the period of 6 months from 1st January 2022 to 30th June 2022. A questionnaire was constructed to fill up the information on patient's socio-demographic characteristics including age, sex, education, occupation, monthly income etc. Information on clinical features, laboratory findings, treatment modalities and their availability and cost effectiveness was also sorted out after person to person interviewing and evaluating patient's previous treatment record files.

3.2 Study population:

Patients between 02-90 years of age admitted in hematology department of Chattogram Medical College suffering from hematological malignancy were included in this study.

3.3 Sampling technique:

Consecutive type of purposive sampling was done according to the availability of the study subjects who fulfilled the inclusion criteria.

3.4 Sample size determination:

To conduct this study, the sample size was determined by using this formula. To estimate single proportion : (Haque, 2009)

$$Sample\ size\ n = \frac{Z^2 pq}{d^2}$$

z = z value of standard normal distribution at given level of significance or 95% Confidence level, Z= 1.96

p= Expected prevalence of hematological malignancy = 0.25

q = 1-p

d = Absolute error or precision (0.5)

All values are taken from (Sarwar et al., 2016)

$$Therefore, \quad \frac{(1.96)^2 \; x \; 0.25 \; (1 - 0.25)}{(0.05)^2}$$

Sample size (n) = 288

Due to time and resource constrains 200 participants were enrolled in this study.

3.5 Selection criteria:

Inclusion criteria:

- Patients aged between 02-90 years diagnosed with hematological malignancy at Chattogram Medical College Hospital.

Exclusion criteria:

- Patients with other than hematological malignancy.
- Respondents who did not give informed written consent and not willing to participate.

3.6 Research instruments:

A semi structured questionnaire with data collection sheet was prepared for this purpose, which included all the variables of interest.

3.7 Data collection technique:

Subjects were selected purposively according to the availability of the respondents. Relevant history and clinical information were obtained by a preformed questionnaire.

3.8 Study procedure:

After obtaining institutional ethical approval from the authorized committee of Chattogram Veterinary and Animal Sciences University (CVASU), Bangladesh, this retrospective observational cross sectional study was conducted among patients aged between 2-90 years admitted in hematology department of Chattogram Medical College Hospital suffering from hematological malignancies. This study was based on the recorded diagnosis of HM. The source of the information included bone marrow morphology databases from the hematopathology laboratory of hospital as well as inpatient and outpatient case registers maintained at the participating hospital. The participating hospital mostly provided both pathological diagnoses and clinical management information. A total 200 patients were enrolled in this study. Data was collected from patient registry books of CMCH. Diagnosis were carried out on the basis of clinical features, blood counts, peripheral blood films and bone marrow morphology including flowcytometry and immunohistochemistry where available. Morphological typing of malignancies was carried out according to "French American British (FAB)" classification system. Although WHO classification (2008) for hematological malignancies is based mostly on immunophenotype and molecular markers, we had to stick to FAB classification because we had limited facilities for immunophenotyping or cytogenetic studies. The diagnoses were supplemented by criteria from the 2008 WHO classification whenever facilities were available to fulfil the criteria. Hematological malignancies were classified according to their cell lineage and all of the patients who were diagnosed with HM was taken for study. From the registry book categorization and identification of various patterns of HM and its age and sex distribution was done. Information regarding clinical features, investigation tools, treatment modalities and its availability and cost effectiveness was obtained through a questionnaire. All data was evaluated and the findings was recorded in a case record form. We interviewed participants to collect information after receiving written consent. Answering a questionnaire was part of the study procedure. The questionnaire included socio-demographic details, clinical features, laboratory findings and various treatment modalities in regard of various types of HM. Participants were included in the study who fulfilled the inclusion criteria.

3.9 Data analysis:

Recorded data were entered into MS Excel 2013, coding, decoding, sorting was done and data was then exported to the Statistical Package for Social Services, SPSS version 23. Descriptive statistical analysis were performed by calculating mean, standard deviation. Qualitative data was analyzed using chi-squared test. Quantitative data was analyzed using student's t- test. The results were presented as tables and charts. P value less than 0.05 was considered as significant.

3.10 Ethical implications

When approaching the study participants, a written informed consent was obtained from all participants or their relatives if participants were unable to respond. All collected data was kept in a secured place under lock and key. Institutional ethical approval was taken from the authorized committee of Chattogram Veterinary and Animal Sciences University (CVASU), Bangladesh. Permission for the study was taken from the concerned department from where we collected our study subjects. The entire study subject was thoroughly appraised about the nature, purpose and implications of the study, as well as the entire spectrum of benefits and risks of the study. The interest of the study subjects was not compromised to safeguard their rights and health. Subjects were assured about their confidentiality and freedom to withdraw them from the study anytime. No invasive procedures were performed on the study subjects. For safeguarding confidentiality and protecting anonymity, each of the patients was given a unique ID number.

Chapter 4

Results

Part A

Overall Scenario of HM patients in Chattogram

Table 2: Socio-demographic characteristics of the patients with HM (N= 200)

Variable	
Age (Mean ± SD) (in Years)	36.03±18.07
Sex (Percentage)	
Male	55.9
Female	43.1
Area (Percentage)	
Rural	60.5
Urban	39.5
Education (Percentage)	
Primary Level	32
Secondary Level	20
Higher Secondary Level	10
Graduation Level	25
No Educational qualification	12.5
Post-Graduation and beyond	0.5
Occupation (Percentage)	
Student	20
Farmer	9.5
Plumber	0.5
Driver	2
Fisherman	5.5
Carpenter	3
Brick Field Worker	0.5
Garments Worker	8
Dry Fish Factory Worker	1
Service Holder	16
Businessman	6.5
Retired	0.5
House wife	18
Teacher	8
Unemployed	1
Monthly Income	
Monthly family income (BDT) minimum (Mean ± SD)	20850.00±20503.16
Monthly family income (BDT) maximum (Mean ± SD)	44882.05±26113.97

(HM=Hematological Malignancy)

Table 2 describes the frequency distribution of socio-economic characteristics of the respondents. The mean age of the respondents was 36.03±18.07 years. No significant age variations were seen in respect of HM. Majority of respondents were male (55.9%) whereas females were about (43.1%). 60.5% of respondents were from rural areas compared to 39.5% of urban dwellers. Most of the respondents have completed only primary school (32%) followed by graduation (25%), secondary education (20%), and higher secondary education (10%), post-graduation (0.5%) and one-fourth of the respondents were illiterate (25%). Most of the respondents (20.0%) were students followed by housewife (18%), service holder (16%), farmer (9.5%), garments worker (8%), teacher (8.0%), businessman (6.5%), fisherman (5.5%), carpenter (3%), driver (2%), dry fish factory worker (1%), plumber (0.5%), retired person (0.5%) and (1%) unemployed.

Table 3: Different types of Hematological Malignancies (N= 200)

Category	Diseases	Percentage
Leukemia (n=143)	Acute Myeloid Leukemia (AML) (Total) 1. AML (except M3) 2. AML (M3)	**33** 23 10
	Acute Lymphoblastic Leukemia (ALL)	**26**
	Chronic Lymphocytic Leukemia (CLL)	**2**
	Chronic Myeloid Leukemia (CML)	**7**
	Myelodysplastic Syndrome (MDS)	**1.5**
	Mixed Leukemia	**2**
Lymphoma (n=41)	Hodgkin Disease (HD)	**8.5**
	Non Hodgkin Lymphoma (NHL)	**12**
Myeloma (n=16)	Multiple Myeloma (MM)	**8**

Table 3 illustrates the frequency distribution of HM patients according to their disease where AML was the leading pattern with (33%) cases, one-fourth of the respondents were suffering from ALL (26%) followed by NHL (12%), HD (8.5%), MM (8%), CML (7%), CLL (2%), Mixed Leukemia (2%) and MDS (1.5%). As the patterns of presentation, mode of treatment and outcome varies significantly from other types of AML, AML M3 was categorized separately. A total number of (10%) was found to be AML (M3) and (23%) was found to be AML (except M3).

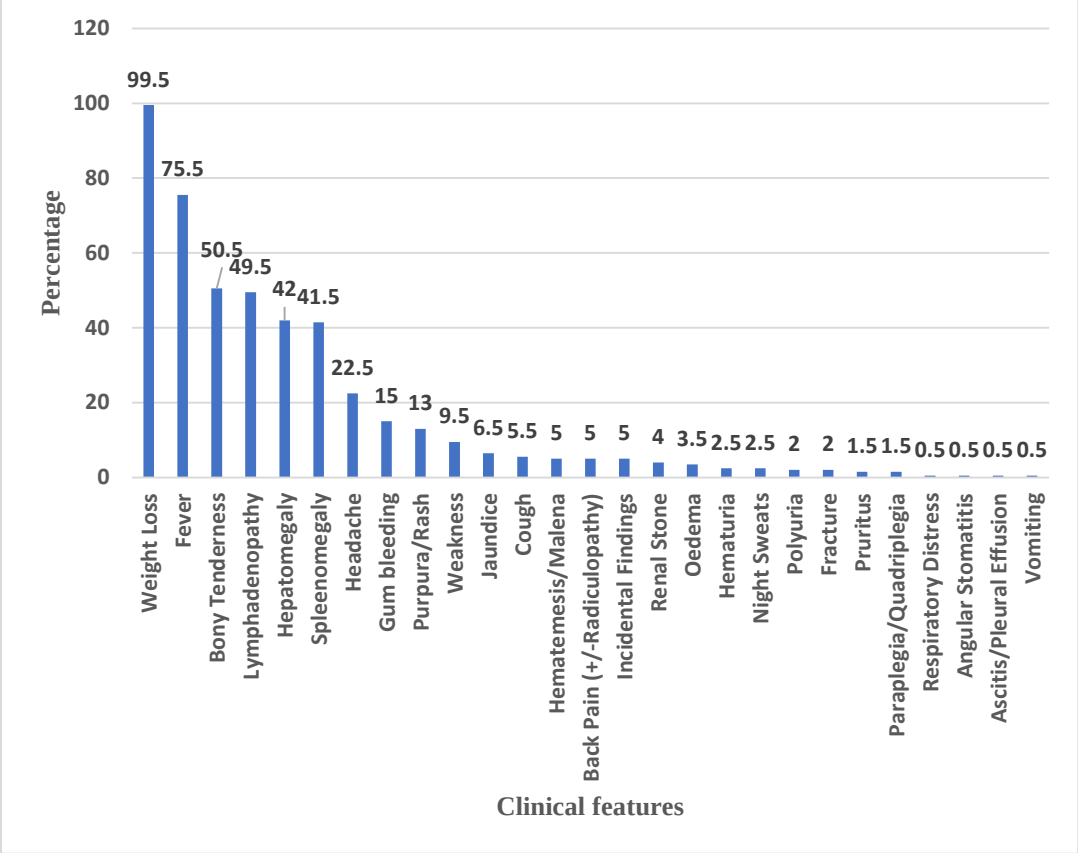

Figure 6: Overall clinical features of HM patients (N=200)

Almost all (99.5%) patients of hematological malignancy had weight loss, fever (75.5%) followed by bony tenderness (50.5%), lymphadenopathy (49.5%), hepatomegaly (42%), splenomegaly (41.5%), headache (22.5%), gum bleeding (15%), purpura/echymosis/rash (13%), weakness (9.5%), jaundice (6.5%), cough (5.5%), back pain(+/- radiculopathy) (5%), hematemesis/melena (5%), incidental findings (5%), renal stone (4%), oedema (3.5%), hematuria (2.5%), night sweats (2.5%), polyuria (2%), fracture (2%), pruritus (1.5%), paraplegia/quadriplegia (1.5%), vomiting/nausea (0.5%), angular stomatitis (0.5%), respiratory distress (0.5%) and ascitis/pleural effusion (0.5%) (Figure 6)

Table 4: Common laboratory Investigations of HM patients (N=200)

Investigations	Leukemia	Lymphoma	Myeloma	Percentage
CBC with ESR	+	+	+	**100**
Peripheral Blood Film (PBF)	+	+	+	**100**
Bone Marrow Study	+	-	+	**79.5**
Flowcytometry	+	-	-	**49.5**
Cytogenetic Study	+	-	+	**30.5**
Lymphnode Biopsy	-	+	-	**20.5**
Immunohistochemistry	-	+	-	**12.5**
Plasma Protein Electrophoresis	-	-	+	**8**

Table 4 summerizes the fact that Leukemia was diagnosed based on CBC with ESR, PBF, Bone marrow study, Flowcytometry and Cytogenetic analysis. Lymphoma was diagnosed on the basis of CBC with ESR, PBF, Lymphnode biopsy and Immunohistochemistry. Diagnosis of Multiple Myeloma was done based on CBC with ESR, PBF, Bone marrow study, Plasma protein electrophoresis and cytogenetic analysis.

Table 5: Overall Complete Blood Count (CBC) finding in patients with HM (N= 200)

Laboratory Findings	Mean ± SD		P value
	Male	Female	
Hemoglobin (gm/dl)	9.45±2.34	9.19±2.03	0.33
ESR (mm in 1st hour)	62.92±30.11	67.74±26.98	0.48
WBC Total Count ($\times 10^9$/L)	60.89±50.78	63.68±53.14	0.70
Platelet Count ($\times 10^9$/L)	233.96±128.65	228.40±109.35	0.56
Blast Cell (%)	37.80± 31.44	40.76± 30.63	0.59

(HM=Hematological Malignancy)

Table 5 revealed that mean Hemoglobin, ESR, WBC and Platelet count of the respondents were 9.45±2.34 gm/dl, 62.92±30.11 mm in 1st hour, 60.89±50.78 $\times 10^9$/Litre, 233.96±128.65 $\times 10^9$/Litre of blood in male patients and 9.18±2.02 gm/dl, 67.74±26.98 mm in 1st hour, 63.68±53.14 $\times 10^9$/Litre and 228.40±109.35 $\times 10^9$/Litre of blood in female patients. There was no significant difference observed between male and female patients. Mean blast cell for male patients was 37.80±31.44 and for female patients was 40.76±30.63 percent respectively. The presence of blast cells in peripheral blood is almost diagnostic in cases of leukemia. It is seen that all the leukemia patients in our study had blast cell in their peripheral blood irrespective of their age and sex.

