Introduction

Myeloproliferative disorders are group of haematological malignancies begin in the bone marrow and may cause a greater than normal number of stem cells to develop into one or more types of blood cells. The disorders usually get worse slowly as the number of extra blood cells increase. Chronic myeloproliferative disorders sometimes become acute leukaemia, in which too many abnormal white blood cells are made ***(Peter et al., 2006).***

Classification of myeloproliferative disorders:

Several classification systems adopted for MPD started by Dameshak in 1951 which include:

- Chronic myeloid leukemia.
- Polycythemia vera.
- Essential thrombocytosis.
- Myelofibrosis.
- Erythroleukemia. ***(Dameshak, 1951)***

According to the French-American-British (FAB) classification, chronic myeloproliferative diseases consist of 4 diseases: chronic myelogenous leukemia (CML); polycythemia vera (PV); essential thrombocythemia (ET); and agnogenic myeloid metaplasia (AMM), which is also known as myelofibrosis (MF). In 2002, the World Health Organization (WHO) proposed an alternate classification scheme for these diseases, adding chronic neutrophilic leukemia (CNL) and chronic eosinophilic leukemia (CEL)/hypereosinophilic syndrome (HES).

A related disorder, systemic mastocytosis (SM), has many features in common with the myeloproliferative diseases and is considered by some authors to belong to this group. In some patients, conditions overlap, and clear categorization may be difficult. Myeloproliferative disease may evolve into one of the other myeloproliferative conditions, transform to acute leukemia, or both ***(Ross and Gilliland, 2008).***

Table (1): Comparison of FAB and WHO Classifications of Chronic Myeloproliferative Diseases.

FAB	WHO, 2002
Chronic myelogenous leukemia	Chronic myelogenous leukemia
Polycythemia vera	Polycythemia vera
Essential thrombocythemia	Essential thrombocythemia
Agnogenic myeloid metaplasia/myelofibrosis	Chronic idiopathic myelofibrosis which previously called agnogenic myeloid metaplasia/myelofibrosis
	Chronic neutrophilic leukemia
	Chronic eosinophilic leukemia/hypereosinophilic syndrome

Recent classification of myeloproliferative disorders released in 2008 by WHO:

Main Categories include: Classic MPD, Atypical MPD

Classic MPD include BCR-ABL-positive Chronic myeloid leukemia (CML) and BCR-ABL-negative which include: Polycythemia vera (~100% JAK2V617F +ve), Essential thrombocythemia (~50% JAK2V617F +ve) and Myelofibrosis (~50% JAK2V617F +ve).

Atypical MPD which include: Chronic myelomonocytic leukemia, Juvenile myelomonocytic leukemia (frequent PTP11, NF1, and RAS mutations +ve), Chronic neutrophilic leukemia (~20% JAK2V617F), Chronic eosinophilic leukemia/eosinophilic MPD which include: PDGFRA-rearranged (e.g., FIP1L1-PDGFRA), PDGFRB-rearranged (e.g., TEL/ETV6-PDGFRB), FGFR1-rearranged (e.g., ZNF198/FIM/RAMP-FGFR1/a.k.a. 8p11 myelo-proliferative syndrome), Molecularly undefined. Hypereosinophilia syndrome, Chronic basophilic leukemia, Systemic mastocytosis which include: PDGFRA-rearranged (e.g., FIP1L1-PDGFRA), KIT-mutated (e.g., KITD816V), Molecularly undefined. Unclassified MPD (~20% JAK2V617F) which include: Mixed/overlap myelodysplastic syndrome/MPD, CML-like but BCR-ABL-negative ***(Cristina and Guang, 2008).***

Pathophysiology

Data from G-6-PD studies, cytogenetic analyses, and molecular methods have established the clonal origin of myeloproliferative diseases; this clonality potentially occurs at different stem cell levels. An attribute common to these disorders appears to be an acquired activating mutation in the gene coding for various tyrosine kinases. In chronic myelogenous leukemia, the tyrosine kinase activity of the *bcr-abl* hybrid gene is increased. In polycythemia vera, essential thrombocythemia, and myelofibrosis, the prevalent genetic lesion appears to be a valine to phenylalanine substitution at amino acid position 617 (V617F) within the Janus kinase 2 (*JAK2*) gene, this produces hypersensitivity to erythropoietin.

