Team Medicine

5.Leukemia

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■ Slides ■ Doctors notes ■ Additional

431 Teams

Definition:
Leukemias are a group of cancers of the blood/ bone marrow and are characterized by an abnormal proliferation (production by multiplication) of blood cells, usually white blood cells (leukocytes).
Leukemia is a broad term covering a spectrum of diseases. Any of various acute or chronic neoplastic diseases of the bone marrow in which unrestrained proliferation of white blood cells occurs and which is usually accompanied by anemia and thrombocytopenia

Because of the infiltration of leukemic cells in the BM causing BM failure, resulting in abnormal growth and proliferation of normal cells which give rise to anemia and thrombocytopenia

Classification:

According to the duration:	**According to the cell of origin:**
Acute leukemias **Acute lymphoblastic leukemia (ALL)** **Acute myelogenous leukemia (AML)** (also "myeloid" or "nonlymphocytic") Chronic leukemias **Chronic lymphocytic leukemia (CLL)** **Chronic myeloid leukemia (CML)**	• Lymphoid cells **ALL** - lymphoblasts **CLL** – mature appearing ymphocytes • Myeloid cells **AML** – myeloblasts **CML** – mature appearing neutrophils

(Within these main categories, there are typically several subcategories)

blastic➔acute or primitive leukemia

BLAST is hallmark of ACUTEleukemias; maturation stops at the blast level

Myeloidvs Lymphoid:

Myeloid	**Lymphoid**
• Any disease that arises from the myeloid elements (white cell, red cell, platelets) is a myeloid disease➔AML, CML	• Any disease that arises from the lymphoid elements is a lymphoid disease➔ALL, CLL

Acute vs. chronic leukemia

Acute leukemias:	**Chronic leukemias:**
• Young, immature, ***blast***cells in the bone marrow(and often blood) • More fulminant presentation • More aggressive course	• Accumulation of mature, ***differentiated***cells • Often subclinical or incidental presentation • In general, more indolent (slow) course • Frequently splenomegaly • Mature appearing cells in the B. marrow

	and blood
• Presence of blast cells in peripheral blood➔it is most commonly ACUTE leukemia • Normal amount of blast cells present in BM is less than <5%, if more than >20% it indicates acute leukemia of BM. • Acute leukemia grows rapidly so patients complain early (short history 1-3 months). • Patients Present with series illnesses such as anemia, bleeding, infection, fever, septicemia, pneumonia, etc.	• Blast cell may be present but less than 5% • Incidentally finding (a routine CBC done and they found out high WBC➔ chronic leukemia). • Chronic leukemia patients are less prone to infection compared to acute leukemia patients because they have less functioning neutrophils, which protect against bacterial and fungal infections.

On a CBC, if you see:

- Predominance of blasts(immature) in blood

consider an acute leukemia

- Leukocytosis with mature*lymphocytosis*

consider CLL

- Leukocytosis with matureneutrophilia

consider CML

ACUTE LEUKEMIA:

Definition:

- Disorders with clonal expansion of precursor cells (myeloid or lymphoid) with reduced capacity to differentiate.
  - Maturation arrest at the blast stage
  - Bone marrow infiltration by blast cells causing suppression of normal precursors (bone marrow failure)
- **Malignancies of immature hematopeotic cells.(> 20% blast cells in the bone marrow).**

Most of the cancers are clonal= arising from single cell
BM failure CBC: low RBC (anemia), low platelet (thrombocytopenia), low WBC (leukopenia) that will give rise to infections

Types: **Acute Myeloid Leukaemia (AML)**
Acute Lymphoblastic leukemia (ALL)

Groups: **Childhood (< 15)** **> 80% ALL**
Adult (> 15) **> 80% AML**
Elderly (> 60 years)

The most common type of leukemia in children is ALL, more than 80%
It is curable in children but have poor prognosis in adults
AML is common in adult

Epidemiology of AML:

- **Incidence – 2.7 per 100,000**
  - **12.6 per 100,000 in those over 65 yrs**
  - **Median age of presentation: 67 yrs**

- **More prevalent in:**
  - **Males**
  - **European descent**
  - **Hispanic/Latino background (promyelocytic leukemia, AML-M3)**

Acute promyelocytic leukemia (APL) is a subtype of AML where there is proliferation of promyelocyte (the one after myeloblast), this type is also called M3.

Etiology: most of the time there is no clear cause = de novo
Chemotherapy drugs in cancer patient may induce AML (e.g patient with breast cancer who develops acute leukemia after treatment)

Etiology:

- Mostly unidentifiable cause i.e., de nove (≥80%)
- Drugs & chemicals
- Alkylating agents (Chlorambucil, N mustard, Melphalan)
- Topoisomerase inhibitors (Etoposide)
- Benzene
- Ionizing radiation> in high doses
- Myelodysplastic syndrome>group of disorder usually present with anemia + BM failure + dysplastic changes (dysplasia) in BM so they have high tendency of developing acute leukemia.
- Myeloproliferative disordersmyelo= BM, group of 4 disorders:pylocythemiavera, thrombocytopenia, CML that has tenednecy to develop into acute leukemia, and mastocytosis.

- Genetic disordersdamage of DNA repair mechanisms
- Down's syndrome
  - Bloom syndrome
  - Faconi anemia
  - Wiskott Aldrich syndrome

The myelodysplatstic syndromes (MDS) are hematological medical conditions with ineffective production (or dysplasia) of the myeloid class of blood cells.
Patients with MDS can develop severe anemia and cytopenia (low blood counts) caused by progressive bone marrow failure.
The myeloproliferative diseases (MPDs) are group of diseases of the bone marrow in which excess cells are produced. They are related to, and may evolve into myelodysplastic syndrome and acute myeloid leukemia(AML)
There are four main MPDs:
leukemia
Polycythemia vera
Essential thrombocytopenia
Mastocytosis = presence of too many mast cells

Clinical presentation:

Symptoms:

- Usual 1-3 Month history: MDS – 1yrpatient who develops leukemia on top of myelodysplastic syndrome MDS they may have longer duration.
- (Features of BM failure)
  - Fatigue, malaise, dyspnea (anemia)
  - Bleeding eg after dental procedure, Easy bruisability, Severe epistaxis
  - Fever (infections)
  - Bone Painduring PE when we press over the sternal area or chin of the long bone patient will feel unusual pain.