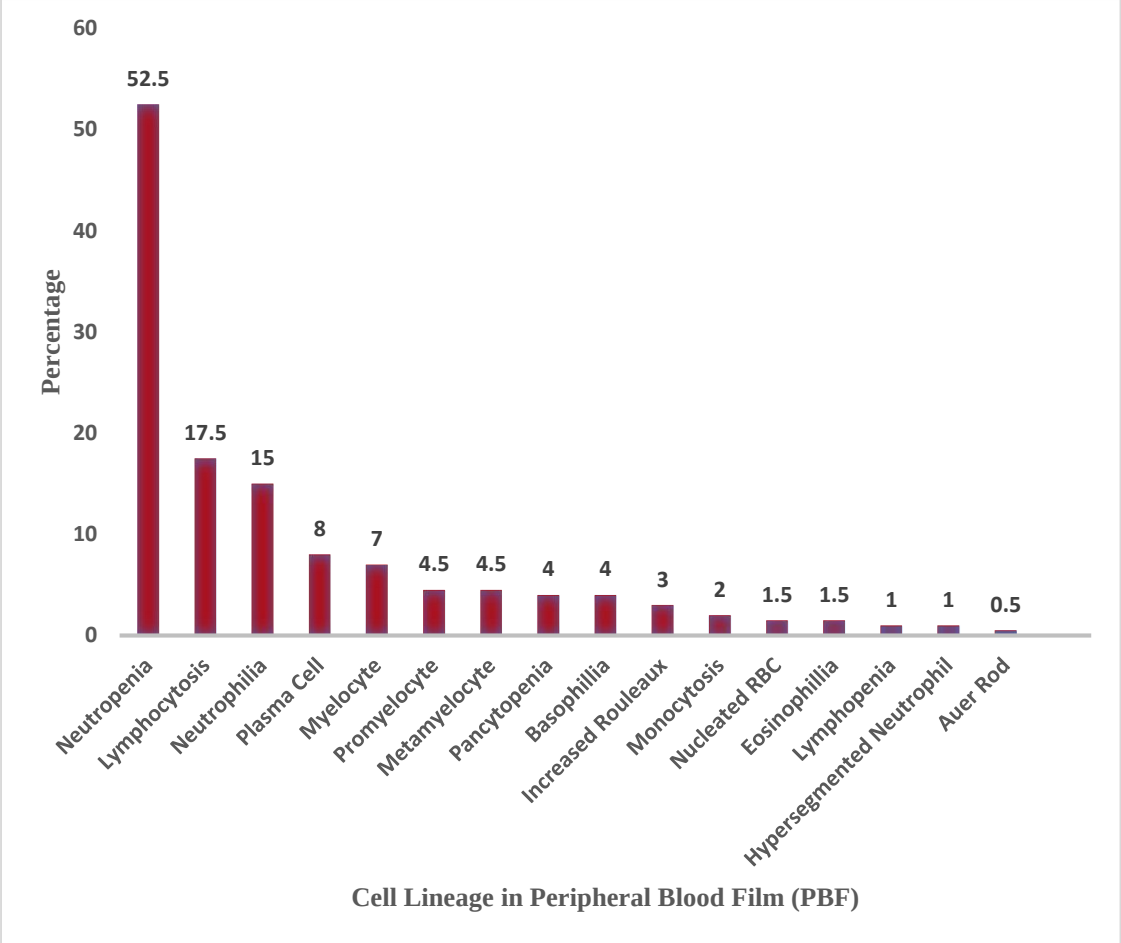

Figure 7: Overall cell lineages found in PBF of HM patients (N=200)

Figure 7 states that more than half of the respondents had Neutropenia (52.5%), followed by Lymphocytosis (17.5%), Neutrophillia (15%), Plasma Cell (8%), Myelocyte (7%), Promyelocyte (4.5%), Metamyelocyte (4.5%), Basophillia (4%), Increased Rouleaux (3%), Monocytosis (2%), Pancytopenia (2%), Eosinophillia (1.5%), Nucleated RBC (1.5%), Lymphopenia (1%), Hyper segmented Neutrophil (1%) and Auer body (0.5%).

Table 6: Overall bone marrow findings of different types of hematological malignancy (HM) patients (n= 159)

Types of malignancy	Percentage
Acute Myeloid Leukemia (Total) • Acute Myeloid Leukemia (AML except M3)) • Acute Myeloid Leukemia (AML M3)	41.5 28.9 12.6
Acute Lymphoblastic Leukemia (ALL)	32.7
Chronic Lymphocytic Leukemia (CLL)	2.5
Chronic Myeloid Leukemia (CML)	8.8
Myelodysplastic Syndrome (MDS)	1.9
Mixed Leukemia	2.5
Multiple Myeloma (MM)	10.1

It was described in table 6 that, in regard of positive bone marrow findings majority of the respondents had AML (Total) (41.5%) which comprises of AML other than M3 (28.9%) and AML M3 (12.6%). The next majority of respondents had ALL (32.7%). The rest are as follows- MM (10.1%), CML (8.8%), CLL (2.5%), Mixed Leukemia (2.5%) and MDS (1.9%).

Table 7: Overall flowcytometry findings of different types of hematological malignancy patients (N= 99)

Flowcytometry findings	Percentage
T lymphocyte Markers	40.4
B lymphocyte Markers	30.3
Myeloid Markers	29.3

According to flow cytometry findings majority of the hematological malignancy patients had T lymphocyte markers (40.4%) followed by B lymphocyte markers (30%) and myeloid markers (29.3%).

Table 8: Overall plasma protein electrophoresis findings of different types of hematological malignancy patients (N= 16)

Plasma Protein Electrophoresis findings	Percentage
Monoclonal Gammopathy	3.5
Monoclonal Gammopathy with Beta 2 Microglobulinemia	2
Light Chain Disease	1
Light Chain Disease with Beta 2 Microglobulinemia	1.5

It is illustrated in table 8 that, in regard of positive plasma electrophoresis findings majority of the respondents had Monoclonal Gammopathy (3.5%) followed by mixed picture of Monoclonal Gammopathy with Beta 2 Microglobulinemia (2%), Light Chain Disease with Beta 2 Microglobulinemia (1.5%) and Light Chain Disease (1%). It is only done in case of multiple myeloma.

Table 9: Overall lymph node biopsy findings of different types of Hematological Malignancy patients (N= 41)

Lymph Node Biopsy findings	Percentage
Hodgkin Disease (HD)	8.5
Non Hodgkin Lymphoma (NHL)	12

It is described in table 9 that, in regard of positive lymph node biopsy findings, which is only done in cases of lymphoma, majority of patients had NHL (12%) followed by HD (8.5%).

Table 10: Overall cytogenetic study findings of different types of Hematological Malignancy patients (N= 61)

Cytogenetic Study Findings	Percentage
Philadelphia Chromosome	15.5
CD 33 Positive	1.5
FLT3 Mutation	1.5
PML RARA Positive	10
CD 20 Positive	1.5
BCL 2 Positive	0.5

(CD=Cluster of Differentiation, FLT3=Fms like Tyrosine kinase receptor 3, PML RARA=Promyelocytic Leukemia Retinoic Acid Receptor Alpha, BCL=B cell lymphoma 2)

It is described in table 10 that, in regard of positive cytogenetic study findings majority of the respondents were positive for Philadelphia Chromosome (15.5%) followed by PML RARA (10%). CD33 and CD 20 positive cases were (1.5%) each and FLT3 Mutation was detected in respondents (1.5%). Only (0.5%) patients were positive for BCL 2.

Table 11: Overall Immunohistochemistry findings of different types of Hematological Malignancy patients (N= 25)

Immunohistochemistry findings	Percentage
CD 30 Positive	6
CD 20 Positive	6.5

(CD=Cluster of Differentiation)

It is described in table 11 that, in regard of positive immunohistochemistry findings majority of patients were positive for CD 20 (6.5%) and (6%) were for CD 30. The rest of patients did not do the test.

Table 12: Overall percentage of treatment modalities taken by Hematological Malignancy patients (N=200)

Treatment Modalities	Given (%)	Not given (%)
Chemotherapy	87	13
Radiotherapy	0	100
Immuno/Target Therapy	46.3	53.7
Bone Marrow Transplantation	0	100

Table 12 states that more than three fourth 87% patients had taken Chemotherapy as a mode of their treatment. 46.3% got Immunotherapy/Target therapy. Radiotherapy and Bone Marrow Transplantation is unavailable in Chittagong. As Radiotherapy machine in CMCH is currently under maintenance and no other private institution provides this facility in Chittagong, it was impossible for the respondents to take radiotherapy. Once the machine in CMCH becomes ready for service, it is literally a daunting task to get the serial as this is the only Radiotherapy facility for whole of Chittagong division.

Part B

Comparative clinical scenario, lab findings and treatment modalities of different types of hematological malignancy patients

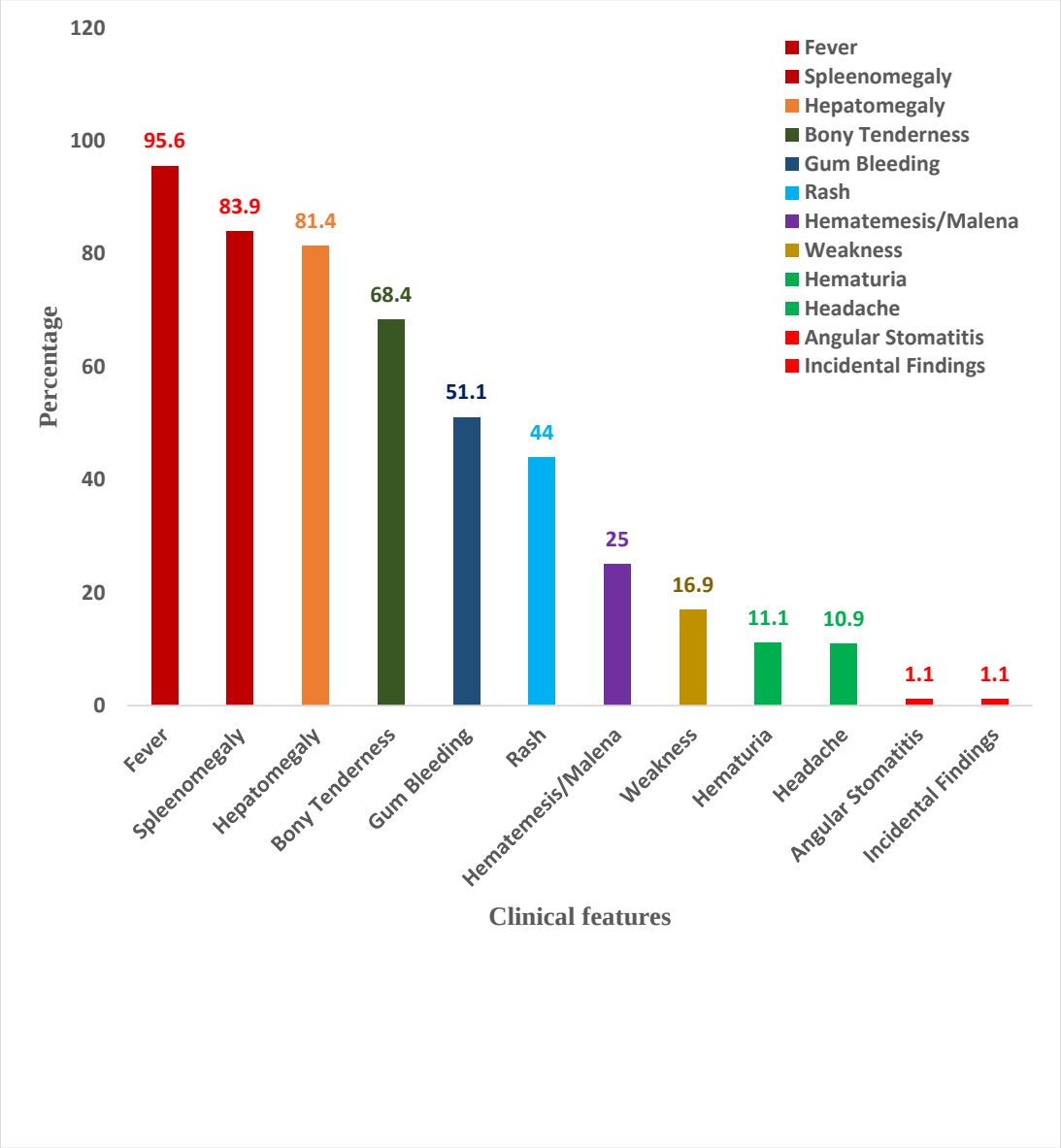

Figure 8: Proportion of different clinical features of Acute Myeloblastic Leukemia (AML) (N=66)

It was described in figure 8 that, among the patients suffering from AML, the most common clinical feature were fever (95.6%), splenomegaly (83.9%), hepatomegaly (81.4.8%) and followed by bony tenderness (68.4.7%), gum bleeding (51.1%), rash (44%), Hematemesis/Malena (25%), Weakness (16.9%), hematuria (11.1%), headache (10.9%) and the least feature was angular stomatitis and incidental findings (1.1% each).

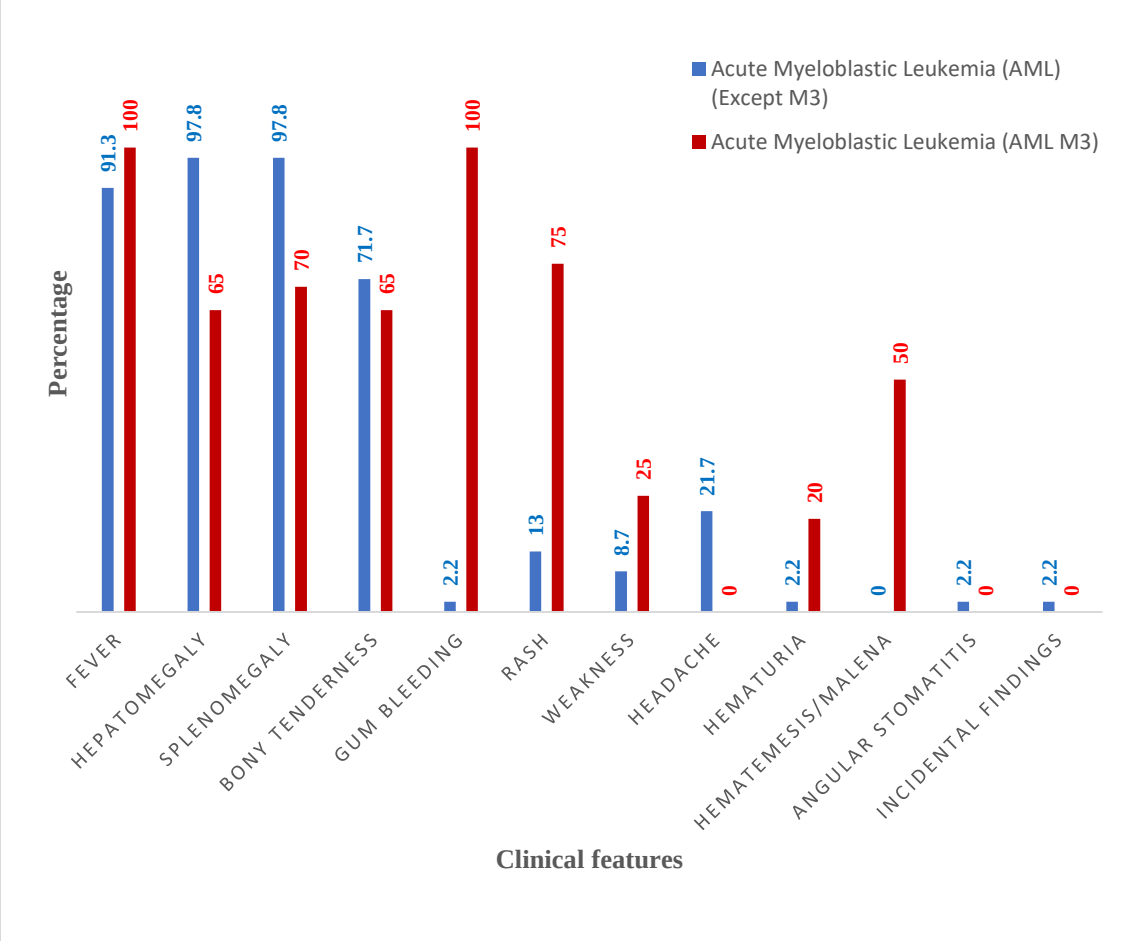

Figure 9: Comparison between clinical features of AML except M3 and AML M3 patients (N=66)

Figure 9 demonstrates a comparison between patients suffering from AML except M3 and AML M3. among the patients suffering from AML, the most common clinical feature was hepatomegaly (97.8%) and splenomegaly (97.8%) followed by fever (91.3%), Bony tenderness (71.7%), Headache (21.7%), rash (13%), Weakness (8.7%), cough (4.3%) and the least feature was Gum bleeding (2.2%), hematuria (2.2%) and incidental findings (2.2%). Whereas the HM patients suffering from AML M3, showed that the most common (100%) clinical feature were fever and Gum bleeding followed by rash (75%), splenomegaly (70%), hepatomegaly (65%), Bony tenderness (65%), Hematemesis (50%), Weakness (25%), and the least feature was hematuria (20%).