At least in myelofibrosis patients the leukemic transformation is probably not related to JAK-2 (V617F) mutation status. Systemic mastocytosis has been linked with the D816 mutation of the *KIT* gene. The *FIP1L1-PDGFR* mutation has been identified in a subgroup of people with systemic mastocytosis with eosinophilia (SM-eos) ***(Norwood, 2008).***

Epidemiological study:

1) Frequency:

Approximately 6-9/100000 are diagnosed as myelo-proliferative disease cases every year ***(Ross and Gilliland, 2008).***

2) Sex:

Males had higher prevalence than females with female-to-male ratio is 1:1.4 ***(Adewale and Dewald, 2003).***

3) Race:

Chronic myelogenous leukemia appears to affect all races with approximately equal frequency. The incidences of polycythemia vera, essential thrombocythemia, and myelofibrosis were ten fold higher among Ashkenazi Jews in northern Israel than in persons of Arabic descent in the region ***(Adewale and Dewald, 2003).***

4) Age:

Most cases encountered in clinical practice are in patients aged 40-60 years. Myeloproliferative diseases are uncommon in people younger than 20 years and are rare in childhood ***(Ross and Gilliland, 2008).***

Clinical picture:

History

- Easy fatigability.
- Anorexia, weight loss.
- Abdominal discomfort and early satiety secondary to splenomegaly is more common in chronic myelogenous leukemia and myelofibrosis.
- Easy bruising, bleeding, and/or symptoms of thrombosis.
- Swollen, painful joint(s) secondary to gouty arthritis secondary to hyperuricemia.
- Priapism, tinnitus, or stupor from leukostasis.
- Left upper quadrant and left shoulder pain as a consequence of splenic infarction and perisplenitis.

In many patients, abnormal blood counts are noted on a blood test performed for other reasons ***(Norwood, 2008)***.

Physical:

- Pallor, except in patients with polycythemia vera.
- Plethora secondary to polycythemia.
- Petechiae and/or ecchymosis.
- Palpable spleen and/or liver.

Occasionally, syndrome of fever accompanied by painful maculopapular violaceous lesions on trunk, arms, legs, and

face, which is called acute febrile neutrophilic dermatosis or Sweet syndrome ***(Norwood, 2008)***.

Lab Studies:

Will be discussed later with each disease.

We will discuss in this essay MPDs diagnostic tools and new therapeutic strategies.

Chronic Myelogenous Leukaemia

Chronic myelogenous leukemia (CML) is a pluripotential stem cell disease characterized by anemia, extreme blood granulocytosis and granulocytic immaturity, basophilia, often thrombocytosis, and splenomegaly.

The presence of *BCR/ABL* rearrangement is the hallmark of CML, although this rearrangement has also been described in other diseases. It is considered diagnostic when present in a patient with clinical manifestations of CML ***(Lee, 2000).***

Incidence

CML accounts for approximately 15 % of all cases of leukemia or approximately 4600 new cases per year. The disease occurs more often in men than in women (ratio 1.5:1.0), NCI's Surveillance, Epidemiology, and End Results (SEER) Cancer Statistics Review, it is estimated that 4830 men and women (2800 men and 2030 women) will be diagnosed with CML. It typically affects middle-aged individuals in the fourth and fifth decades of life but The median age at presentation is around 66 years, Younger patients aged 20-29 years may be affected and may present with a more aggressive form, such as in accelerated phase or blast crisis.