Signs:

- Pallor
- Hemorrhage from the gums, epistaxis, skin, fundus, GI tract, urinary tract
- Hepato-splenomegaly➔more common in chronic leukemia.
- Enlarged lymph nodes➔commonly seen in CLL.
- Gum (hypertrophy) or skin infiltration (M5)➔subtype of AML.
- Fever (sepsis, pneumonia, peri-rectal abscess)

Differential diagnosis

- Aplastic anemia
- Myelodysplastic syndromes
- Multiple myeloma➔cancer of plasma cells
- Lymphomas
- Severe megaloblastic anemia➔can present with pancytopenia although there will be no increase of the blast cells.
- Leukemoid reaction. ➔Some patients occasionally due to severe stress or infection may develop high WBC count more than 50.000 with some immature cells.(WBC is 50.000 or above with some immature cells)

Diagnosis:

- CBC
  - Anemia
  - Trombocytopenia
  - WBC (**High,** Normal, or Low) ➔WBC is always high in chronic leukemia, but in acute leukemia it may be normal or low because the BM is so infiltrated with blast cells, so cells are tightly packed and can't leave the BM.
- Coagulation Studies (M3-DIC)common sub type of AML
- Biochemical Studies (U/E, LFT)
- Peripheral Blood Smear – blasts in almost all cases. We take a drop of blood>spread it on a slide> stain it>and view it under the microscope. Done before BM biopsy.
- Bone Marrow Examination (>20% blasts)➔diagnostic
- Flow cyometry (Surface immunophenotype of blast cells)➔most important test to diagnose types and subtypes of leukemia.
- Cytogenetics (chromosomal analysis).

- CSF analysis (all ALL patients, some AML)➔usually in lymphoid leukemia.

***The 1st step after the CBC is BM biopsy that will tell us this is acute leukemia not the type, but we don't know whether it is lymphoid or myeloid and their subtypes, which is important in treatment and prognosis.**
***Flow cytometry is a machine like a blood counter and it will tell us the type of the cell and it measures the different antigens since they have different clusters of differentiation (e.g CD19 and 20 are B-cell marker).**
***Chromosomal analysis is very important.**

Diagnostic methods of importance

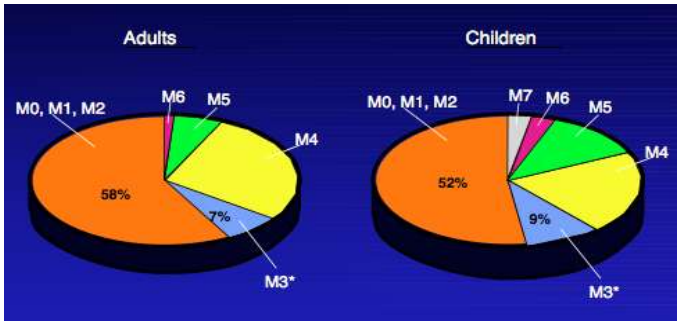
- Bone marrow aspirate &Romanowsky stain (morphology):
  Enumeration of blasts, maturing cells, recognition of dysplasia
- Cytochemistry:
  Myeloperoxidase, Sudan Black B, esterases to determine involved lineages

We still do it but it is not very accurate, the different types of cells (such myeloid or lymphoid) are stained in different colours.

- Immunophenotyping: (the cell markers) > diagnostic technique of choice
  Defines blast cell lineage commitment as myeloid, lymphoid or biphenotypic
- Cytogenetics& molecular studies (FISH, PCR):
  Detects clonal chromosomal abnormalities, including those of prognostic importance

FAB Classification of AML:(French–American–Britishclassification),it is an old classification.

- **MO** undifferentiated acute myeloblastic leukemia (5%)
- **M1**AML with minimal maturation (20%)
- **M2**AML with maturation (30%)
  - t(8;21)
- **M3** Acute promyelocytic leukemia (5%)
  - t(15;17)
- **M4**Acute myelomonocytic leukemia (20%)
- **M4**eos Acute myelomonocytic leukemia with eosinophilia (5%)
  - inv (16**)**
- **M5** Acute monocytic leukemia (10%)
  - t(9;11)
- **M6** Acute erythroid leukemia (3%) arises from RBC precursor
- **M7**Acute megakaryoblastic leukemia (3%) which give rise to platelets



We don't need to know it

WHO Classification of AML(The new classification).

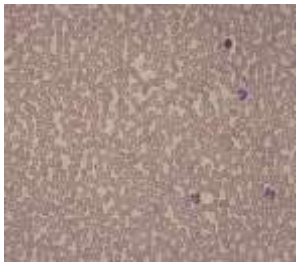
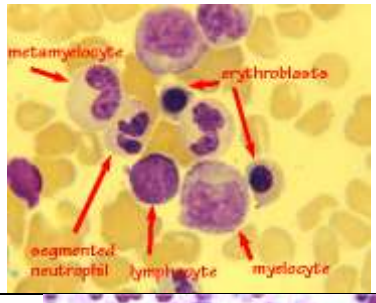
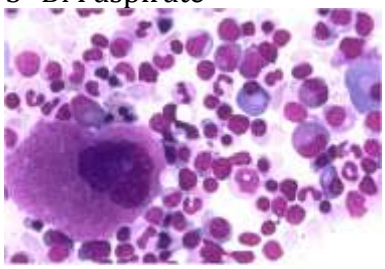
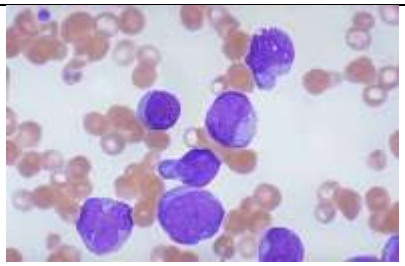
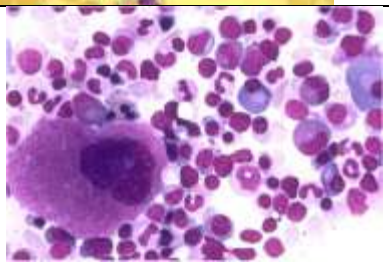
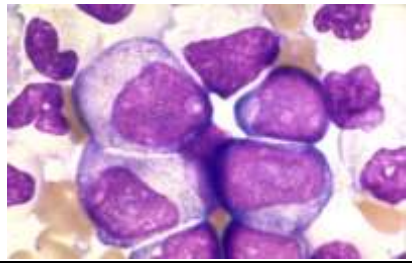
-The exchange of materials between 2 chromosomes is called translocation.
-The first noticed translocation was philadalphia chromosome t(9;22) which presents mainly in CML

- **AML with recurrent genetic abnormalities**
  - t(8;21), t(16), inv(16), chromosome 11 changes
  - t(15;17) as usually seen with AML M3
- **AML with multilineage dysplasia** (more than one abnormal myeloid cell type is involved)those patient who have AML on top of MDS
- **AML related to previous chemotherapy or radiation**
- **AML not otherwise specified**classified based on their appearance (morphology), they don't arise on top of any secondary causes.
  - Undifferentiated AML (MO)they are called biphenotypic it is difficult to tell if it is myeloid or lymphoid
  - AML with minimal maturation (M1)
  - AML with maturation (M2)
  - Acute myelomonocytic leukemia (M4)
  - Acute monocytic leukemia (M5)
  - Acute erythroid leukemia (M6)
  - Acute megakaryoblastic leukemia (M7)
  - Acute basophilic leukemia (M8)
  - Acute panmyelosis with fibrosis
  - Myeloid sarcoma (also known as granulocytic sarcoma or chloroma)
- **Undifferentiated or biphenotypic acute leukemias**(leukemias that have both lymphocytic and myeloid features. Sometimes called ALL with myeloid markers, AML with lymphoid markers, or mixed lymphoid lineage leukemias).