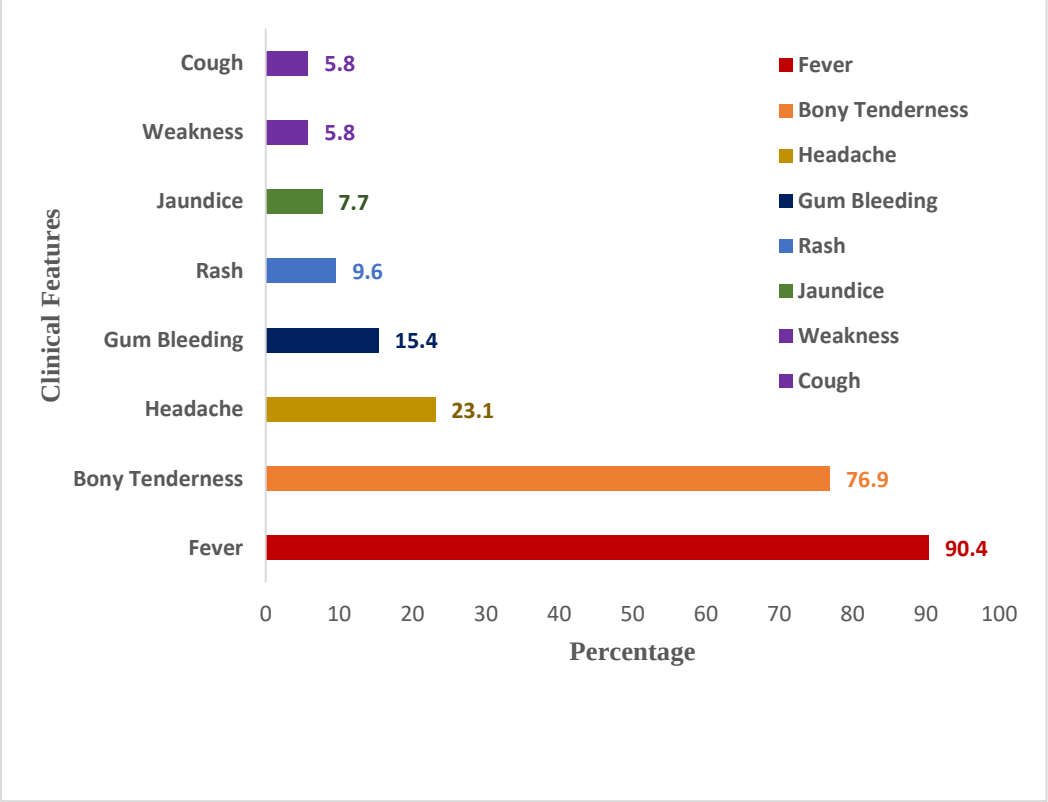

Figure 10: Percentage of different clinical features of Acute Lymphoblastic Leukemia (ALL) (N=52)

Figure 9 describes that among the patients suffering from ALL, most of them had fever (90.4%) followed by bony tenderness (76.9%), headache (23.1%), gum bleeding (15.4%), rash (9.6%), weakness (5.8%) and cough (5.8%).

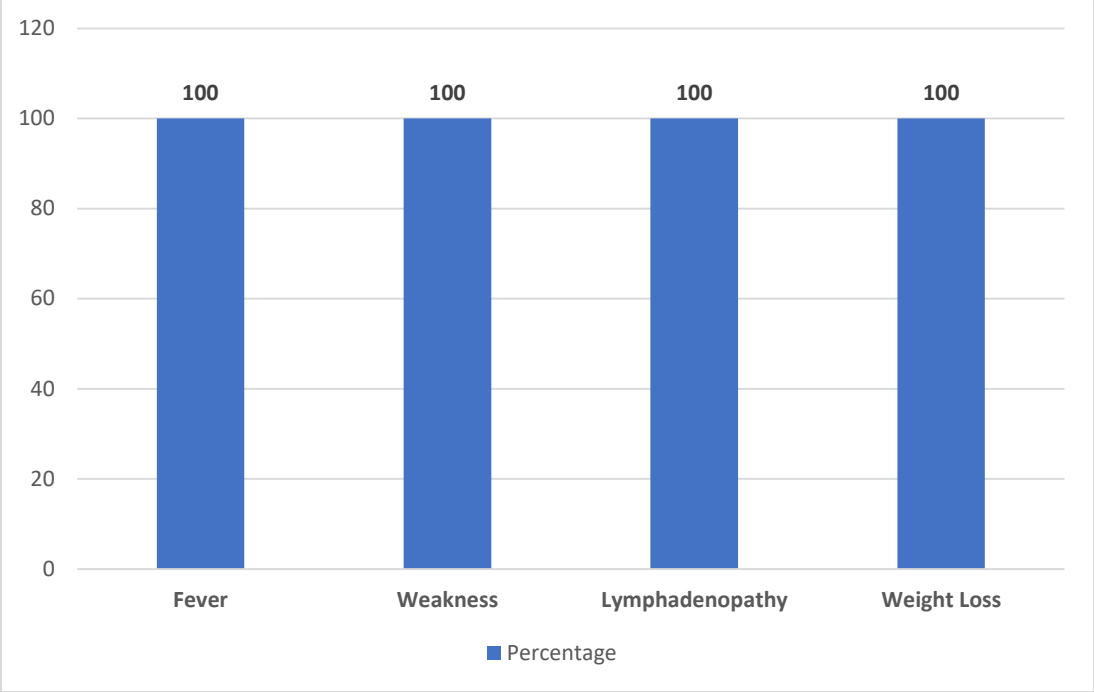

Figure 11: Proportion of different clinical features of Chronic Lymphocytic Leukemia (CLL) (N=4)

All patients (100%) demonstrates the four features such as fever, weakness, lymphadenopathy and weight loss as depicted in figure 11.

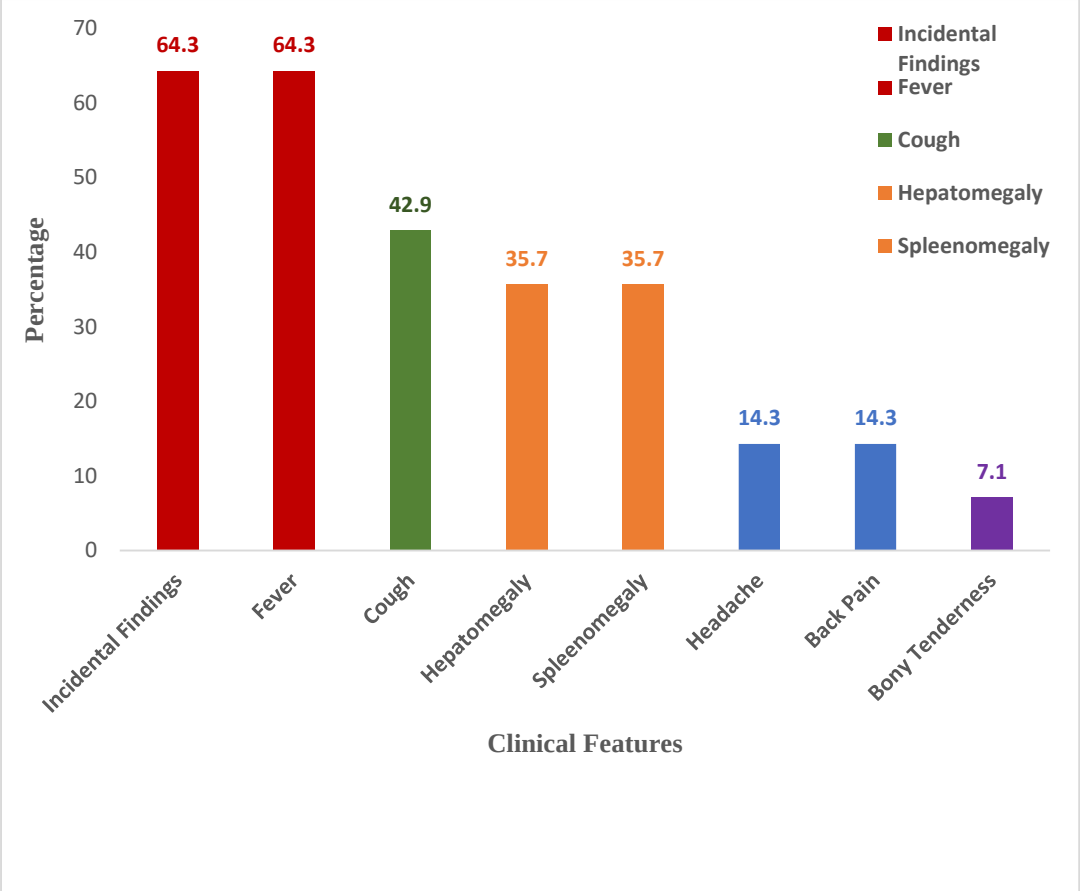

Figure 12: Percentage of different clinical features of Chronic Myeloid Leukemia (CML) (N=14)

In figure 12, it was shown that in cases of CML patients the most common (64.3%) clinical feature were fever and incidental findings followed by cough (42.9%), splenomegaly (35.7%), hepatomegaly (35.7%), headache (14.3%), back pain (14.3%) and the least feature was Bony tenderness (7.1%).

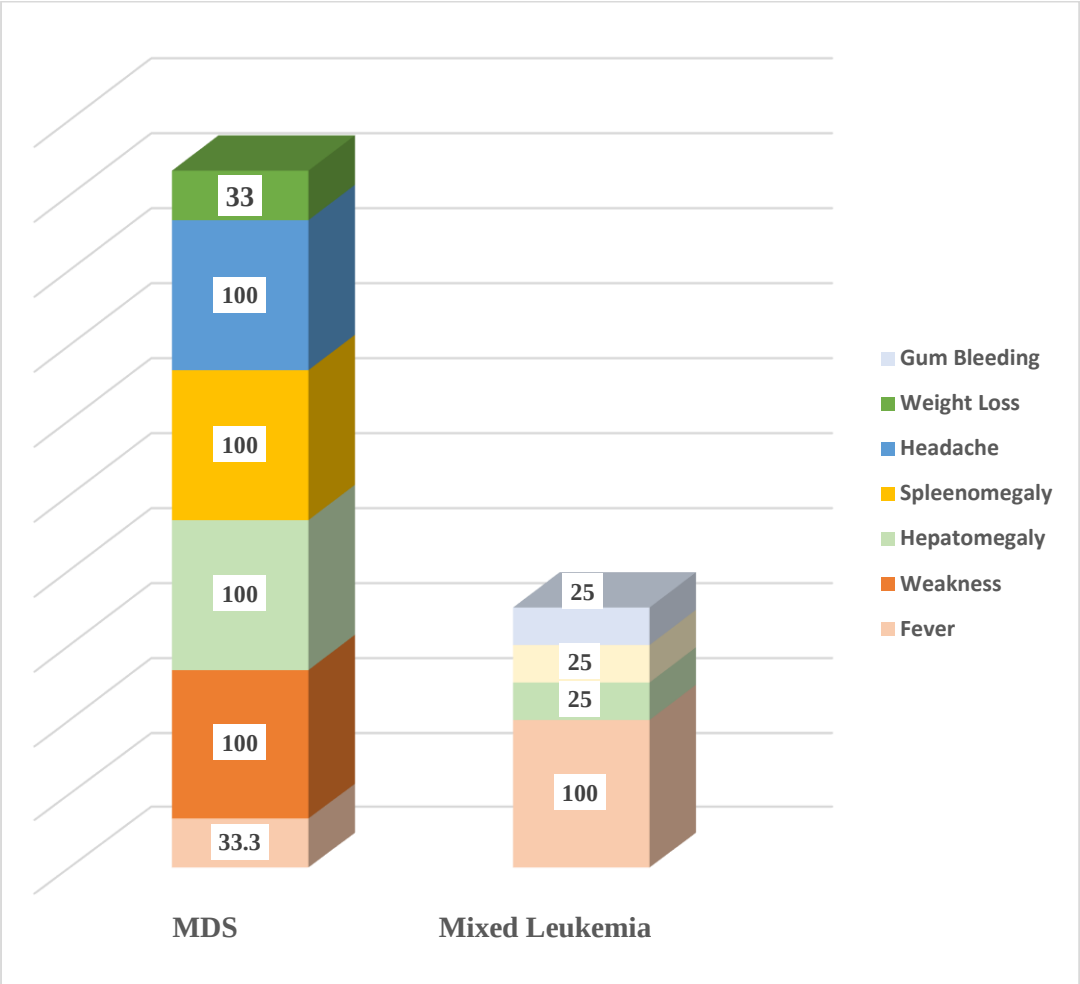

Figure 13: Comparison between different clinical features of Myelodysplastic Syndrome (MDS) (N=3) and Mixed Leukemia (N=4)

It is described in figure 14 that, among the respondents suffering from MDS, the most common (100%) clinical feature was Weakness, Headache, Hepatomegaly and Splenomegaly. The least feature (33.3%) was fever and weight loss. Among the respondents suffering from mixed leukemia, the most common (100%) clinical feature was Fever followed by Hepatomegaly (75%), Splenomegaly (75%), Lymphadenopathy (50%) and the least feature (25%) was Headache and Gum Bleeding.

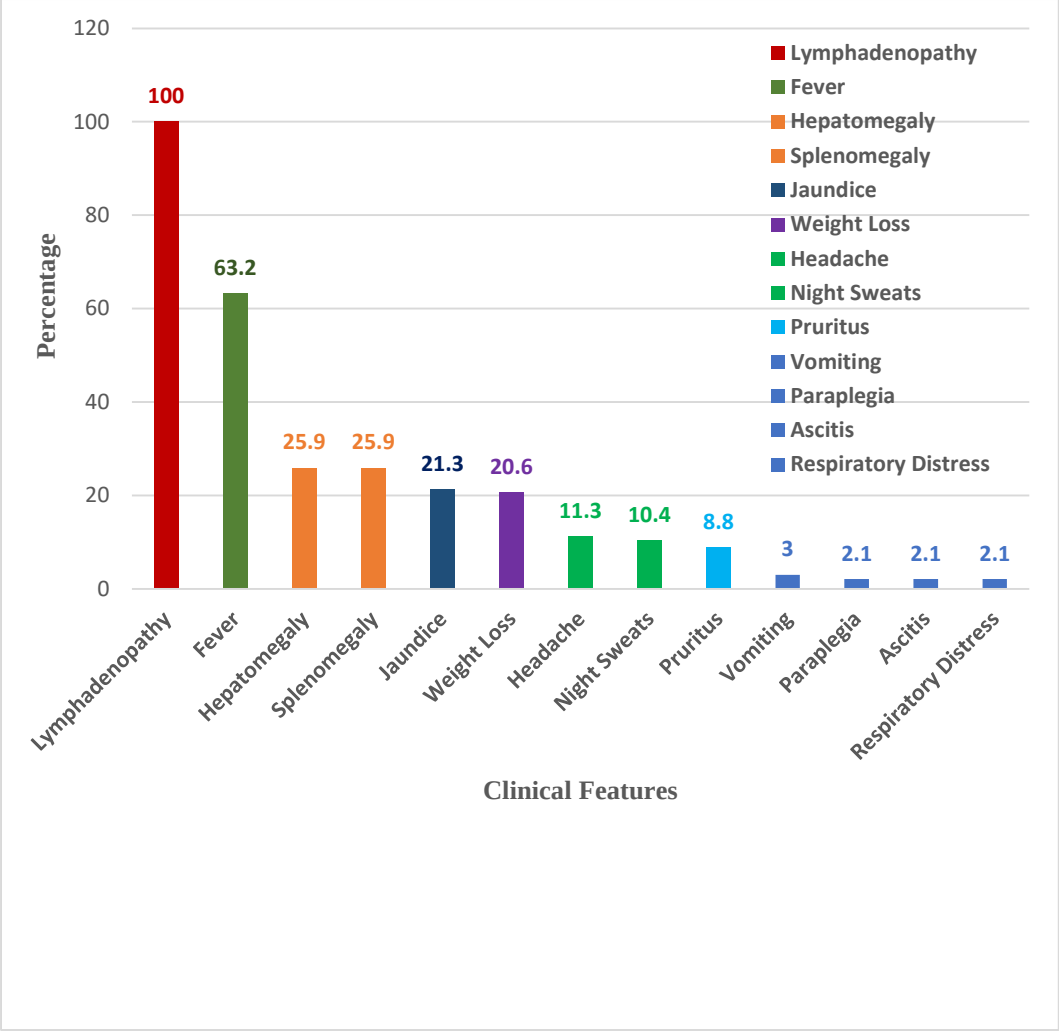

Figure 14: Proportion of different clinical features of Lymphoma (N=41)

In this figure 14, it was shown that lymphadenopathy is the most common feature (100%) followed by fever (63.2%), hepatomegaly and splenomegaly each with 25.9%, jaundice (21.3%), weight Loss (20.6%), headache (11.3%), night sweats (10.4%) Pruritus (8.8%), vomiting (3%) and each and paraplegia, respiratory distress and ascites each with 2.1%.