The lowest incidence is in Sweden and China (approximately 0.7 per 100,000 persons), and the highest incidence is in Switzerland and the United States approximately 1.7 per 100,000 persons. Internationally, Increased incidence

was reported among individuals exposed to radiation in Nagasaki and Hiroshima after the dropping of the atomic bomb (***Lichtman et al., 2007a).***

Risk factors

The initiating factor of CML is still unknown, Exposure to very high doses of ionizing radiation can increase the occurrence of CML above the expected frequency in comparable populations such as atomic bomb survivors were exposed to in (World War II), Chemical leukemogens, such as benzene and alkylating agents have not been identified as causative agents of CML, although they are well established to produce a dose-dependent increase in acute myelogenous leukemia. DNA topoisomerase II inhibitors may be an exception, as they have been found to have a propensity to induce t(9;22)-positive leukemia. Chronic myeloid leukaemia, like other cancers is not infectious and can't be passed on to other people. It is not caused by an inherited faulty gene, The Philadelphia chromosome is not an inherited fault that can be passed on from one generation to the next. However, it causes the production of a particular enzyme called tyrosine kinase, which leads to the development of CML. Research has shown that exposure to electromagnetic fields, living near high-voltage electricity cables, and household radon do not increase a person's risk of developing chronic myeloid leukaemia ***(Lichtman et al., 2007a).***

Mortality/morbidity

Generally, 3 phases of the disease are recognized: chronic, accelerated and blastic crisis. The general course of the disease is characterized by an eventual evolution to a refractory form of acute myelogenous or, occasionally, lymphoblastic leukemia. The median survival of patients using older forms of therapy was 3-5 years in chronic phase and 1 year in blastic crisis ***(Lee, 2000).***

Origin from a stem cell clone

CML results from the malignant transformation of a single stem cell. The disease is acquired (somatic mutation). The origin of CML from a single hematopoietic stem cell is supported by the following lines of evidence:

1. Involvement of erythropoiesis, neutrophilopoiesis, eosinophilopoiesis, basophilopoiesis, monocytopoiesis, and thrombopoiesis in chronic phase CML.
2. Presence of the Ph chromosome in erythroblasts; neutrophilic, eosinophilic, and basophilic granulocytes; macrophages; and megakaryocytes.
3. Presence of a single glucose-6-phosphate dehydrogenase isoenzyme in red cells, neutrophils, eosinophils, basophils, monocytes, and platelets, but not in fibroblasts or other somatic cells in women with CML who are heterozygotes for isoenzymes A and B.
4. Presence of the Ph translocation only on a structurally anomalous chromosome 9 or 22 of each chromosome pair in

every cell analyzed in occasional patients with a structurally dissimilar 9 or 22 chromosome within the pair.

5. Presence of the Ph chromosome in one but not the other cell lineage of patients who are a mosaic for sex chromosomes, as in Turner syndrome (45X/46XX) and Klinefelter syndrome (46XY/47XXY).
6. Molecular studies showing variation in the breakpoint of chromosome 22 among different patients with CML but precisely the same breakpoint among cells within a single patient with CML.
7. Combined DNA hybridization-methylation analysis of women who have restriction fragment length polymorphisms at the X-linked locus for hypoxanthine phosphoribosyl-transferase (HPRT), which enables distinction of the two alleles of the HPRT gene in heterozygous females, coupled with methylation-sensitive restriction enzyme cleavage patterns, which permits delineation of whether cells contain either the maternally derived or the paternally derived copy of the gene ***(Lichtman et al., 2007a).***

Molecular Genetics of Chronic Myelogenous Leukemia

The Philadelphia (Ph) chromosome was originally detected by workers in Philadelphia as an abnormally short G-group chromosome in analysis of bone marrow metaphases from CML patients, it has the distinction of being the first genetic abnormality to be associated with a human cancer. Subsequently, advances in chromosome banding techniques

demonstrated that the Ph chromosome was the result of a balanced translocation between chromosomes 9 and 22, denoted t(9;22) (q34.1;q11.21), where the derivative chromosome 22 is significantly smaller. The Ph chromosome is present in hematopoietic cells from patients with CML but not in non hematopoietic tissues, including bone marrow fibroblasts (***Tefferi and Gilliland, 2006).***