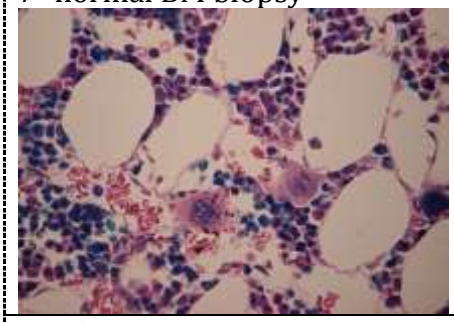
Just read them the doctor didn't go through them

Blood film:

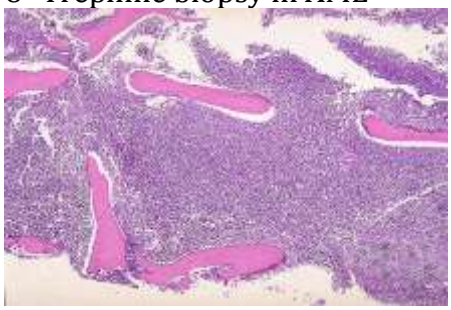
The doctor skipped 10-16

1- normal blood film 	2- normal BM cells 	3- BM aspirate 
4- 	5- 	6- AML 

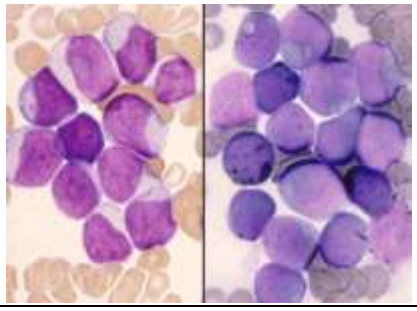
7- normal BM biopsy



8- Trephine biopsy in AML

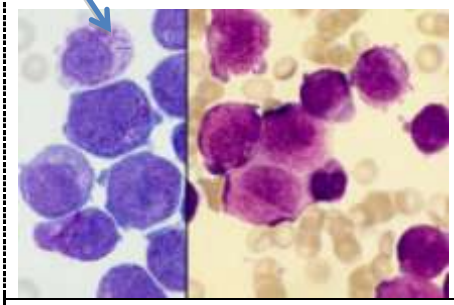


9- AML

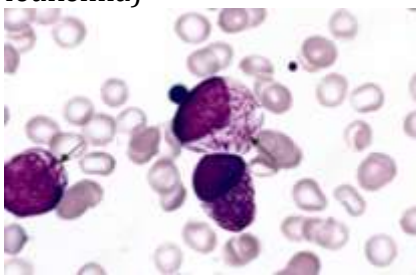


10- AML

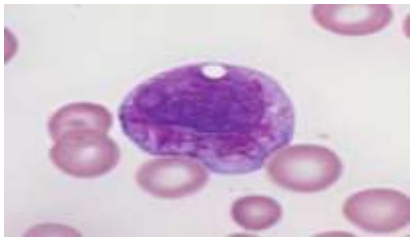
Auer rods is a hallmark of AML



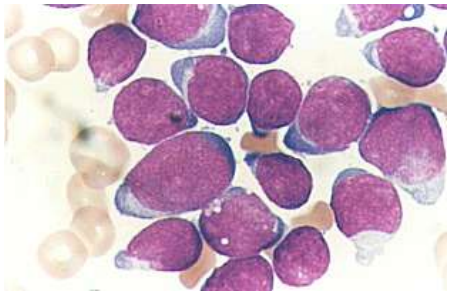
11- AML-M3 (Acute promyelocytic leukemia)



12-

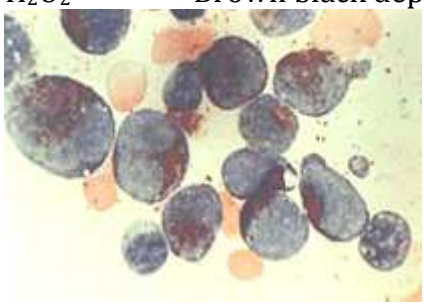


13- ALL



14- Myeloperoxidase (MPO)

p-Phenylenediamine + Catecol + H_2O_2 —MPO—> Brown black deposits



15- Chloracetate (Specific) Esterase Myeloid Cell Line

Naphthol-ASD-chloracetate —CAE—> Free naphthol compounds

+ Stable diazonium salt (eg, Fast Corinth)

>Red deposit

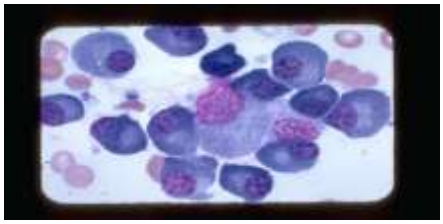
16- Non-Specific Esterase Monocytic Line

aNaphthyl acetate —ANAE—> Free naphthyl compounds

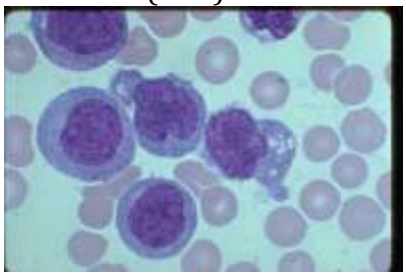
+Stable diazonium salt (eg, Fast blue RR)

——>**Brown deposits**

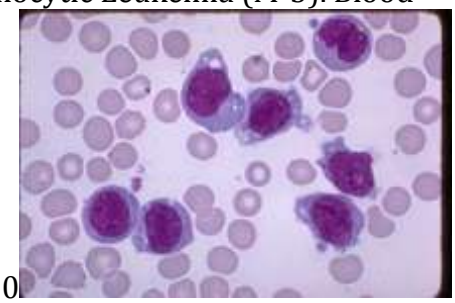
17- Clump of Plasma Cells most of which are small with a deep basophilic blue cytoplasm. Two cells in the center are partially smudged and show a paler cytoplasm and less dense and redder staining nuclear chromatin. Acute myelomonocytic (M-4) leukemia. Marrow - 100X