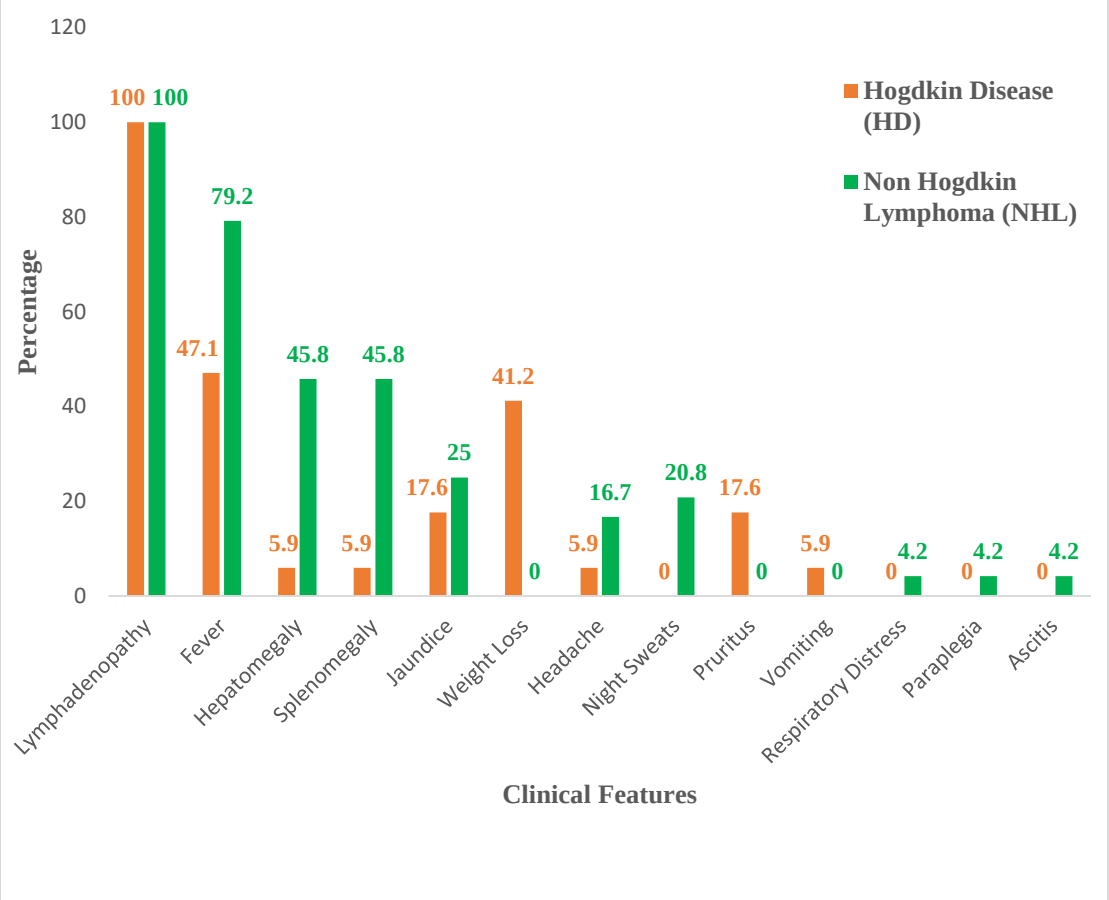

Figure 15: Comparison of clinical features between HD and NHL (N=41)

In figure 15 that, it is shown that in case of HD lymphadenopathy is the most common symptom (100%) followed by fever (47.1%), weight loss (41.2%). pruritus and jaundice were (17.6%) each and vomiting, headache, hepatomegaly and splenomegaly were (5.9%) each. Among the patients suffering from NHL, the most common (100%) clinical feature was lymphadenopathy followed by fever (79.2%), weight Loss (45.8%), hepatomegaly (45.8%), splenomegaly (45.8%), night Sweats (20.8%), headache (16.7%), jaundice (25%) and the least feature (4.2%) were ascites, headache, paraplegia and respiratory distress.

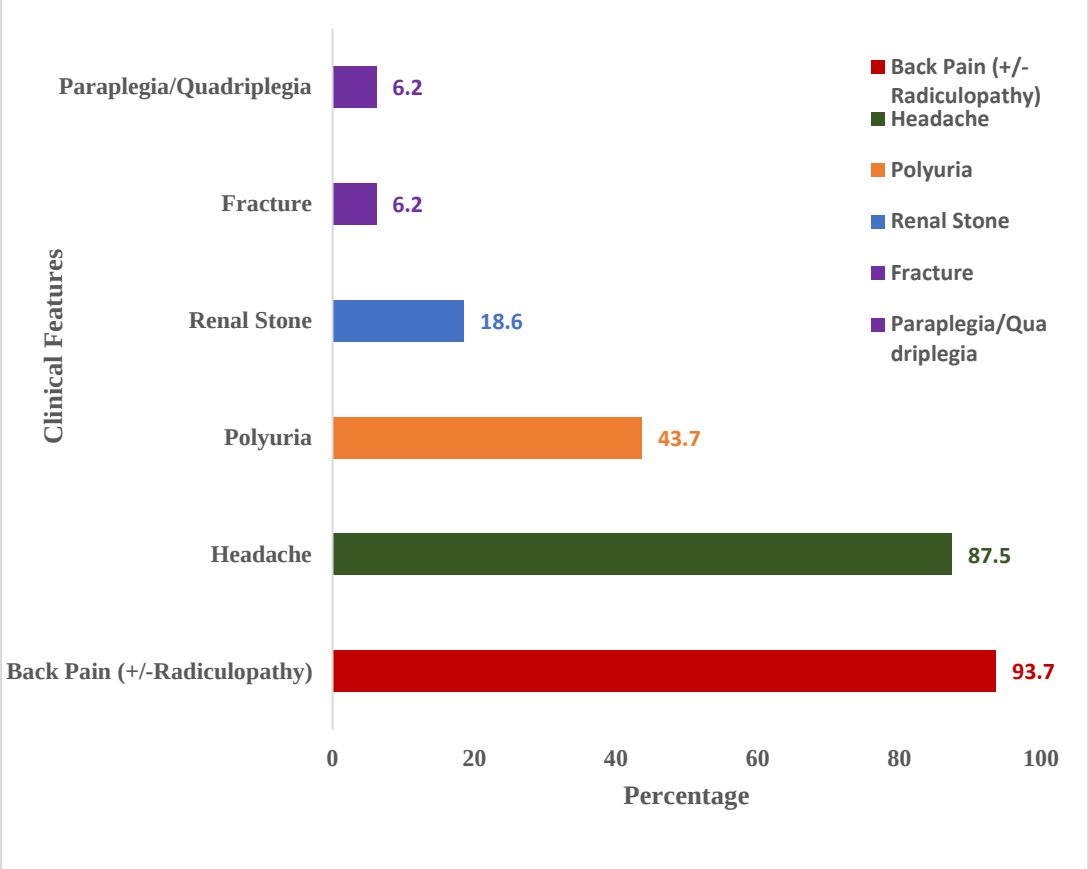

Figure 16: Percentage of different clinical features of Multiple Myeloma (MM) (N=14)

In figure 16, it was shown that most patients of MM presents with Back pain (+/- Radiculopathy) (93.7%) followed by Headache (87.5%), Polyuria (43.7%), Renal Stone (18.6%). Fracture and Paraplegia/Quadriplegia is the least of it (6.2%) each.

Table 13: Percentage of different cell lineages in PBF findings among different types of hematological malignancy patients-A comparison

Variables	AML	AML M3	ALL	CML	CLL	MDS	Mixed Leukemia	HD	NHL	MM
Neutropenia	100	100	9.6	-	-	-	-	-	-	-
Lymphocytosis	-	-	61.5	-	100	-	50	100	100	-
Neutrophilia	-	-	19.2	100	-	-	50	-	-	-
Plasma Cell	-	-	-	-	-	-	-	-	-	100
Myelocyte	-	-	-	12	-	-	-	-	-	-
Pro myelocyte	-	-	-	64.3	-	-	-	-	-	-
Meta myelocyte	-	-	-	64.3	-	-	-	-	-	-
Pancytopenia	-	-	7.7	-	-	-	-	-	-	-
Basophillia	-	-	-	57.1	-	-	-	-	-	-
Increased Rouleaux Formation	-	-	-	-	-	-	-	-	-	46
Monocytosis	-	-	-	28.6	-	-	-	-	-	-
Neucleated RBC	-	-	-	2	-	100	-	-	-	-
Eosinophillia	-	-	-	21.4	-	-	-	-	-	-
Lymphopenia	-	-	3.8	-	-	-	-	-	-	-
Hyper segmented Neutrophil	-	-	-	-	-	66.7	-	-	-	-
Auer Rod	-	-	-	-	-	33.3	-	-	-	-

(PBF=Peripheral blood film, AML=Acute Myeloid Leukemia, ALL=Acute Lymphoblastic Leukemia, CML=Chronic Myeloid Leukemia, CLL=Chronic Lymphocytic Leukemia, MDS=Myelodysplastic Syndrome, HD=Hogdkin Disease, NHL=Non Hogdkin Lymphoma, MM=Multiple Myeloma)

Table 13 shows that all respondents (100%) suffering from AML (both AML M3 and AML other than M3) had Neutropenia in their PBF findings. It also illustrates that among the respondents suffering from ALL, more than half (61.5%) of them had lymphocytosis followed by Neutrophilia (19.2%), Neutropenia (9.6%), Pancytopenia (7.7%) and Lymphopenia (3.8%). It shows that all respondents (100%) suffering from CLL, HD and NHL had lymphocytosis in their CBC findings whereas all of MM patients had Plasma cell in PBF (100%) and Increased Rouleaux Formation (46%). In case of Mixed Leukemia we found that Neutrophillia and Lymphocytosis were (50%) each. Table also illustrates that among the respondents suffering from MDS, all of them (100%) had Nucleated RBC followed by Hyper-segmented Neutrophil (66.7%) and Auer Rod (33.3%). illustrates that among the respondents suffering from CML, all of them (100%) had Neutrophilia followed by Promyelocyte (64.3%), Metamyelocyte (64.3%), Basophillia (57.1%), Monocytosis (28.6%), and Eosinophillia (21.4%).

Table 14: Blast cell picture of different types of hematological malignancy patients (N=138)

Category	Minimum (%)	Maximum (%)	Mean±SD
AML (Except M3)	34	91	63.78±15.05
AML M3	35	88	61.35±16.52
ALL	30	91	60.29±15.47
CML	15	30	22.29±4.75
CLL	0	0	0
MDS	0	0	0
Mixed Leukemia	46	61	22.29±4.75
HD	0	0	0
NHL	0	0	0
MM	0	0	0

(AML=Acute Myeloid Leukemia, ALL=Acute Lymphoblastic Leukemia, CML=Chronic Myeloid Leukemia, CLL=Chronic Lymphocytic Leukemia, MDS=Myelodysplastic Syndrome, HD=Hogdkin Disease, NHL=Non Hogdkin Lymphoma, MM=Multiple Myeloma)

Table 14 shows that blast cells are almost always present in Leukemia cases. The most blast cells were found in AML except M3 (63.78±15.05) where minimum percentage was (34%) and maximum was (91%) followed by AML M3 (61.35±16.52), minimum (35%) and maximum (88%) and ALL (60.29±15.47) with minimum (30%) and maximum (91%). In MDS (22.29±4.75), we found that minimum and maximum percentage of blast were (46%) and (61%). The rest had no blast cell in peripheral blood.

Table 15: Comparison of flowcytometry findings among different types of hematological malignancy patients (N=149)

Disease	Flow cytometry findings	Percentage
AML	T lymphocyte markers	4.3
	Myeloid markers	60.9
ALL	T lymphocyte markers	90
CLL	T lymphocyte markers	100
MDS	Myeloid markers	33.3
NHL	B lymphocyte markers	54.2

(AML=Acute Myeloid Leukemia, ALL=Acute Lymphoblastic Leukemia, CLL=Chronic Lymphocytic Leukemia, MDS=Myelodysplastic Syndrome, NHL=Non Hogdkin Lymphoma)

Table 15 states the percentage of respondents according to finding of flowcytometry study of AML where more than half (60.9%) had myeloid lineage and only (4.3%) had T cell lineage. Rest of the patients could not do the test due to high cost. In case of ALL T cell lineage is (90%). In case of MDS only (33.3%) had Myeloid Lineage rest of them did not do the test due to its high cost. It also states that in CLL we find the predominant lineage is T cell lineage (100%). Other varieties of HM patients did not do Flowcytometry as in AML M3, HD, NHL, MM and CML it is not relevant. Among the respondents in regards of NHL more than half (54.2%) have done this test and found B Cell Lineage and rest did not go through flowcytometry due to its high cost.

Table 16: Comparison of findings of cytogenetic study among different types of hematological malignancy patients (N=136)

Disease	Cytogenic finding	Percentage
AML except M3	Philadelphia Chromosome	**58.8**
	CD33 positive	**17.6**
	FLT 3 Mutation	**23.5**
AML M3	PML RARA	**100**
ALL	Philadelphia Chromosome	**13.5**
CML	Philadelphia Chromosome	**100**
CLL	CD 20 Positive	**75**
	BCL 2 Positive	**25**

(AML=Acute Myeloid Leukemia, ALL=Acute Lymphoblastic Leukemia, CML=Chronic Myeloid Leukemia, CLL=Chronic Lymphocytic Leukemia, CD=Cluster of Differentiation, FLT3=Fms like Tyrosine kinase receptor 3, PML RARA=Promyelocytic Leukemia Retinoic Acid Receptor Alpha, BCL=B cell lymphoma 2)

Table 16 says the frequency distribution of respondents according to finding of cytogenetic study of AML Other than M3 where most common finding was Philadelphia Chromosome (58.8%) followed by FLT 3 Mutation (23.5%) and CD33 positive (17.6%). In case of AML M3, we find PML RARA in all patients (100%). In regard of Cytogenetic study among the respondents suffering from ALL less than one fourth (13.5%) respondents had done this study and the finding was Philadelphia Chromosome. Rest of the respondents did not go through this study due to its high cost. In case of CLL we found that in Cytogenetic study it was CD20 Positive (75%) and BCL 2 positive (25%). In case of CML we found all the patients to be positive for Philadelphia Chromosome (100%).

Table 17: Comparison of findings of immunohistochemistry among different types of hematological malignancy patients (N=41)

Disease	Immunohistochemistry	Percentage
Hodgkin Disease	CD30 positive	70.6
Non Hodgkin Lymphoma	CD 20 positive	54.2

Among the patients in regard of HD more than half (70.6%) have done this test and found CD30 positive. Only (29.4%) did not go through Immunohistochemistry due to its high cost (Table 17). It also shows that among the patients of NHL more than half (54.2%) have done this test and found CD 20 Positive. The rest did not go through Immunohistochemistry due to its high cost. It is not done in case of Leukemia and Myeloma.