The structure of the Ph chromosome was established by initial studies demonstrating translocation of the c-ABL gene on chromosome 9q34 to the Ph chromosome and the subsequent discovery of breakpoints near the 5' end of the c-ABL gene in leukemic cells from patients with CML. The c-ABL gene had been previously identified as the cellular homologue of the transforming gene of Abelson murine leukemia virus an acutely transforming retrovirus that induces B-lymphoid leukemia in mice. The c-ABL gene on chromosome 9 has eleven exons with two alternative 5' first exons, and a very large first intron of over 250 kilobases (kb). The c-ABL gene encodes a non-receptor protein-tyrosine kinase, c-Abl) ***(Huntly et al., 2003).***

DNA sequences immediately 5' to the ABL gene on the Ph chromosome were derived from chromosome 22 sequences, and DNA probes from a small region on chromosome 22 detected genomic rearrangements by Southern analysis of genomic DNA in virtually all CML samples. This locus on chromosome 22 was named the breakpoint cluster region, or bcr. Subsequent characterization of this locus demonstrated that the bcr region was in the middle of a large protein-coding gene

of 25 exons now called the BCR gene. Five small exons were initially identified in the bcr region and denoted exons b1 through b5; They are now known to be exons 12 through 16 of BCR. The BCR gene product is a 160 kilodalton cytoplasmic phosphoprotein denoted Bcr ***(Susan, 2007).***

Depending upon the location of the breakpoint within the bcr region, the consequence of the t(9;22) translocation in CML is to fuse the first 13 or 14 exons of the BCR gene upstream of the second exon of the c-ABL gene, with the breakpoint on chromosome 9 falling in the large first intron region. The two alternative fusion genes are traditionally described according to the original bcr exon nomenclature as b2a2 and b3a2 fusions. Transcription of the fusion gene followed by RNA splicing leads to the generation of a novel 8.5 kb fusion BCR-ABL mRNA that encodes a fusion protein of 210 kilodaltons designated p210BCR-ABL or p210 Bcr-Abl The protein product of the b3a2 fusion is 25 amino acids longer than the b2a2 product due to the inclusion of the b3 exon ***(Toru et al., 2008).***

The Bcr-Abl fusion protein contains the entire tyrosine kinase catalytic domain from c-Abl and has constitutively increased tyrosine kinase activity relative to c-Abl. Bcr-Abl-expressing cells have increased levels of tyrosine-phosphorylated proteins and aberrant activation of multiple cellular signaling pathways, some of which have been implicated directly in the pathogenesis of CML ***(Jerald, 2007).***

A central role for the Bcr-Abl tyrosine kinase in the pathogenesis of CML has been established by the therapeutic efficacy of small molecule inhibitors of the Abl tyrosine kinase. Specific Bcr-Abl tyrosine kinase inhibitors decrease cellular proliferation of Bcr-Abl-expressing cells in vitro by more than 90 % but have no effect on normal cells. (Toru et al., 2008).

The reciprocal translocation product on the derivative chromosome 9 [der(9)] is an ABL-BCR fusion gene that could generate an Abl-Bcr fusion protein. However, the inconsistent expression of the ABL-BCR gene in patients with CML, along with survival data in ABL-BCR negative patients, suggest that this product does not play a major role in the pathogenesis of this disorder although der(9) deletions may confer a worse prognosis ***(Reid et al., 2003).***

Several years after the description of BCR-ABL in patients with CML, it was discovered that the majority of pediatric and adult patients with Ph-positive B-ALL had chromosome 22 breakpoints within the large first intron of the BCR gene rather than the classic bcr region, leading to formation of a smaller BCR-ABL fusion gene consisting of the first exon of BCR fused upstream of ABL exon 2 (e1a2). The bcr region in CML was renamed the major or M-bcr, while the first intron breakpoints were designated the minor or m-bcr. This fusion generates a protein of 190 kD in size, p190BCR-ABL (also referred to as p185 in some references) that contains BCR first exon-encoded sequence fused to the same amount of Abl found in p210. A third minor bcr region on chromosome 22

resulting in fusion of BCR exon 19 to ABL exon 2 (e19a2) has been described in several patients, leading to generation of a p230 form of Bcr-Abl, (In addition to these more common variant forms, rare patients with fusion of BCR exon 1 or exon b2 to ABL exon 3 (e1a3 and b2a3) and BCR exon 6 to ABL exon 2 (e6a2) have been described, where both fusion mRNAs are predicted to have an intact translational reading frame ***(Toru et al., 2008).***