18- Four Immature Monocytes. Two of the cells are shedding large Cytoplasmic Fragments which can resemble a Megathrombocyte and/or Platelets. Acute Monocytic Leukemia (M-5). Blood - 100X



19- Cytoplasmic Fragments from Leukemic Monocytes that resemble Platelets. Two fragments (top right and lower left) are probably valid platelets. Seven immature monocytes. Acute Monocytic Leukemia (M-5). Blood –

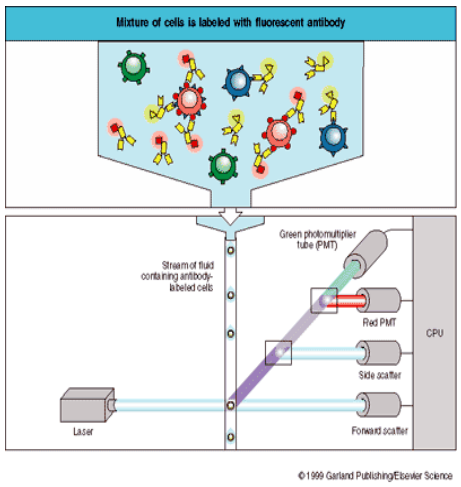


x100

Immunophenotyping:

Immunophenotyping detects the presence or absence of white blood cell (WBC) antigens.
Most of the antigens that immunophenotyping detects are identified by a CD (clusters of differentiation or cluster designation) number, such as: CD1a, CD2, CD3, CD4, CD8, CD13, CD19, CD20, CD33, CD61, etc.

- Flow cytometry is performed by processing blood or marrow samples and adding specific antibodies tagged with fluorescent markers.
- These antibodies attach to corresponding antigens on the WBCs when the antigens are present, and are analyzed.
- Results are then graphed and compared to "normal" results and to patterns that are known to be associated with different leukemias

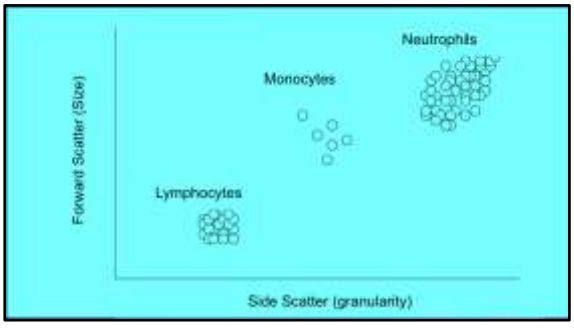


Flow cytometry:

- First labeled with fluorescent dyes.
- Forced through a nozzle in a single-cell stream passing through a laser beam
- The laser is focused to a known wavelength
- Excitation of a specific fluorochrome
- Photo-multiplier tubes detect the scattering of light and emission from the fluorescent dye

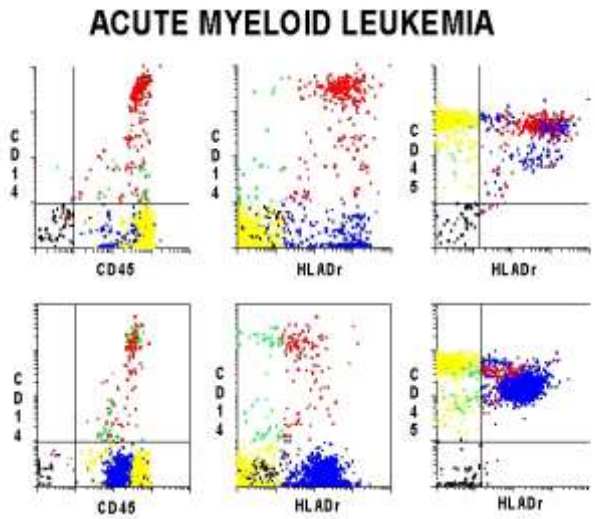
How does it work?
http://www.unsolvedmysteries.oregonstate.edu/flow_06

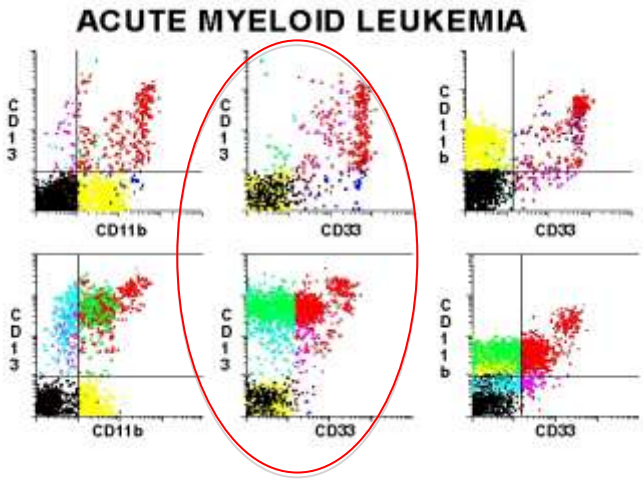
Flow cytometryForward and Side Scatter



This is how the graph looks like (each group of cells are gathered together).

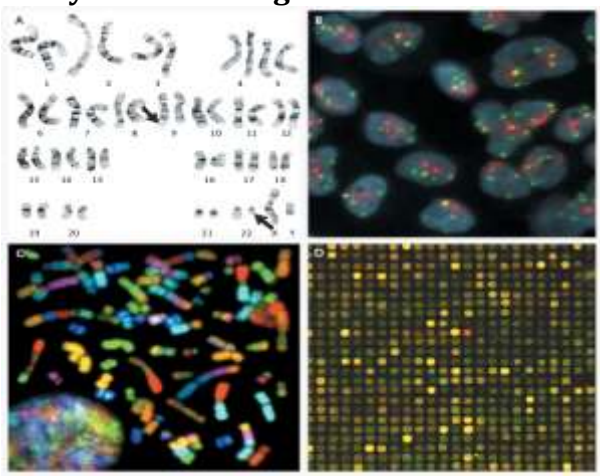
Immunophenotyping in leukemias:

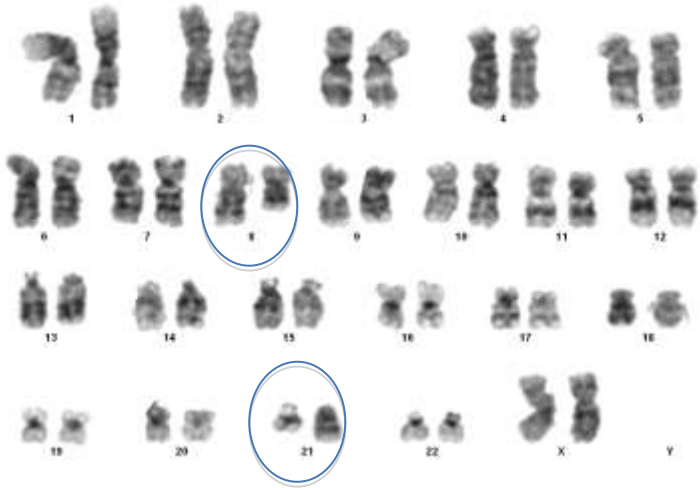




-These are myeloid markers.
-Presence of abnormal cells

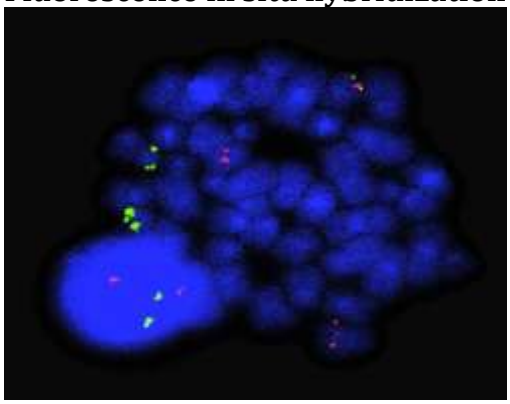
Application of Cytogenetic, FISH and Microarray Analysis in the Diagnosis of Leukemia





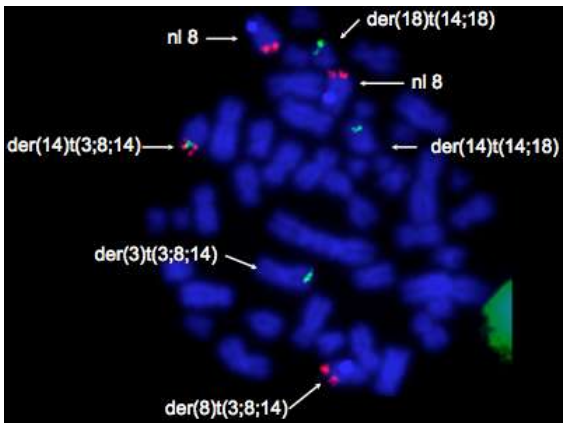
ISCN:46,XX,t(8;21)(q22;q22)[19]/46,XX[1]

Fluorescence in situ hybridization (FISH)



45,X,Y,t(8;20;21)(q22;p13;q22),del(11)(q21q25)
FISH with AML1/ETO –DF probe:
76% of cells show one fusion, two red and two green signals
Three way translocation of t(8;21)(q22;q22)

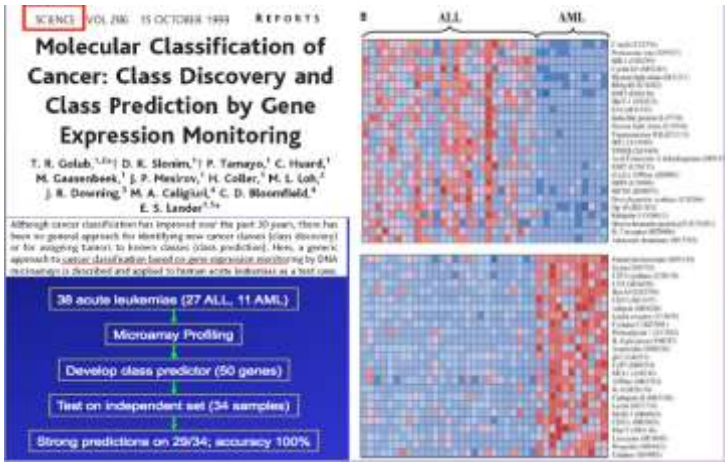
FISH
One group of chromosomes should appear either red or green "same color". If tow colors appear it means there is translocation.

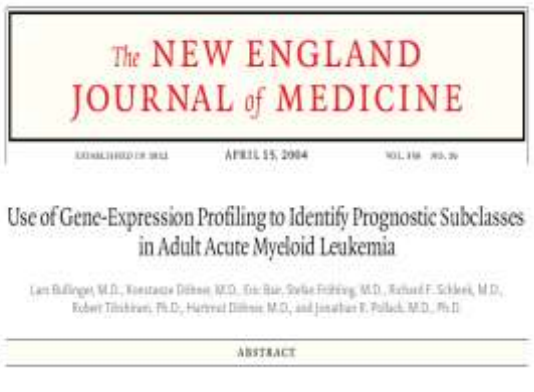


Karyotype is a test to identify and evaluate the size, shape, and number of chromosomes in a sample of body cells.

Application of genomic array analysis in leukemias (microarrays)

Microarray = measuring or mapping different genes in our cells
It will be used in the clinical practice soon





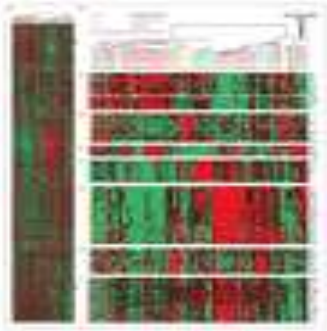
- Some publications

Use of gene-expression profiling to identify prognostic subclasses in adult acute myeloid leukemia

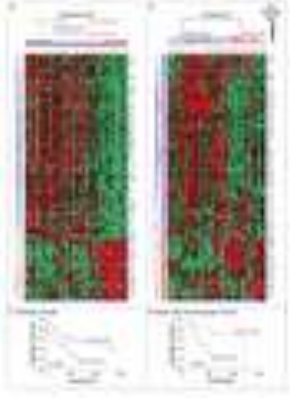
- N Engl J Med. 2004 Apr 15;350(16):1605-16.
- Bullinger L, Döhner K, Bair E, Fröhling S, Schlenk RF, Tibshirani R, Döhner H, Pollack JR.
- Source
- Department of Pathology, Stanford University, Stanford, Calif, USA.
- Abstract
- RESULTS:
- Unsupervised analysis identified new molecular subtypes of AML, including two prognostically relevant subgroups in AML with a normal karyotype. Using the supervised learning algorithm, we constructed an optimal 133-gene clinical-outcome predictor, which accurately predicted overall survival among patients in the independent validation group (P=0.006), including the subgroup of patients with AML with a normal karyotype (P=0.046). In multivariate analysis, the gene-expression predictor was a strong independent prognostic factor (odds ratio, 8.8; 95 percent confidence interval, 2.6 to 29.3; P<0.001).
- CONCLUSIONS: The use of gene-expression profiling improves the molecular classification of adult AML.