Table 18: Percentage of different chemotherapeutic drugs used by hematological malignancy patients suffering from different types of hematological malignancy (N= 174)

Chemotherapeutic drugs	AML Other than M3	AML M3	ALL	MDS	Mixed Leukemia	HD	NHL	MM	% Overall
Doxorubicin	-	-	-	-	**100**	-	**100**	-	**3.5**
Bleomycin	-	-	**1.9**	-	-	**100**	-	-	**8.5**
Vinblastine	-	-	-	-	-	**100**	-	-	**8**
Dacarbazine	-	-	-	-	-	**100**	-	-	**8**
Cyclophosphamide	-	-	-	-	-	-	**100**	**25**	**12**
Vincristine	-	-	**90.4**	-	-	-	**100**	-	**35.5**
Prednisolone	-	-	**100**	-	**100**	-	**100**	-	**40**
Daunorubicin	**100**	-	**100**	**100**	**100**	**100**	-	-	**51**
Cytarabine	**100**	-	**9.6**	-	**100**	-	-	-	**27.5**
Arsenic Trioxide (ATO)	-	**100**	-	-	-	-	-	-	**10**
Vesanoid (ATRA)	-	**100**	-	-	-	-	-	-	**10**
Bordezomib	-	-	-	**4**	-	-	-	**100**	**8**
Linamide	-	-	-	-	-	-	-	**100**	**8**
Dexamethasone	-	-	-	**100**	-	-	-	**100**	**9.4**
L Aspaginase	-	-	**88.5**	-	-	-	-	-	**23**
6 Mercaptopurine	-	-	**9.6**	-	-	-	-	-	**2.5**
Methotrexate	-	-	**98.1**	-	**25**	-	-	-	**26**

(AML=Acute Myeloid Leukemia, ALL=Acute Lymphoblastic Leukemia, CML=Chronic Myeloid Leukemia, CLL=Chronic Lymphocytic Leukemia, MDS=Myelodysplastic Syndrome, HD=Hogdkin Disease, NHL=Non Hogdkin Lymphoma, MM=Multiple Myeloma)

It is illustrated in table 18 that in the treatment of AML (other than AML M3), most commonly used (100%) drugs were Daunorubicin and Cytarabine and in the treatment of AML M3, all patients received (100%) Arsenic Trioxide and ATRA. In the treatment of ALL where all of them had Prednisolone (100%) and Daunorubicin (100%) followed by Methotrexate (98.1%), Vincristine (90.4%), L Aspaginase (88.5%), Cytarabine (9.6%), 6-Mercaptopurine (9.6%) and Bleomycin (1.9%). In the treatment of MDS, most commonly used (100.0%) drugs were Daunorubicin and Dexamethasone. The table also states that in the treatment of Mixed Leukemia, most commonly used (100%) drugs were Doxorubicin, Prednisolone, Daunorubicin, Cytarabine and only (25%) Methotrexate. We can see that in the treatment of HD, all (100%) drugs were Daunorubicin, Bleomycin Vinblastine and Dacarbazine. (ABVD Protocol) and in the treatment of NHL, most commonly used (100%) drugs were Doxorubicin, Cyclophosphamide, Vincristine and Prednisolone (As part of R-CHOP Protocol). According to the table above all patients received Bordezomib (100%), Linamide (100%), Dexamethasone (100%) and only (25%) got Cyclophosphamide in the treatment of MM. CLL and CML patients did not receive chemotherapy rather received target therapies which are discussed later.

In Overall perspective, the most commonly used chemotherapeutic drug among the respondents suffering from hematological malignancy was Daunorubicin (51%) followed by Prednisolone (40%), Vincristine (35.5%), Cytarabine (27.5%), Methotrexate (26%), L Asparginase (23%), Cyclophosphamide (12%), Arsenic Trioxide (ATO) (10%), Vesanoid (ATRA) (10%), Dexamethasone (9.4%), Bleomycin (8.5%), Vinblastine (8%), Dacarbazine (8%), Bordezomib (8%), Linamide (8%), Doxorubicin (3.5%) and 6 Mercaptopurine (2.5%).

Table 19: Supply of chemotherapeutic drugs used for treatment of hematological malignancy patients (N=174)

Drug name	Government Supply	Private Supply/Patients own fund
	Percentage	Percentage
Doxorubicin(n=7)	57.5	42.5
Bleomycin (n=17)	5.9	94.1
Vinblastine (n=16)	18.7	81.3
Dacarbazine (n=16)	18.7	81.3
Cyclophosphamide (n=24)	33.3	66.7
Vincristine (n=71)	38	62
Prednisolone (n=80)	31.3	68.7
Daunorubicin (n=102)	51	49
Cytarabine (n=55)	9.1	90.9
Arsenic Trioxide (ATO) (n=20)	0	100
Vesanoid (ATRA) (n=20)	0	100
Bortezomib (n=16)	0	100
Linamide (n=16)	0	100
Dexamethasone (n=19)	57.9	42.1
L asparginase (n=46)	0	100
Methotrexate (n=52)	0	100

Table 19 says that among the respondents that used Doxorubicin, (57.5%) got that from Govt. supply and (42.5%) got from private supply. It also states that among the respondents that used Bleomycin, majority of them (94.1%) got that from Govt. supply and only (5.9%) got from private supply. Among the respondents that used Vinblastine

and Dacarbazine, majority of them (81.3%) got that from private supply and (18.7%) got from Govt. supply. Among the respondents that used Cyclophosphamide, majority of them (66.7%) got that from private supply and (33.3%) got from Govt. supply. We can see that among the respondents that used Vincristine, majority of them (62%) got that from private supply and (38%) got from Govt. supply. It also describes that among the respondents that used Prednisolone, majority of them (68.7%) got that from private supply and (31.3%) got from Govt. supply.

Only Daunorubicin picture states otherwise. Majority of them (51%) got from Govt. supply and (49%) got from private supply. Among the respondents that used Arsenic Trioxide and Vesanoid (ATRA), all of them (100%) got that from private supply. Among the respondents that used Bordezomib and Linamide, all of them (100%) got that from private supply.

On the contrary, respondents that used Dexamethasone, majority of them (57.9%) got that from Govt. supply and (42.1%) got from private supply. This table also states that among the respondents that used L Aspaginase, 6 Mercaptopurine all of them (100%) got that from private supply. Finally we can see that all of the respondents got their supply of Methotrexate from private supply (100%).

Table 20: Overall percentage of the use of different immune therapy/ target therapy by the hematological malignancy patients (N= 87)

Immunotherapy/ Target Therapy	AML	ALL	CML	CLL	MDS	HD	NHL	Overall %
Rituximab	-	-	-	**50**	-	-	**54.2**	**21.5**
Imatinib	**83.3**	**13.5**	**100**	-	-	-	-	**15.5**
Azacytidine	-	-	-	-	**60**	-	-	**1.50**
Venetoclox	-	-	-	**50**	-	-	-	**1.0**
Brentuximab Vedotin	-	-	-	-	-	**29**	-	**3.0**
Midostaurine	**16.7**	-	-	-	-	-	-	**1.0**

(AML=Acute Myeloid Leukemia, ALL=Acute Lymphoblastic Leukemia, CML=Chronic Myeloid Leukemia, CLL=Chronic Lymphocytic Leukemia, MDS=Myelodysplastic Syndrome, HD=Hogdkin Disease, NHL=Non Hogdkin Lymphoma)

Table 20 states that in the treatment of AML where more than three fourth (83.3%) respondents have taken Imatinib and only (16.7%) have taken Midostaurin. Midostaurin is unavailable in Bangladesh. Patient who went for further follow up abroad has received this drug. In treatment of ALL, only (13.5%) patients received Imatinib owing to their Philadelphia chromosome positive reports in cytogenetic study. In the treatment of CML, all patients (100%) received Imatinib as target therapy. The table also illustrates that half of the respondents (50%) suffering from CLL have taken Venetoclox as immunotherapy. Rest (50%) got Rituximab. In case of MDS and HD, most of the patients received Azacytidine (60%) and Brentuximab Vedotin (29%) respectively. These two drugs are also unavailable in Bangladesh and patients who went abroad got that drug. In case of NHL, (54.2%) received Rituximab as part of treatment. It also shows that highest respondents took Rituximab (21.5%) followed by Imatinib (15.5%), Brentuximab Vedotin (3%), Azacytidine (1.5%), Midostaurine (1%) and Venetoclox (1%). All the drugs were obtained from patient's own expense and none of the drugs were supplied by Government fund.

Chapter 5

Discussion

This retrospective observational study was carried out to determine the patterns of common Hematological Malignancies in Chattogram. A total of 200 patients admitted in admitted in hematology department of Chattogram Medical College suffering from Hematological Malignancies were included in this study.

In present study the mean age of the respondents was 36.03 ±18.07 years and minimum age was 2 years and maximum age was 80 years. No significant age variations were seen in respect of HM. Majority of our respondents were male (55.9%) whereas females were about (43.1%). (60.5%) of respondents were from rural areas compared to (39.5%) of urban dwellers. Most of the respondents have completed only primary school (32%) followed by graduation (25%), Secondary education completion (20%), Higher Secondary education completion (10%). Completion of Post-graduation and above was (0.5%) and one-fourth of the respondents was illiterate (25%). Most of the respondents (20%) were students followed by housewife (18%), service holder (16%), farmer (9.5%), garments worker (8%), teacher (8%), businessman (6.5%), fisherman (5.5%), Carpenter (3%), Driver (2%), Dry Fish Factory Worker (1%), plumber (0.5%), retired person (0.5%) and (1%) were unemployed. A similar study revealed that the combined median age at diagnosis for all hematological malignancies was 42 years. Age-group specific distribution showed that three types of HM including AML (64.6%), CML (66.2%), ALL (66.4%), and HL (62.7%) were predominantly observed in the young adults aged 20-49 years. Interestingly, four other HM were mostly occurred among older patients aged 50-over 70 years. These include CLL (83.1%), MM (76.7%) and MDS (76.9%). On the other hand, NHL was almost evenly distributed among 20–49 years (50.7%) and 50-over 70 years (49.3%) age group. Among childhood HM cases 257 cases were in 15–19 year age group, while 81cases were in 0–14 year age group (Hossain et al., 2014).

In this study AML was the leading pattern with (33%) cases, one-fourth of the respondents were suffering from ALL (26%) followed by NHL (12%), HD (8.5%), MM

(8%), CML (7%), CLL (2%), Mixed Leukemia (2%) and MDS (1.5%). As the patterns of presentation, mode of treatment and outcome varies significantly from other types of AML, AML M3 was categorized separately. A total number of (10%) cases was found and the rest (23%) of AML other than M3 was found. In WHO prediction, the commonest type of HM was NHL, which was followed by Leukemias, HL and Multiple Myeloma. In Pakistan, NHL is the most prevalent type of HM (Ferlay et al., 2013). In US, NHL was the commonest cancer among HM, which was 1.5 times that of all leukemia (Alteri et al., 2020). Another study reported that acute leukemias including AML and ALL were the most prevalent HM affecting Bangladeshi population, accounting for (42.4%) of all HM cases, while these two constituted (66%) of leukemia cases. The frequency of AML was two times higher than that of ALL in Bangladesh (Hossain et al., 2014) which was similar with this study. Schottenfeld and Fraumeni Jr informed that the incidence of AML is relatively common in North America, Europe, and Oceania, while adult AML was rare in Asia and Latin America (Schottenfeld and Fraumeni, 2006). These findings were not similar with present study and this unexpected discrepancy might be due to lack of proper referral system in some participating centers. Another reason for AML being the predominant HM in this study was that in a country like Bangladesh people seeked advice from doctors when there was acute symptoms of disease. Many chronic conditions which does not present with acute symptoms were usually under reported. Although lymphoma was a haematological disorder, a small number of patients might have been admitted to the medical oncology department. Moreover, year-specific data was not available for the participating center. Another study reported NHL was one of the commonest cancers in developed countries, but the incidence was relatively lower in Asia (Mozaheb, 2012) which supports the finding of present study.

In present study almost all (99.5%) respondents had weight loss, three fourth of the respondents complained of fever (75.5%) followed by Bony Tenderness (50.5%), Lymphadenopathy (49.5%), Hepatomegaly (42%), Headache (22.5%), Gum Bleeding (15%), Purpura/Echymosis/Rash (13%), Weakness (9.5%), Jaundice (6.5%), Cough (5.5%), Back Pain(+/- Radiculopathy) (5%), Hematemesis/Malena (5%), Incidental findings (5%), Renal stone (4%), Oedema (3.5%), Hematuria (2.5%), Night Sweats (2.5%), Polyuria (2%), Fracture (2%), Pruritus (1.5%), Paraplegia/Quadriplegia (1.5%),

Vomiting/Nausea (0.5%), Angular Stomatitis (0.5%), Respiratory Distress (0.5%) and Ascitis/Pleural effusion (0.5%).

In regard of AML, in present study, the most common clinical feature was hepatomegaly (97.8%) and splenomegaly (97.8%) followed by fever (91.3%), Bony tenderness (71.7%), Headache (21.7%), rash 6 (13%), Weakness (8.7%), cough (4.3%) and the least feature was Gum bleeding (2.2%), hematuria (2.2%) and incidental findings (2.2%).

In terms of AML M3 most common (100%) clinical feature were fever and Gum bleeding followed by rash (75%), splenomegaly (70%), hepatomegaly (65%), Bony tenderness (65%), Hematemesis (50%), Weakness (25%), and the least feature was hematuria (20%).

In regard of ALL, in this study most of them had fever (90.4%) followed by bony tenderness (76.9%), headache (23.1%), gum bleeding (15.4%), rash (9.6%), weakness (5.8%) and cough (5.8%). More than half (61.5%) of them had lymphocytosis followed by Neutrophilia (19.2%), Neutropenia (9.6%), Pancytopenia (7.7%) and Lymphopenia (3.8%). A similar study reported that the most common symptoms include fever (caused by leukemia or a secondary infection secondary to neutropenia), fatigue and lethargy (as a result of anemia), bone and joint pain, and a bleeding diathesis. The most common laboratory abnormalities in ALL include anemia, thrombocytopenia, neutropenia, and leucopenia or leukocytosis, with hyperleukocytosis (>100×109 /L) present in approximately 15% of the pediatric patients. Other common laboratory abnormalities include elevated serum uric acid and lactose dehydrogenase levels. The lineage of ALL established in this manner subdivides this disease into two broad, clinically and biologically meaningful categories: precursor B-cell ALL (B-ALL) and precursor T cell ALL (T-ALL) (Onciu, 2009).

In this study in terms of CML most common 9 (64.3%) clinical features were fever and incidental findings followed by cough 6 (42.9%), splenomegaly 5 (35.7%), hepatomegaly 5 (35.7%), headache 2 (14.3%), back pain 2 (14.3%) and the least feature was Bony tenderness 1 (7.1%).

In regard of CLL the most common 4 (100%) clinical feature were fever, Weakness, Lymphadenopathy and Weight loss.

In terms of MDS most common 3 (100%) clinical feature was Weakness, Headache, Hepatomegaly and Splenomegaly. The least feature 1 (33.3%) was fever and weight loss.

Among the respondents suffering from mixed leukemia, the most common (100%) clinical feature was Fever followed by Hepatomegaly (75%), Splenomegaly (75%), Lymphadenopathy (50%) and the least feature (25%) was Headache and Gum Bleeding.

In regards of HD, the most common 17 (100%) clinical feature was Lymphadenopathy followed by fever 8 (47.1%), Weight Loss 7 (41.2%), Jaundice 3 (17.6%), Pruritus3 (17.6%) and the least feature 1 (5.9%) was Vomiting, Headache, Hepatomegaly and Splenomegaly.

In regards of NHL most common 24 (100%) clinical feature was Lymphadenopathy followed by fever 19 (79.2%), Weight Loss 11 (45.8%), Hepatomegaly 11 (45.8%), Splenomegaly 11 (45.8%), Night Sweats 5 (20.8%), Headache 4 (16.7%), Jaundice 6 (25%) and the least feature 1(4.2%) was Ascites, Headache, Paraplegia and Respiratory Distress.