Type of BCR-ABL fusion may influence the clinical manifestations, The various types of BCR-ABL fusion may be associated with distinct leukemia phenotypes; this is most apparent with p190BCR-ABL. Determining whether different forms of BCR-ABL are associated with distinct leukemic phenotypes and clinical outcomes is important, since it is plausible that optimum therapy (eg, response to interferon alfa, imatinib mesylate, or donor leukocyte infusions post-transplant) might be in part determined by the BCR-ABL genotype. Experiments in tissue culture and animal models of Ph-positive leukemia suggest that different forms of BCR-ABL do have distinct leukemogenic properties ***(Ravandi et al., 1999).***

P190BCR-ABL is found in about 80 % of childhood and 50 % of adult Ph-positive B-ALL, and is infrequently observed in CML ***(Ren, 2005).***

Several patients with e19/a2 BCR-ABL fusions and the p230 form of the Bcr-Abl protein have been described with a disorder similar to classic CML but with mild clinical

symptoms, these include lower peripheral blood leukocyte counts consisting principally of neutrophils, thrombocytosis, less severe splenomegaly, and delayed or absent transformation to blast crisis. It was proposed that patients with the e19/a2 fusion comprise a distinct clinical entity called neutrophilic CML or chronic neutrophilic leukemia, with a much more benign clinical course than that associated with traditional b2a2 or b3a2 p210BCR-ABL fusions However, other patients with the e19/a2 fusion appear to have typical CML ***(Ren, 2005).***

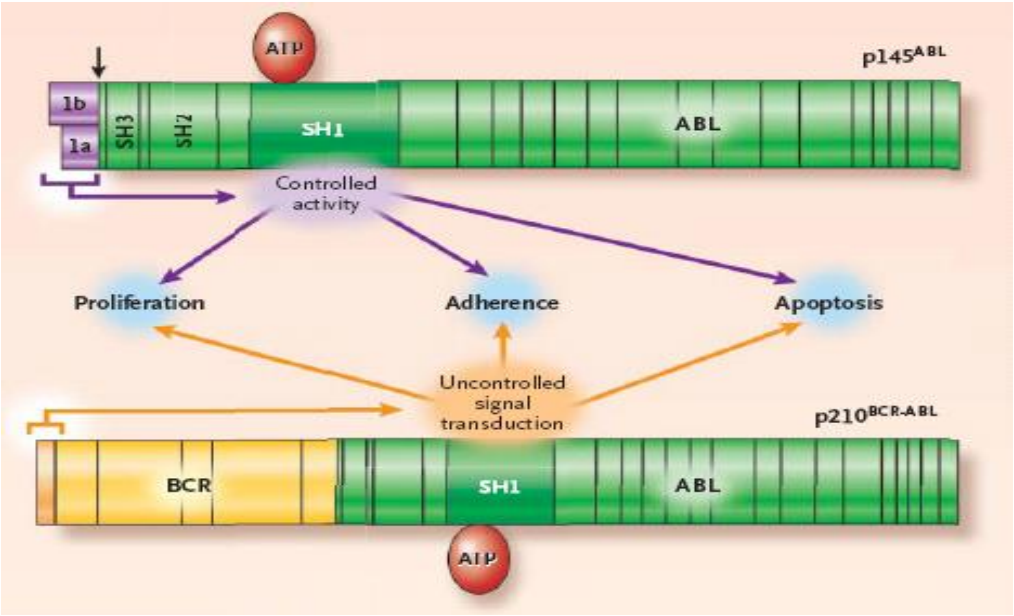

Figure (1): Illustration of BCR-ABL protein effect.