This is how it looks like, we see tow patterns one is normal and the other one is abnormal and this how we predict the outcome

Hierarchical Cluster Analysis of Diagnostic AML Samples



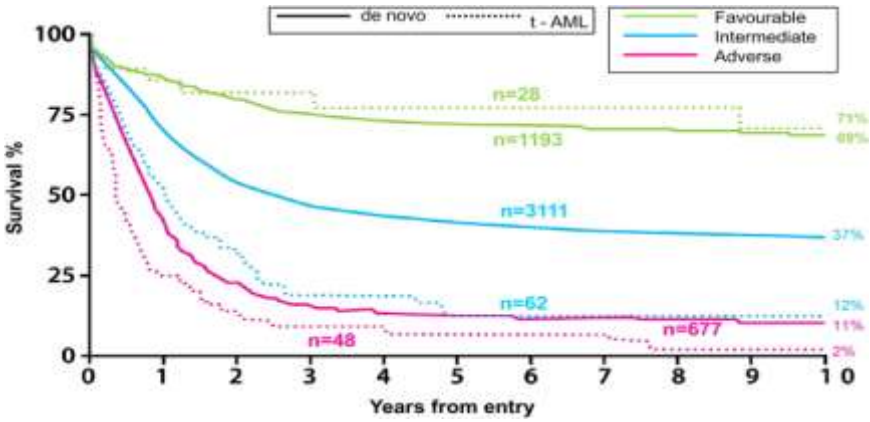
Outcome prediction



Acute Myeloid Leukaemia (AML)

Prognostic factors:

- **Age**
  Above the age of 50 years the complete remission rate falls progressively
- **Cytogenetics**
  Three risk groups defined
  - Good risk: patients with t(8;21), t(15;17) and inv/t(16)
  - Intermediate risk: Normal, +8, +21, +22, 7q-, 9q-, abnormal 11q23, all other
  - Poor risk: patients with -7, -5, 5q-, abnormal 3q and complex karyotypes
- **Treatment response**
  - Patients with >20% blasts in the marrow after first course of treatment have short remissions (if achieved) and poor overall survival
- **Secondary AML**
  - Patients with AML following chemotherapy or myelodysplasia respond poorly
- **Trilineagemyelodysplasia**
  - Patients with trilineagemyelodysplasia have a lower remission rate



The lower the curve the lesser survival chance
Green: shows those with good risk chromosomes, the chance of survival for 10 years is 70-80%
Pink: those with bad prognostic chromosomes have poor survival rate 10%
Blue: is the intermediate
And this is how we classify them into different groups

Treatment and prognosis of AML

- **Intensive chemotherapy**
  - Patients < 55 years old: 80% remissions
  - Patients > 55 years old: progressive reduction in remission rate
- **Bone marrow (stem cell) transplantation**
  - Autologous and allogeneic transplants reduce the relapse rate
- Importance of cytogenetics for prognosis in children and adults < 55 years old
- Good risk cytogenetic group
  - 91% remissions, 65% five year survival

In young patients who have poor risk chromosomes they need BM transplant.
The donated bone marrow will fight the leukemia and this is called graft versus host disease (GVHD).

Autologous➔ auto means self. stem cells are removed from the same patient.

Allogeneic➔ allo means other. Stem cells are removed from another person, called a donor.

Acute Lymphoblastic Leukemia (ALL)

Some of ALL patients may have high number of chromosomes (more than 46) but they do well and have good prognosis, others with lower number (40-43) have poor prognosis.

Prognostic factors:

Poor Prognostic Factors

- Age < 2 yrs and > 10 yrs
- Male sex
- High WBC count (> 50 x10^9/L)
- Presence of CNS disease
- Cytogenetics
  - Good risk➔Hyperdiploid (>50 ch)
  - Poor risk➔Hypodiploid,t(9: 22), t(4:11)
- Bone Marrow: Blasts present on day 14
- Day 28:No complete response

Treatment of acute leukemias:

1-Specific therapy (chemotherapy)

Stages of Therapy:

a. Induction
b. Consolidation
c. Maintenance

A) Induction:

Obtained by using high doses of chemotherapy

1. Severe bone marrow hypoplasia
2. Allowing regrowth of normal residual stem cells to regrow faster than leukemic cells.

Remission:

- Normal neutrophil count
- Normal platelet count
- Normal hemoglobin level

Remission defined as < 5% blast in the bone marrow

-Remission = clearance of blast cells but still not cured
-The initial treatment is to clear the blast cell > induction
Then if it is cleared we give them repeated chemotherapy > consolidation
For 2-3 years to prevent the recurrence of the disease > maintenance mainly used in ALL

B) Consolidation:

- Repeated cycles of different or same drugs to those used during induction
- Higher doses of chemotherapy
- Advantage: Delays relapse and improved survival

C) Maintenance:

- Smaller doses for longer period
- Produce low neutrophil counts & platelet counts
- Objective is to eradicate progressively any remaining leukemic cells.

2- Supportive Care

- Vascular access (Central line)
- Prevention of vomiting
- Blood products (Anemia, ↓Plat)
- Prevention & treatment of infections(Antibiotics)
- Management of metabolic complications

ALL	AML
▪ **Induction** ▪ **Consolidation** ▪ **Maintenance** ▪ **CNS prophylaxis all patients> common**	▪ **Induction** ▪ **Consolidation** ▪ **No maintenance** ▪ **CNS – Selected group only**

CHRONIC LEUKEMIAS:

Definition:
Neoplasticproliferationsof mature haemopoeiticcells.
Types:

Chromic Lymphocytic Leukemia (CLL)

disease of old age

CLL isvery similar tolymphoma,soifthe WBC countis high itis CLL, butifthethe WBC countis normal it islymphoma

- Neoplasticproliferationsofmaturelymphocytes.
- Distinguishedfrom ALL by
  - **Morphologyofcells.**
  - **Degreeofmaturationofcells.**
  - **Immunologicallyimmatureblasts in ALL.**
  - CLL affectsmainlyelderly.

Symptoms:

- May beentirelyabsent in 40%
- Weakness, easy fatigue, vague senseofbeingill
- Night sweats
- Feeling oflumps
- Infectionsesp pneumonia

In late stagewhentheydevelopbonemarrowfailuretheypresentwithanemia,thrombocytopeniaandinfection

Physical Examination:

- Pallor
- Lymphoadenopathy
  - Cervical, supraclavicular nodes more commonlyinvolvedthanaxillaryor inguino-femoral
  - Non-tender, notpainful, discrete, firm, easilymovableonpalpation
- Splenomegely, mildtomoderate
- Hepatomegaly

Clinical staging-CLL:

We treat at late stage, because if we treat at early stage it might be harmful to the patient.