In MM cases, most common feature was back pain (93.7%) followed by headache (87.5%), polyuria (43.7%), renal stone (18.6%) with paraplegia and fracture (6.2% each) being the least common.

A similar study revealed that most prevalent disease-related symptoms were: tiredness; feeling unwell; breathlessness; lack of energy; and back pain. The most prevalent treatment side effects were: tiredness; feeling sick; disturbance in sense of taste; and breathlessness (Goswami et al., 2020).

In case of diagnostic tools it is seen that CBC, PBF is compulsorily done for all 3 types of HM (100% each). Bone marrow study, Flowcytometry, Cytogenetic Study is necessary for Leukemia patients and Lymph node biopsy and immunohistochemistry along is necessary for Lymphoma patients. Whereas Plasma protein electrophoresis along with Bone marrow Study and Cytogenetic study are necessary in cases of Multiple Myeloma.

In this study, mean Hemoglobin, ESR, WBC and Platelet count of the respondents was (9.45±2.34) gm/dl, (62.92±30.11) mm in 1st hour, (60.89±50.78)×10^9/Litre, (233.96±128.65)×10^9/Litre of blood in male patients and (9.18±2.02)gm/dl, (67.74±26.98)mm in 1st hour, (63.68±53.14)×10^9/Litre and (228.40±109.35)×109/Litre of blood in female patients. There was no significant difference observed between male and female patients. Mean blast cell for male patients was (37.80±31.44) and for female patients was (40.76±30.62) percent respectively with no significant sex difference.

The most blast cells were found in AML except M3 (63.78±15.05) where minimum percentage was (34%) and maximum was (91%) followed by AML M3 (61.35±16.52), minimum (35%) and maximum (88%) and ALL (60.29±15.47) with minimum (30%) and maximum (91%). In MDS (22.29±4.75), we found that minimum and maximum percentage of blast were (46%) and (61%). Blast cells are not found in cases of Lymphoma and Myeloma.

In PBF, More than half of the respondents had Neutropenia (52.5%), followed by Lymphocytosis (17.5%), Neutrophillia (15%), Plasma Cell (8%), Myelocyte (7%), Promyelocyte (4.5%), Metamyelocyte (4.5%), Basophillia (4%), Increased Rouleaux (3%), Monocytosis (2%), Pancytopenia (2%), Eosinophillia (1.5%), Nucleated RBC (1.5%), Lymphopenia (1%), Hyper-segmented Neutrophil (1%) and Auer body (0.5%).

In cases of AML other than M3, AML M3, CLL, HD and NHL, All respondents (100%) had lymphocytosis. In ALL, More than half (61.5%) of them had lymphocytosis followed by Neutrophilia (19.2%), Neutropenia (9.6%), Pancytopenia (7.7%) and Lymphopenia (3.8%). In CML, all of them (100.0%) had Neutrophilia followed by Promyelocyte (64.3%), Metamyelocyte (64.3%), Basophillia (57.1%), Monocytosis (28.6%), and Eosinophillia (21.4%). In cases of MDS, All of them (100%) had Nucleated RBC followed by hyper segmented Neutrophil (66.7%), Auer Rod (33.3%). A similar study reported peripheral blood findings (PBF) of patients suffering from HM which showed Pancytopenia/bicytopenia (56%), Mixed picture (61%), Circulating blasts (100%), Leukocytosis (33%), Polycythemia (33%), Anemia (56%), Neutropenia (17%), Thrombocytosis (33%), Microcytosis (10%) and within normal limits was (50%). Patients with anemia, cytopenias, leukocytosis, polycythemia, and mixed presentations also had a significant rate of positivity, ranging between 20% and 69%.

The lowest yield was among cases of thrombocytopenia and neutropenia (0% and 14%, respectively) (Northrup et al., 2020).

In this study more than three fourth of the respondents (79.5%) had positive bone marrow findings and only (20.5%) had negative bone marrow findings. Majority of the respondents had AML (33%), among them AML other than M3 was (28.9%) and AML M3 (12.6%) followed by ALL (26%), MM (10.1%), CML (8.8%), CLL (2.5%), Mixed leukemia (2.5%) and MDS (1.9%). Ebrahim et al. in their study reported that 11.4% respondents have positive bone marrow findings. The prevalence of hematological malignancies was (3.5%) CML, (2.6%) AML, (1.8%) CLL and MM, (0.9%) ALL and undifferentiated acute leukemia. In patients with HM, (66.7%) of AML, (100%) of CML and CLL, and (75%) of MM patients had increased total WBC count, whereas (66.7%) of AML, (62.5%) of CML, (75%) of CLL, and (50%) of MM patients had decreased hemoglobin level. On the other hand, (66.7%) of AML and (50%) of CML, ALL, and CLL patients had decreased platelet count (Ebrahim et al., 2022).

In this study almost half of the respondents (49.5%) had positive flow cytometry findings and half of the respondents (50.5%) had negative flow cytometry findings. Majority of the respondents had T cell lineage (40.4%) followed by B cell lineage (30%) and Myeloid lineage (29.3%).

In ALL, predominantly T cell lineage was found (90%) in flowcytometry findings. On the contrary, (60.9%) showed myeloid lineage and only (4.3%) showed T cell lineage. In CLL, common finding in Flow Cytometry was B Cell Lineage (100%). In MDS, predominant lineage was myeloid lineage (33.3%). Zhang et al. in a similar study reported a significantly higher proportion of patients with a history of HM had positive peripheral blood flowcytometry results and independently identified history of HM as a significant predictor of a positive test result (Zhang et al., 2020).

In present study almost all respondents (92%) had negative plasma electrophoresis findings and only (8%) had positive plasma electrophoresis findings. In case of MM all the patients had done this investigation. In rest of the cases it is not relevant. Majority of the respondents had Monoclonal Gammopathy (3.5%) followed by Monoclonal Gammopathy with Beta 2 Microglobulinemia (2%), Light Chain Disease with Beta 2

Microglobulinemia (1.5%) and Light Chain Disease (1%). A study reported that among the positive plasma electrophoresis 10.66% cases had monoclonal gammopathy. Ten percent cases were diagnosed to be multiple myeloma (Tripathy, 2012) which does not support the findings of present study and this discrepancy might be due to racial variation.

In this study three fourth of the respondents (79.5%) had positive lymph node biopsy findings and only (20.5%) had negative lymph node biopsy findings. Only the Lymphoma cases had a lymph node biopsy done. In regard of positive lymph node biopsy findings majority of the respondents had NHL (12%) followed by HD (8.5%). It is not done in cases of Leukemia and Myeloma.

This study demonstrates that more than half of the respondents (69.5%) had negative cytogenetic biopsy findings and only (30.5%) had positive cytogenetic findings. In regard of positive cytogenetic study findings majority of the respondents had Philadelphia Chromosome (15.5%) followed by PML RARA positive cases (10%), CD 20 positive (1.5 %), CD 33 Positive (1.5%), FLT 3 Mutation (1.5%) and BCL 2 Positive case (0.5%).

In AML except M3, cytogenic study finding were Philadelphia Chromosome (58.8%), FLT 3 Mutation (23.5%) and CD33 positive (17.6%). All the patients of AML M3 were positive for PML RARA (100%). All cases (100%) were positive for Philadelphia chromosome in cases of CML. In case of ALL, Cytogenetic study finding was Philadelphia Chromosome (13.5%). Many of them could not afford to do the test. In cytogenetic study of CLL, it was CD20 Positive (75%) and BCL 2 positive (25%). A study revealed that discovery of the Philadelphia chromosome and other chromosomal aberrations paved the way for the characterization of molecular lesions that lead to phenotypic characteristics of a number of hematologic malignancies. Metaphase cytogenetics (MC) has proven an extremely valuable diagnostic tool, providing definite diagnoses, for example, in the case of balanced pathognomonic translocations in chronic myeloid leukemia (CML), acute promyelocytic leukemia (APL), or core binding factor acute myelogenous leukemias (AMLs). Chromosomal aberrations support the cytomorphologic diagnosis and provide prognostic information, for example, in myelodysplastic syndromes (MDSs), AML with unbalanced translocations,

multiple myeloma (MM) and chronic lymphocytic leukemia (CLL). Irrespective of their location, size, and corresponding phenotype, chromosomal defects also represent suitable markers of clonality suggestive of clonal dominance by the malignant clone or, in rare circumstances, of oligoclonality due to a profound depletion of stem cell reserves (Maciejewski and Mufti, 2008) which supports the findings of present study.

In this study more than half of the respondents (87.5%) had negative immuno histo chemistry findings and only (12.5%) had positive immuno histo chemistry findings. In regard of positive immuno histo chemistry findings majority of the respondents had CD 20 positive findings (6.5%) and (6%) had CD 30. In cases of HD (70.6%) respondents were positive for CD 30 whereas (54.2%) NHL respondents were positive for CD20.

In present study more than three fourth (87%) patients had taken Chemotherapy. A total of 17 types of chemotherapeutic drugs were used in patients with HM who are enrolled in this study. Most commonly used chemotherapeutic drug among the respondents suffering from hematological malignancy was Daunorubicin (51%) followed by Prednisolone (40%), Vincristine (35.5%), Cytarabine (27.5%), Methotrexate (26%), L Aspaginase (23%), Cyclophosphamide (12%), Arsenic Trioxide (10%), Vesanoid (ATRA) (10%), Dexamethasone (9.4%), Bleomycin (8.5%), Vinblastine (8%), Dacarbazine (8%), Bordezomib (8%), Linamide (8%), Doxorubicin (3.5%) and 6 Mercaptopurine (2.5%).

In present study, among the respondents that used Doxorubicin, Daunorubicin and Bleomycin were mostly supplied by Government fund. Vinblastine, Vincristine, Dacarbazine, Prednisolone, Cytarabine, Arsenic Trioxide, Vesanoid, Bordezomib, Linamide, Dexamethason, L Aspaginase, 6 Mercaptopurine, Methotrexate were mostly had to be acquired by patient's own fund from private sources. Radiotherapy and Bone Marrow Transplantation is unavailable in Chittagong. As Radiotherapy machine in CMCH is currently under maintenance and no other private institution provides this facility in Chittagong, it was impossible for the respondents to take radiotherapy. Once the machine in CMCH becomes ready for service, it is literally a daunting task to get the serial as this is the only Radiotherapy facility for whole of Chittagong division.

In AML cases, All (100%) of them underwent through chemotherapy, none of them had bone marrow transplant (0%) and only (26.1%) had immunotherapy. Most commonly used chemotherapeutic drugs were Daunorubicin and Cytarabine (100%) each. In terms of immunotherapy more than three fourth (83.3%) respondents have taken Imatinib and only (16.7%) have taken Midostaurin. In AML M3 cases, all (100%) patients received Arsenic Trioxide (ATO) and Vesanoid (ATRA).

In regard of ALL, All of them underwent through chemotherapy, none of them had bone marrow transplant (0%) and only (13.5%) had immunotherapy. In the term of treatment of ALL, all of them had Prednisolone (100%) and Daunorubicin (100%) followed by Methotrexate (98.1%), Vincristine (90.4%), L Aspaginase (88.5%), Imatinib (13.5%), Cytarabine (9.6%), 6-Mercaptopurine (9.6%) and Bleomycin (1.9%).

In terms of CML, all (100%) of them underwent through immunotherapy which was Imatinib, none of them (0%) had chemotherapy and bone marrow transplantation. In regard of CLL, all (100%) of them underwent through immunotherapy (50%) of them was Venetoclox and the rest (50%) of them received Rituximab. None of them had chemotherapy, radiotherapy and bone marrow transplant. A study reported that approximately two thirds of patients with CLL presented with Binet stage-A (66.5%) and/or Rai stage 0 or 1 (63%). Most patients (92.3%) had an ECOG PS of 0 or 1 (range, 81.3% in Chile to 93.6% in Brazil) at diagnosis. The most common initial treatment of CLL was chlorambucil with or without prednisone (191; 46%) followed by fludarabine, cyclophosphamide, and rituximab (FCR; 87; 21%) and fludarabine and cyclophosphamide (34; 8.2%). The majority of patients in Brazil (108 of 181; 59.7%) and Mexico (57 of 95; 60%) received chlorambucil with or without prednisone. FCR was the most frequent initial chemotherapy regimen in Argentina (53.6%), Panama/Guatemala (50%), Colombia (40%), and Chile (38.5%) (Tietsche et al., 2019). Chlorambucil was previously used before the development of specific target therapies. We found that treatment modalities of CLL in CMCH is similar to the new advanced protocols.

In terms of MDS, (60%) of them received Azacytidine. None of them (0%) had bone marrow transplant. Other most commonly used chemotherapeutic drugs were Daunorubicin and Dexamethasone. In regards of mixed Leukemia, All (100%) of them

underwent through chemotherapy, none of them had bone marrow transplant 0 (0%) and immunotherapy. Most commonly used (100%) chemotherapeutic drugs were Doxorubicin, Prednisolone, Daunorubicin, Cytarabine and only (25%) Methotrexate.

In regards of HD, All (100%) of them underwent through chemotherapy, none of them had bone marrow transplant (0%) and only (35.3%) had immunotherapy (Brentuximab Vedotin). Most commonly used chemotherapeutic drugs (100%) were Daunorubicin, Bleomycin and Vinblastine and Dacarbazine. (ABVD Protocol).

In regards of NHL All 24 (100%) of them underwent through chemotherapy, none of them had bone marrow transplant 0 (0%) and only 13 (54.2%) had immunotherapy (Rituximab). Most commonly used Chemotherapeutic (100%) drugs were Doxorubicin, Cyclophosphamide, Vincristine and Prednisolone. (R-CHOP) A similar study reported that more than two thirds of patients with NHL (68.6%) had either diffuse large B-cell lymphoma (DLBCL; 1,457; 49.1%) or follicular lymphoma (FL; 578; 19.5%). The majority of patients with DLBCL or FL received rituximab plus cyclophosphamide, doxorubicin, vincristine, and prednisone (Tietsche et al., 2019). In cases of MM, All (100%) received Bordezomib, Linsmide and Dexamethasone and only (25%) got Cyclophosphamide.

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Chapter 6

Conclusion

This study of a large number of HM patients is a very first step in understanding the patterns and distribution of HM in Chittagong, Bangladesh. Initially overall picture was described and then category wise HM subtypes were divided into leukemia, lymphoma and myeloma. Then clinical features, laboratory investigations and their findings, treatment modalities were individually described for each type of leukemia, lymphoma and myeloma. It was seen that AML was the leading pattern with of all HM, one-fourth of the respondents were suffering from ALL and MDS was the least occurring one. Among them almost all respondents had weight loss, three fourth of the respondents complained of fever followed by bony tenderness, lymphadenopathy, hepatomegaly, splenomegaly etc. In case of diagnostic tools it was seen that CBC, PBF was compulsorily done for all 3 types of HM. There was no significant difference observed between male and female patients. Mean blast cell for was 36.03±18.07. The presence of blast cells in peripheral blood is almost diagnostic in cases of leukemia. Bone marrow study, Flowcytometry, Cytogenetic Study is necessary for Leukemia patients and Lymph node biopsy and immunohistochemistry along is necessary for Lymphoma patients. Whereas Plasma protein electrophoresis along with Bone marrow Study and Cytogenetic study are necessary in cases of Multiple Myeloma. More than half of the respondents had Neutropenia, followed by Lymphocytosis, Neutrophillia, plasma Cell etc. In flow cytometry findings majority of the respondents had T lymphocyte markers followed by B lymphocyte markers and myeloid markers. Among the Leukemia patients, cytogenetic study findings states that majority of the respondents were positive for Philadelphia Chromosome followed by PML RARA. CD33 and CD 20 positive cases and FLT3 Mutation was among the least. Among the lymphoma patients immunohistochemistry findings states that majority of patients were positive for CD 20. Among our study population more than three fourth patients had taken Chemotherapy as a mode of their treatment. Only one third of them got Immunotherapy/Target therapy. Radiotherapy and Bone Marrow Transplantation is unavailable in Chattogram.