- (0-1) lymphocytosis± LNS.
- (II) above + hepatosplenomagely.
- (III-IV)
  - Anaemia. Hb< 10 g/l
  - Thrombocytopenia.
  - Plateletcount : <100x10^9/L.

Diagnosis-CLL:

- CBC
  - Lymphocytecount> 5 x 10^9/L (5 -500 x 10^9/L).
  - Plateletsmaybedecreased

    - Hbmaybelow
    - Bloodfilm
- PB immunophenotyping
- Bonemarrowbiopsy (neededbeforestartingtreatment)>toknowthepresenceof BM infiltration
- Imaging>toseelymphadenopaty

Treatment-CLL:

- Observation> in earlystages it maybeenough
- Chemotherapy.
    - Oral chlorambucil
    - Fludarabine, Cyclo
- Immunotherapy
    - Anti-CD 20 (Rituximab),
    - Anti-CD 52 (Alemtuzumab)
- FC-R isthecurrent standard

CLL is B-cellmalignancy where they express CD20 on their surface, so when we give Rituximab it will target these malignant cells

Indications for startingchemotherapy

- ProgressiveSymptoms. Weightloss, fever, andnightsweat.
- Progressive Anemia orThrombocytopenia
- Bulky LN, largespleen
- RecurrentInfections

Chronic Myeloid Leukemia (CML)

- CML is a clonal stemcelldisordercharacterisedbyincreasedproliferationofmyeloidelement satallstagesofdifferentiation.
- Incidenceincreaseswith age, M > F.

CML ischaracterisedby 3 distinctphases

Nowadays it is a very curable disease

- **ChronicPhase:** >mostofthepatients
    - Proliferationofmyeloidcells, which show a full range ofmaturation.
- **AcceleratedPhase**decrease in myeloiddifferentiationoccurs.
- **Blastcrisis (acuteleukemia)**

Symptoms:

- Asymptomatic (50% ofpatients)
- Fatigue
- Weightloss
- Abdominal fullnessand anorexia
- Abdominal pain, espsplenicarea
- Increasedsweating
- Easybruisingorbleeding

Signs:

- Splenomegaly (95%)
    - (50% ofpatientshave a palpablespleen ≥ 10 cm BCM, usuallyfirmand non-tender)
- Hepatomegaly (50%)

DDx of splenomegaly is very IMP:
1- CML: in young patient
2- Myelofibrosis: in old patient
3- Infection (Leishmaniasis, chronic malaria)
4- Portal vein obstruction

- Sternal tenderness is a reliable sign of disease. Is usually limited to a small area, most commonly the midbody.(fifth intercostal disease).

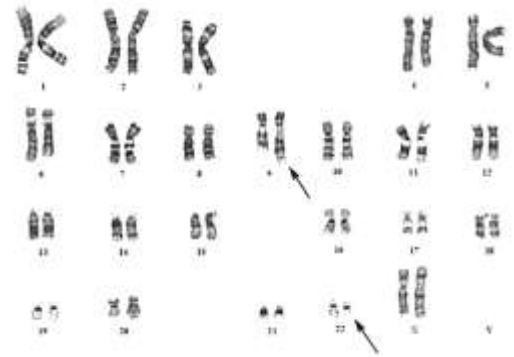
Diagnosis:

Chronicphase:

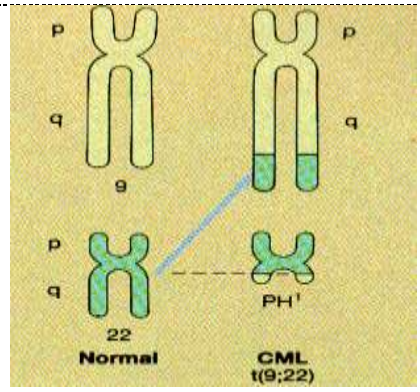
- Peripheralblood – neutrophilleukocytes 20,000 ->500, 000/μ L basophilia
  - ↓ LAP score
  - blasts< 5%
  - NucleatedRBCs
  - Thrombocytosis
  - Anaemia

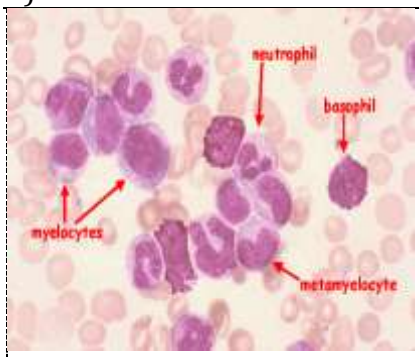
The leukocyte alkaline phosphatase (LAP) test is a laboratory test that measures the amount of a certain enzyme (alkaline phosphatase) in white blood cells.

- **Cytogenetic of CML:**
  - **Philadelphia (Ph) chromosome**isanacquiredcytogeneticabnormality in allleukaemiacells in CML.
  - Reciprocaltranslocationofchromosomal material betweenchromosome 22 andchromosome 9.t(9;22)

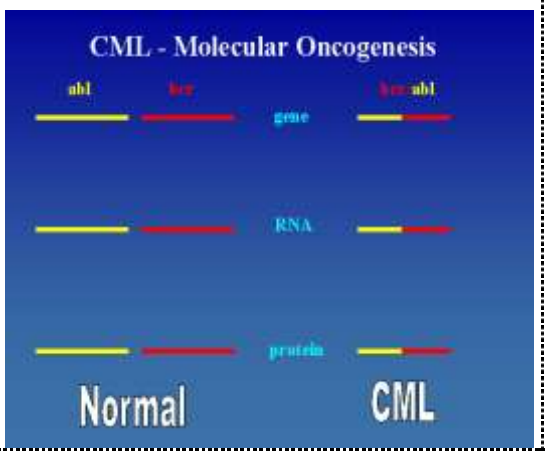


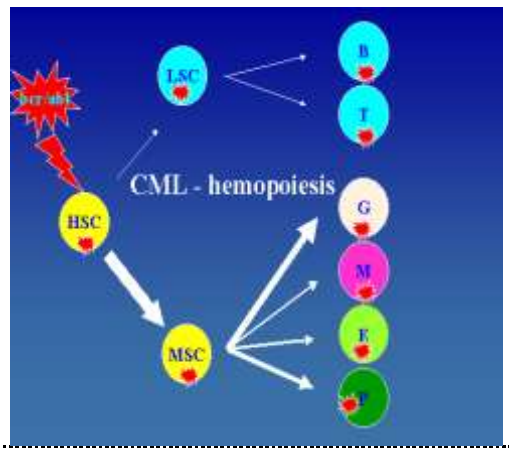
Karyotype 46,XX,t(9;22)(q34;q11.2) -- Ph chromosome