When comparing various types of HM we found that in AML, the most common clinical feature was hepatomegaly and splenomegaly followed by fever and Bony

tenderness. Different picture was found in a variety of AML, AML M3 where patients most commonly had fever and Gum bleeding followed by rash, splenomegaly and hepatomegaly. In case of patients suffering from ALL, most of them had fever followed by bony tenderness. In CML patients the most common clinical feature were fever and incidental findings. Fever and Lymphadenopathy was almost always present for CLL patients. In patients of HD, lymphadenopathy is the most common symptom followed by fever and weight Loss. The patients suffering from NHL most commonly had lymphadenopathy followed by fever and weight loss. Hepatomegaly and Splenomegaly were found to be similar in CLL and HD patients. In cases of MM, patients presents with back pain (+/-radiculopathy) followed by headache, polyuria and renal Stone.

Regarding treatment, AML patients except AML M3 has all got Daunorubicin and Cytarabine and in terms of immunotherapy more than three fourth patients have taken Imatinib and only 2 patients have taken Midostaurin procured from abroad. In case of AML M3 all patients received ATRA and Arsenic Trioxide. In the term of treatment of ALL, all of them had Prednisolone and Daunorubicin followed by Methotrexate and Vincristine, L Aspaginase, Cytarabine, 6-Mercaptopurine and Bleomycin. In case of HD, only one third had immunotherapy (Brentuximab Vedotin) procured from abroad. Most commonly used chemotherapeutic drugs were Daunorubicin, Bleomycin and Vinblastine and Dacarbazine which is the standard protocol for HD (ABVD Protocol). In case of NHL only half of the patients had immunotherapy (Rituximab). Most commonly used chemotherapeutic drugs were Doxorubicin, Cyclophosphamide, Vincristine and Prednisolone. It is the standard protocol for NHL treatment (R-CHOP Protocol). In case of CML Imatinib was the mainstay of therapy and in CLL cases half of them took Rituximab owing to their CD 20 positive report. Then the availability of chemotherapeutic drugs, target therapies were sorted out and any scarcity of drugs are addressed. Most chemotherapeutic drugs and all immunotherapeutic drugs were supplied from private source. Radiotherapy is currently unavailable in Chattogram division and it is a serious concern related to the successful treatment of HM. Similarly bone marrow transplantation is also unavailable. It is a major modality of treatment in relapse or refractory cases of HM. HM can occur in any age group, in both male and female group. Further investigations are necessary to understand the epidemiology, potential risk factors, biology and genetics of hematological malignancies in this country in rapid transition.

Chapter 7
Limitation

- As there is no proper and effective hospital record keeping system in Bangladesh, there are always possibilities that the same patient might visited multiple times in the same hospitals and/or different hospitals and a large pool of patients might have missed being enrolled.
- Many patients of HM go to abroad and we have no data regarding their disease frequency and outcome.
- Due to time and resource constrain, larger sample could not be studied.
- Treatment response could not be evaluated due to scarcity of fund and lack of Laboratory facilities.
- As radiotherapy is currently unavailable in Chittagong, many patients needed chemotherapy but could not get one.
- It was not possible to get information on the cancer-associated complications and deaths due to lack of proper record keeping system.
- Population-based studies that examine these additional factors are needed to understand the root causes for disparities**.**
- Risk factor of HM, its association with lifestyle, food habit, stress, pollution and its genetic and epigenetic variations could not be focused as these were beyond the limit of this study. Further research regarding Population-based studies that examine these additional factors are needed to understand the root causes for disparities and we are in dire need of both government and private collaboration in this sector.

Chapter 8

Recommendations

- As there are always possibilities of duplication due to visit of same patient multiple times in the same hospitals, to eliminate these possibilities, raw data should be thoroughly analyzed to clean up such duplicated, re-enrolled cases.
- Although small in quantity, some anticancer drugs and vaccines are manufactured by some the local companies to cool down the current situation. The government should focus on this sector to tackle the massive need for these chemotherapy drugs.
- National data base must be build up in order to enroll all patients with HM, so that a clear disease burden could be identified and policy could be made in order to tackle this situation.
- Government should ensure more supply of chemotherapeutic drugs in hospital settings for majority poor people of this country. Simultaneously government should ensure and encourage production of existing chemotherapy drugs and newer drugs of target therapy.
- In case laboratory facilities, more and more well equipped centers must be developed. Government should encourage private funding in this sector as much as possible.
- Urgent set up of a cancer center with well-equipped facility is a must if one wants to enrich HM cancer treatment facility in Bangladesh.

Chapter 9

References

Abdulsalam. 2010. Laboratory diagnosis of acute leukemia in Iraq. Turkish Journal of hematology, 27(4): 320-321

Alteri R, Christy M, Daniels E, Daniels K, Delfavero M, Fedewa S, Hargrove T, Kalidas M, Koerner TJ, McMahon C, Tyson DM, Pinheiro P, Richard C, Sauer AG, Simpson S, Singleton S, Sloan K, Torre L, Wagner D, Young K. 2020. Cancer Facts and Figures for Hispanic/Latinos 2018-2020. American Cancer Society, Atlanta: 1-52, available from: http://www.cancer.org/research/cancer-facts-statistics/hispanics-latinos-facts-figures.html

Arber DA, Orazi A, Hasserjian R, Thiele J, Borowitz MJ, Le Beau MM, Bloomfield CD, Cazzola M, Vardiman. 2016. The 2016 revision to the World Health Organization classification of myeloid neoplasms and acute leukemia. The Journal of the American Society of Hematology, 127(20): 2391-2405.

Bach PB, Schrag D, Brawley OW, Galaznik A, Yakren S, Begg. 2002. Survival of blacks and whites after a cancer diagnosis. Journal of the American Medical Association, 287(16): 2106-2113.

Bain BJ. 2005. Diagnosis from the blood smear. New England Journal of Medicine, 353(5): 498-507.

Chabner BA, Longo DL. 2015. Cancer Chemotherapy, Immunotherapy and Biotherapy, 6th edn., Wolters Kluwer, Singapore, 1-561p

Cokkinides V, Albano J, Samuels A, Ward M, Thum J. 2005. Cancer facts and figures. American cancer society, 5008(05): 1-64

Curado MP, de Souza. 2014. Cancer burden in Latin America and the Caribbean. American Cancer Society, 80(5): 370-377.

Das P, Horton. 2013. Bangladesh: innovating for health. The Lancet, 9906(382): 1681-1682.

De Lange K, de Boer EN, Bosga A, Alimohamed MZ, Johansson LF, Mulder AB, Vellenga E, van Diemen CC, Deelen P, van den Berg. 2020. Targeted RNA-Sequencing Enables Detection of Relevant Translocations and Single Nucleotide Variants and Provides a Method for Classification of Hematological Malignancies–RANKING. Clinical Chemistry, 66(12): 1521-1530.

Delsol G, Ralfkiaer E, Stein H, Wright D, Jaffe. 2007. Pathology and genetics of tumors of haematopoietic and lymphoid tissues: World Health Organization Classification of Tumours. National Library of Medicine, 110(7): 230-235.

Ebrahim H, Fisha T, Debash H, Bisetegn. 2022. Patterns of Bone Marrow Confirmed Malignant and Non-Malignant Hematological Disorders in Patients with Abnormal Hematological Parameters in Northeast Ethiopia. Journal of Blood medicine, 13: 51-60.

Farhad MN, Islam GY, Wasim M, Biswas AR, Tompa. 2021. Pattern of Haematological Malignant Patients Attending OPD in a Tertiary Level Hospital in A Calendar Year. Hematology Journal of Bangladesh, 5(01): 20-26.

Ferlay J, Soerjomataram I, Ervik M, Dikshit R, Eser S, Mathers C, Rebelo M, Parkin D, Forman D, Bray F. 2013. Cancer incidence, mortality and prevalence worldwide in 2012: GLOBOCAN 2012. International Journal of Cancer, 136(5):E359-86. doi: 10.1002/ijc.29210

Flowers CR., Glover R, Lonial S, Brawley. 2007. Racial differences in the incidence and outcomes for patients with hematological malignancies, Current Problems in Cancer, 31(3): 182-201.

Goswami P, Oliva, E.N, Ionova T, Else R, Kell J, Fielding AK, Jennings DM, Karakantza M, Al-Ismail S, Collins GP, McConnell S, Langton C, Salek S. 2020. Quality-of-life issues and symptoms reported by patients living with haematological malignancy: a qualitative study, Therapeutic Advances in Hematology, 07(11): 2040620720955002. doi: 10.1177/2040620720955002.

Greer JP, Foerstar J, Rodgars GM, Paraskevas F, Glader B, Arber DA, Means Jr RT. 2009. Wintrobe's clinical hematology. 12th edn., Lippincott Williams & Wilkins, Philadelphia, 2236p

Haque M. 2009. ABC of research methodology and biostatistics, 5th edn., Dolphin Medical books, Dhaka, 1-400 p

Hicks L. K, Bering H, Carson KR, Haynes AE, Kleinerman J, Kukreti V, Ma A, Mueller BU, O'Brien SH, Panepinto P. 2014. Five hematologic tests and treatments to question. The Journal of the American Society of Hematology, 124(24): 3524-3528.

Hochhaus A, Larson RA, Guilhot F, Radich JP, Branford S, Hughes TP, Baccarani M, Deininger MW, Cervantes F, Fujihara S, Ortmann CE, Menssen HD, Kantarjian H, O'Brien SG, Druker BJ. 2017. Long-term outcomes of imatinib treatment for chronic myeloid leukemia. New England Journal Medicine, 376(10): 917-927.

Hossain MS, Iqbal MS, Khan MA, Rabbani MG, Khatun H, Munira S, Miah M, Kabir AL, Islam N, Dipta TF, Rahman F, Mottalib A, Afrose S, Ara T, Biswas AR, Rahman M, Abedin AM, Rahman M, Yunus AB, Niessen LW, Sultana TA. 2014. Diagnosed hematological malignancies in Bangladesh-a retrospective analysis of over 5000 cases from 10 specialized hospitals. BMC Cancer, 14(1): 1-7.

Howlader N, Noone AM, Krapcho M, Miller D, Brest A, Yu M, Ruhl J, Tatalovich Z, Mariotto A, Lewis DR, Chen HS, Feuer EJ, Cronin KA. 2019. SEER data submission for SEER Cancer Statistics Review (1975-2016), November 2018, National Cancer Institute. Bethesda, Maryland,USA, 13:31-35.

Hussain SA, Sullivan RJ. 2013. Cancer control in Bangladesh. Japanese journal of clinical oncology, 43(12): 1159-1169.

Jemal A, Siegel R, Ward E, Murray T, Xu J, Smigal C, Thun MJ. 2006. Cancer statistics. CA cancer journal for clinicians, 56(2):106-130.

Lichtman MA. 2008. Battling the hematological malignancies: the 200 years' war. The Oncologist, 13(2): 126-138.

Maciejewski JP, Mufti G. 2008. Whole genome scanning as a cytogenetic tool in hematologic malignancies. The Journal of the American Society of Hematology, 112(4): 965-974.

Mozaheb Z. 2012. Epidemiology of Lymphoid Malignancy in Asia, In: Maria De Lourdes Ribeiro de Souza da Cunha, (ed.). Epidemiology Insights, IntechOpen, London, 325-354p, doi 10.5772/31746.

Northrup V, Maybank A, Carson N, Rahmeh T. 2020. The value of next-generation sequencing in the screening and evaluation of hematologic neoplasms in clinical practice. AM Journal Clinical Pathology, 153(5): 639-645.

Onciu MJ. 2009. Acute lymphoblastic leukemia. Hematology/Oncology Clinics of North America, 23(4): 655-674.

Paudyal P, Pradhan A, Pokharel S, Shah N, Pradhan B, Poudel P. 2018. Pattern of hematological malignancies diagnosed by peripheral smear examination. Journal of BP Koirala Institute of Health Sciences, 1(1): 25-35.

Ramdass B, Chowdhary A, Koka PS. 2013. Hematological malignancies: disease pathophysiology of leukemic stem cells. Journal of Stem cells, 8(3/4): 151.

Rodriguez-Abreu D, Bordoni A, Zucca E. 2007. Epidemiology of hematological malignancies. Annals of Oncology, 18: i3-i8. doi: 10.1093/annonc/mdl443.

Ruppert AS, Dixon JG, Salles G, Wall A, Cunningham D, Poeschel V, Haioun C, Tilly H, Ghesquieres H, Ziepert MJ. 2020. International prognostic indices in diffuse large B-cell lymphoma: a comparison of IPI, R-IPI, and NCCN-IPI. The American Society of hematology-Blood, 135(23): 2041-2048.

Sarwar SMR, Lona H, Halder D, Selim S. 2020. Hematological Malignancies Pattern among Bangladeshi Adults. Journal of Medical Science and Clinical Research, 5: 6-9.

Schottenfeld D, Fraumeni Jr. 2006. Cancer epidemiology and prevention, 3rd edn., Oxford University Press, 841-871p

Sharmin T, Nikhat N, Rayna SE, Khalequzzaman M, Khan FA, Rahman KT, Islam T, Haque SE, Ahsan H, Kamal M. 2022. Types and distribution of cancer patients attending in a tertiary care hospital of Bangladesh. BSMMU Journal, 15(1): 43-49.

Taylor J, Xiao W, Wahab A. 2017. Diagnosis and classification of hematologic malignancies on the basis of genetics. The Journal of the American Society of Hematology, 130(4): 410-423.

Tietsche de Moraes Hungria V, Chiattone C, Pavlovsky M, Abenoza LM, Agreda GP, Armenta J, Arrais C, Avendaño Flores O, Barroso F, Basquiera AL. 2019. Epidemiology of hematologic malignancies in real-world settings: findings from the Hemato-oncology Latin America observational registry study. Journal of Global Oncology, 5: 1-19.

Tripathy SJ. 2012. The role of serum protein electrophoresis in the detection of multiple myeloma: An experience of a corporate hospital. Journal of Clinical Diagnosis and Research, 6(9): 1458.

Vardiman JW, Thiele J, Arber DA, Brunning RD, Borowitz MJ, Porwit A, Harris NL, Le Beau MM, Hellström-Lindberg E, Tefferi A, Bloomfield CD. 2009. The 2008 revision of the World Health Organization (WHO) classification of myeloid neoplasms and acute leukemia: rationale and important changes. The Journal of the American Society of Hematology, 114(5): 937-951.

Zhang ML, Guo AX, Kadauke S, Dighe AS, Baron JM, Sohani AR. 2020. Machine learning models improve the diagnostic yield of peripheral blood flow cytometry. American Journal of Clinical Pathology, 153(2): 235-242.

Appendix-I

Informed written consent form for subjects

Title: Hematological Malignancies in Chattogram region: A cross sectional study

Principal Investigator: Dr. Ambarish Mitra

Name of participant: ----------

Name of Investigator: Dr. Ambarish Mitra

1. I consent to participate in the project named above, the particulars of which including details of interviews and questionaries' have been explained to me. A written copy of the information's has been given to me to keep with.
2. I authorize the researcher to use with me the interviews and questionaries' referred to under (1) above.
3. I acknowledge that:
   a. The possible effects of the interviews and questionaries' have been explained to me to my satisfaction.
   b. I have been informed that I am free to withdraw myself from the project at any time without explanation or prejudice and to withdraw any unprocessed data previously supplied.
   c. The project is for purpose of research.
   d. I have been informed that the confidentiality of the information's will be safeguarded.
   e. I have been informed regarding the interviews. I have also been informed that because of the number of people to be interviewed are small ; it is possible that someone may still be able to identify me on the basis of any references to personal information that might allow someone to guess my identity. However, I will be referred by pseudo name or identified by a different name in any publications arising from the research.

Signature date

(Participant)

Signature date

(Witness to consent)

Appendix - II

Questionnaire (English)

"Patterns of Common Hematological Malignancies in Chattogram: An Overview"

Thesis for MPH One Health done by

Principal investigator: Dr. Ambarish Mitra

SUPERVISER: PROF. DR. S K M AZIZUL ISLAM
Professor, Department of Physiology and Pharmacology, CVASU, Chattogram

1. Socio Demographic Distribution:

Name:	
1.1 Patient's ID	
1.2 Age (In years, completed)	
1.3 Sex	1.3.1 Male ☐ 1.3.2 Female ☐
1.4 Address	Upazilla…….. District…… Rural ☐ Urban☐
1.5 Occupation	1.5.1 Student ☐ 1.5.2 Farmer ☐ 1.5.3 Painter ☐ 1.5.4 Plumber ☐ 1.5.5 Driver ☐ 1.5.6 Fisherman ☐ 1.5.7 Carpenter Teacher ☐ 1.5.8 Steel Mill Worker ☐ 1.5.9 Brick Field worker ☐ 1.5.10 Garments Factory Worker ☐ 1.5.11 Dry Fish Factory worker ☐ 1.5.12 Service Holder ☐ 1.5.13 Businessman ☐ 1.5.14 Retired ☐ 1.5.15 House wife ☐ 1.5.16 Teacher ☐ 1.5.17 Unemployed ☐ 1.5.18 Radiology Technologist ☐ 1.5.19 Engineer ☐ 1.5.20 Physician ☐
1.6 Monthly Income	1.6.1 5000-25,000/- BDT ☐
	1.6.2 25,001-50,000/- BDT ☐
	1.6.3 50,001-1,00,000/- BDT ☐
	1.6.4 More than 1,00,000/- BDT ☐
1.7 Family History	Present ☐ Absent☐

2. Clinical Features leading to diagnosis:

2.1	Fever	☐
2.2	Gum Bleeding	☐
2.3	Purpura/Echymosis/Rash	☐
2.4	Hematemesis/Malena	☐
2.5	Hematuria	☐
2.6	Weakness	☐
2.7	Vomiting/Nausea	☐
2.8	Oral Ulcer	☐
2.9	Headache	☐
2.10	Oedema	☐
2.11	Hepatomegaly	☐
2.12	Spleenomegaly	☐
2.13	Lymphadenopathy	☐
2.14	Angular stomatitis	☐
2.15	Bony Tenderness	☐
2.16	Renal stone	☐
2.17	Fracture	☐
2.18	Back Pain(+/- Radiculopathy)	☐
2.19	Paraplegia/Quadriplegia	☐
2.20	Cough	☐
2.21	Respiratory Distress	☐
2.22	Jaundice	☐
2.23	Pruritus	☐
2.24	Incidental Finding	☐
2.25	Weight Loss	☐
2.26	Night Sweat	☐
2.27	Ascitis/Pleural Effusion	☐
2.28	Polyuria	☐

3. Investigation tools:

3.1	Complete Blood Count and Peripheral Blood Film	3.3.1 Neutropenia 1=yes☐ 2=No ☐ 3.3.2 Neutrophillia 1=yes☐ 2=No ☐ 3.3.3 Lymphocytosis 1=yes☐ 2=No ☐ 3.3.4 Lymphopenia 1=yes☐ 2=No ☐ 3.3.5 Eosiniphillia 1=yes☐ 2=No ☐ 3.3.6 Monocytosis 1=yes☐ 2=No ☐ 3.3.7 Basophillia 1=yes☐ 2=No ☐ 3.3.8 Pancytopenia 1=yes☐ 2=No☐ 3.3.9 Promyelocyte 1=yes☐ 2=No ☐ 3.3.10 Myelocyte 1=yes☐ 2=No ☐ 3.3.11 Metamyelocyte 1=yes☐ 2=No☐ 3.3.12 Plasma Cell 1=yes☐ 2=No☐ 3.3.13 Increased Rouleax Formation 1=yes☐ 2=No☐ Hb%.....gm/dl ESR.....mm in 1st Hour WBC Total Count.....X10^9/L WBC Differential Count....check on above box Platelet Count.....X10^9/L

		Blast Cell……. %
3.2	Bone Marrow Study	3.2.1 Acute Lymphoblastic Leukemia ☐ (ALL) 3.2.2 Acute Myeloblastic Leukemia ☐ (AML) 3.2.3 Chronic Lymphoblastic Leukemia(CLL) ☐ 3.2.4 Chronic Myeloid Leukemia (CML) ☐ 3.2.5 Myelodysplastic Syndrome ☐ 3.2.6 Hodgkin Lymphoma ☐ 3.2.7 Non Hodgkin Lymphoma ☐ 3.2.8 Multiple Myeloma ☐
3.3	Flow cytometry	Is it done? 1=yes☐ 2=No☐ If yes, 3.3.1.1 T Cell Lineage 1=yes☐ 2=No☐ 3.3.1.2 B Cell Lineage 1=yes☐ 2=No ☐ 3.3.1.3 Myeloid Lineage 1=yes☐ 2=No ☐ If No, Not done due to : 3.3.2.1 Unavailability 1=yes☐ 2=No☐ 3.3.2.2 High Cost 1=yes☐ 2=No☐ 3.3.2.3 Not Relevant ☐
3.4	Plasma Protein Electrophoresis	Is it done? 1=yes☐ 2=No☐ If Yes, 3.4.1 Monoclonal Gammopathy 1=yes☐ 2=No☐ 3.4.2 Light Chain Disease 1=yes☐ 2=No☐ 3.4.3 Beta 2 Microglobulin 1=yes☐ 2=No☐ If No, Not done due to :

		3.4.2.1 Unavailability 1=yes☐ 2=No☐ 3.4.2.2 High Cost 1=yes☐ 2=No☐ 3.4.2.3 Not Relevant ☐
3.5	Lymph node Biopsy	Is it done? 1=yes☐ 2=No☐ If Yes 3.5.1 Hodgkin Lymphoma ☐ 3.5.2 Non Hodgkin Lymphoma ☐ If No, Not done due to : 3.5.2.1 Unavailability 1=yes☐ 2=No☐ 3.5.2.2 High Cost 1=yes☐ 2=No☐ 3.5.2.3 Not Relevant ☐
3.6	Cytogenetic Study	Is it done? 1=yes☐ 2=No☐ If Yes, 3.6.1.1 Philadelphia chromosome ☐ 3.6.1.2 CD33 ☐ 3.6.1.3 FLT3 Mutation ☐ 3.6.1.4 PML RARA ☐ 3.6.1. 5 CD20 ☐ 3.6.1.6 BCL 2 ☐ If No, Not done due to : 3.6.2.1 Unavailability 1=yes☐ 2=No☐ 3.6.2.2 High Cost 1=yes☐ 2=No☐ 3.6.2.3 Not Relevant ☐
3.7	Immunohistochemistry	Is it Done? 1=yes☐ 2=No☐ If Yes, 3.7.1.1 CD30 ☐ 3.7.1.2 CD20 ☐ If No, Not done due to : 3.7.2.1 Unavailability 1=yes☐ 2=No☐ 3.7.2.2 High Cost 1=yes☐ 2=No☐ 3.7.2.3 Not Relevant ☐

4. Availability of investigation facility:

4.1 CBC, PBF

4.1.1	Available in Chattogram	4.1.1.1 At Chittagong Medical College 1=yes☐ 2=No☐ 4.1.1.2 At Private Laboratory 1=yes☐ 2=No☐
4.1.2	Available in Dhaka	1=yes☐ 2=No☐
4.1.3	Unavailable in Bangladesh	1=yes☐ 2=No☐

4.2 Bone Marrow Study

4.2.1	Available in Chattogram	4.2.1.1 At Chittagong Medical College 1=yes☐ 2=No☐ 4.2.1.2 At Private Laboratory 1=yes☐ 2=No☐
4.2.2	Available in Dhaka	1=yes☐ 2=No☐
4.2.3	Unavailable in Bangladesh	1=yes☐ 2=No☐

4.3 Flow Cytometry

4.3.1	Available in Chattogram	4.3.1.1 At Chittagong Medical College 1=yes☐ 2=No☐ 4.3.1.2 At Private Laboratory 1=yes☐ 2=No☐
4.3.2	Available in Dhaka	1=yes☐ 2=No☐
4.3.3	Unavailable in Bangladesh	1=yes☐ 2=No☐

4.4 Plasma protein Electrophoresis

4.4.1	Available in Chattogram	4.4.1.1 At Chittagong Medical College 1=yes☐ 2=No☐ 4.4.1.2 At Private Laboratory 1=yes☐ 2=No☐
4.4.2	Available in Dhaka	1=yes☐ 2=No☐
4.4.3	Unavailable in Bangladesh	1=yes☐ 2=No☐

4.5 Lymph node Biopsy

4.5.1	Available in Chattogram	4.5.1.1 At Chittagong Medical College 1=yes☐ 2=No☐ 4.5.1.2 At Private Laboratory 1=yes☐ 2=No☐
4.5.2	Available in Dhaka	1=yes☐ 2=No☐
4.5.3	Unavailable in Bangladesh	1=yes☐ 2=No☐

4.6 Cytogenetic Study

4.6.1	Available in Chattogram	4.6.1.1 At Chittagong Medical College 1=yes☐ 2=No☐ 4.6.1.2 At Private Laboratory 1=yes☐ 2=No☐
4.6.2	Available in Dhaka	1=yes☐ 2=No☐
4.6.3	Unavailable in Bangladesh	1=yes☐ 2=No☐

4.7 Immunohistochemistry

4.7.1	Available in Chattogram	4.7.1.1 At Chittagong Medical College 1=yes☐ 2=No☐ 4.7.1.2 At Private Laboratory 1=yes☐ 2=No☐
4.7.2	Available in Dhaka	1=yes☐ 2=No☐
4.7.3	Unavailable in Bangladesh	1=yes☐ 2=No☐

5 Specific Treatment Modalities:

5.1	**Chemotherapy** 1=yes☐ 2=No☐ If Yes, then which drugs are given		
	5.1.1 Doxorubicin ☐	Govt supply=1☐	Private fund=2☐
	5.1.2 Bleomycin ☐	Govt supply=1☐	Private fund=2☐
	5.1.3 Vinblastin ☐	Govt supply=1☐	Private fund=2☐
	5.1.4 Dacarbazine ☐	Govt supply=1☐	Private fund=2☐
	5.1.5 Cyclophosphamide ☐	Govt supply=1☐	Private fund=2☐
	5.1.6 Vincristine ☐	Govt supply=1☐	Private fund=2☐
	5.1.7 Prednisolone ☐	Govt supply=1☐	Private fund=2☐
	5.1.8 Daunorubicin ☐	Govt supply=1☐	Private fund=2☐
	5.1.9 Cytarabine ☐	Govt supply=1☐	Private fund=2☐
	5.1.10 Arsenic Trioxide ☐	Govt supply=1☐	Private fund=2☐
	5.1.11 Vesanoid (ATRA) ☐	Govt supply=1☐	Private fund=2☐
	5.1.12 Bortezomib ☐	Govt supply=1☐	Private fund=2☐
	5.1.13 Linamide ☐	Govt supply=1☐	Private fund=2☐
	5.1.14 Dexamethason ☐	Govt supply=1☐	Private fund=2☐
	5.1.15 L Asparginase ☐	Govt supply=1☐	Private fund=2☐
	5.1.16 6 Mercaptopurine ☐	Govt supply=1☐	Private fund=2☐
	5.1.17 Methotrexate ☐	Govt supply=1☐	Private fund=2☐
	If No, Not done due to : 5.1.2.1 Unavailability 1=yes☐ 2=No☐ 5.1.2.2 High Cost 1=yes☐ 2=No☐ 5.1.2.3 Not Relevant ☐		

5.2	**Radiotherapy** 1=yes☐ 2=No☐ If Yes, Then 5.2.1 Brachytherapy ☐ 5.2.2 Teletherapy ☐ If No, Not done due to : 5.2.2.1 Unavailability 1=yes☐ 2=No☐ 5.2.2.2 High Cost 1=yes☐ 2=No☐ 5.2.2.3 Not Relevant ☐		
5.3	**Specific Target therapy/Immunotherapy** 1=yes☐ 2=No ☐ If yes, Then which drugs are given		
	5.3.1 Rituximab ☐	Govt supply=1 ☐	Private fund=2☐
	5.3.2 Imatinib ☐	Govt supply=1 ☐	Private fund=2☐
	5.3.3 Azacytidine ☐	Govt supply=1 ☐	Private fund=2☐
	5.3.4 Venetoclox ☐	Govt supply=1 ☐	Private fund=2☐
	5.3.5 Brentuximab Vedotin	Govt supply=1 ☐	Private fund=2☐
	5.3.6 Midostaurin	Govt supply=1 ☐	Private fund=2☐
	If No, Not done due to : 5.3.2.1 Unavailability 1=yes☐ 2=No☐ 5.3.2.2 High Cost 1=yes☐ 2=No☐ 5.3.2.3 Not Relevant ☐		

6 Availability of Specific Treatment Facility:

6.1 Chemotherapy

6.1.1	Available in Chattogram	1=yes☐ 2 No=☐ If yes, Drugs are supplied to the patient… 6.1.1.1 Entirely At Chittagong Medical College 1=yes ☐ 2=No ☐ 6.1.1.2 Entirely At Private Health Care Facility 1=yes ☐ 2=No ☐ 6.1.1.3 Partly from Government supply and partly from private source 1=yes☐ 2=No☐

6.1.2	Available in Dhaka	1=yes ☐ 2=No ☐
6.1.3	Unavailable in Bangladesh	1=yes ☐ 2=No ☐
6.1.4	Not Given to Patient	☐

6.2 Radiotherapy

6.2.1	Available in Chattogram	6.2.1.1 At Chittagong Medical College 1=yes☐ 2=No☐ 6.2.1.2 At Private Health Care Facility 1=yes☐ 2=No☐
6.2.2	Available in Dhaka	1=yes☐ 2=No☐
6.2.3	Unavailable in Bangladesh	1=yes☐ 2=No☐
6.2.4	Not Given to Patient	☐

6.3 Specific Target Therapy/ Immunotherapy

6.3.1	Available in Chattogram	1=yes☐ 2 No=☐ If yes, Drugs are supplied to the patient… 6.3.1.1 Entirely At Chittagong Medical College 1=yes ☐ 2=No ☐ 6.3.1.2 Entirely At Private Health Care Facility 1=yes ☐ 2=No ☐ 6.3.1.3 Partly from Government supply and partly from private source 1=yes☐ 2=No☐
6.3.2	Available in Dhaka	1=yes☐ 2=No☐
6.3.3	Unavailable in Bangladesh	1=yes☐ 2=No☐
6.3.4	Not Given to Patient	☐

7 Affordability of investigations

7.1 CBC with ESR, PBF

7.1.1	Do you feel that the current cost of this investigation related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐

7.2 Bone Marrow Study

7.2.1	Do you feel that the current cost of this investigation related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐

7.3 Flow Cytometry

7.3.1	Do you feel that the current cost of this investigation related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐

7.4 Plasma Protein Electrophoresis

7.4.1	Do you feel that the current cost of this investigation related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐

7.5 Lymph node Biopsy

7.5.1	Do you feel that the current cost of this investigation related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐

7.6 Cytogenetic Study

7.6.1	Do you feel that the current cost of this investigation related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐

7.7 Immunohistochemistry

7.7.1	Do you feel that the current cost of this investigation related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐

8 Affordability of Specific Treatment Facility:

8.1Chemotherapy

8.1.1	Do you feel that the current cost of this treatment related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐

8.2Radiotherapy

8.2.1	Do you feel that the current cost of this treatment related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐

8.3Specific Target Therapy/ Immunotherapy

8.3.1	Do you feel that the current cost of this treatment related to Hematological Malignancies is affordable?	1=yes☐ 2=No☐ Not Applicable ☐