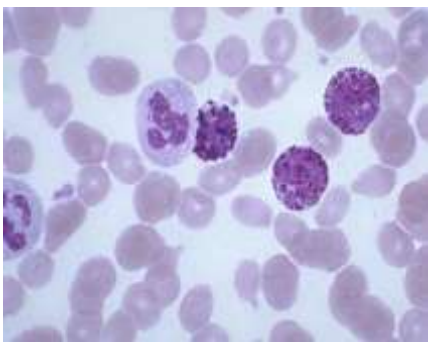




ABL and BCR are normally present but they don't have many functions; yet when they combine together they become oncogenes where they stimulate the proliferation of cancer cells







Philadelphia Chromosome, the result of a reciprocal translocation between chromosomes 9 and 22. More specifically the breakpoint cluster region (BCR) of chromosome 22 is fused with part of the Abelson (ABL) gene on chromosome 9. The resulting BCR-ABL genetic domain now located within chromosome 22 and codes for a mutant tyrosine kinase also known as BCR-ABL. Under normal circumstances tyrosine kinase proteins respond to external cellular messaging proteins, and ultimately initiate a series of reactions that culminate in cellular replication. Conversely, BCR-ABL is constitutively active, meaning it does not require activation by the aforementioned cellular messaging proteins in order to stimulate cellular replication. This results in acceleration of cell division, an inhibition of DNA repair, overall genomic instability, and the fatal blast crisis characteristic of chronic myelogenous leukemia.

CML-Treatment Response Criteria

- Hematological response
  - Normalisation of blood count
- Cytogenetic response
  - Major cytogenetic response 1-35% Ph +ve cells in metaphase
  - Minor cytogenetic response36-65% Ph +ve cells in metaphase
- Molecular response
  - Absence of BCR/ABL gene

There are 3 phases of response to treatment, the first thing is WBC count should be back to the normal level, the next phase is called cytogenetic response where Philadelphia chromosome should disappear, if more than 65% of cells become –ve we call it major cytogenetic response, if all of them become –ve we call it complete cryptogenic response. The last thing is BCR/ALB should disappear, we do it once we diagnose and start the treatment then after 3 months if it is going down that's mean the patient is responding well to treatment, if it is not we may need to change the treatment.

CML-Principles of Treatment

- Control & prolong chronic phase (non-curative)
  - Tyrosine kinase inhibitors-Imatinib (Glivec)
  - Alpha-Interferon
  - Oral chemotherapy (Hydroxyurea, ARA-C)
- Eradicate malignant Clone (curative)
  - Allogeneic BM/stem cell transplantation
  - Alpha Interferon
  - Imatinib 2nd line TKIs

Treatment of CML:

- Tyrosinekinaseinhibitor (TKI) Imatinib (Glivec) isthefirstlinetreatment
- In resistent cases 2nd lineTKIs (Nilotinib, Dasatinib, Bosutinib) veryuseful (approved as 1st line)
- Allogenicbonemarrowtrasnsplantationcanbecurative in ptsresisranttoTKIsbuthassignificantcomplications&mortality
- Acceleratedandblastphase
  - Imatinib, 2nd lineTKIs
  - Treatlike AML or ALL followedby BMT

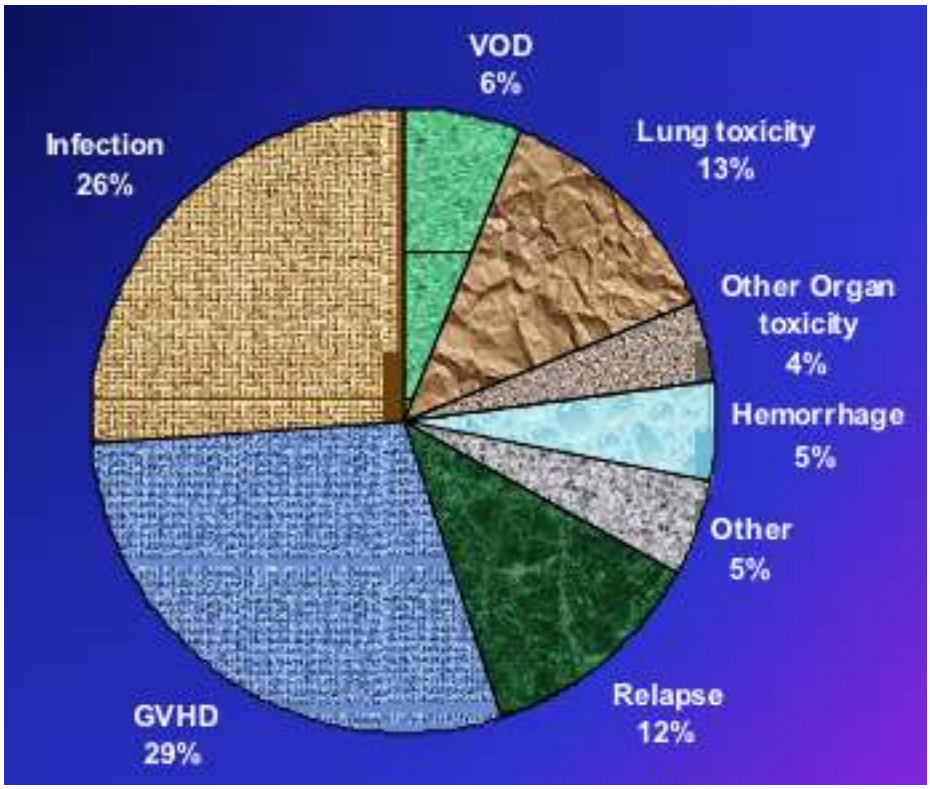
CML VS Leukmoid reaction:

1- LA P Score
2- Philadelphia chromosome
3- Basophilia
4- splenomegaly

Bone marrow or PBSC transplantation in leukemias

- Types of transplant

    - Autologous transplant
    - Allogeneic Transplant
- Purpose of transplant
    - Autologous
        - To deliver a high dose of chemo to kill any residual cancer (lymphoma, multiple myeloma)
    - Allogeneic
        - To eradicate residual leukemia cells
        - Graft vs leukemia effect
- Technique of transplantation
    - MHC + HLA matching
    - Chemotherapy
    - Total body irradiation
    - GVHD prophylaxis
- Complications of transplantation
    - Prolonged BM suppression (graft failure)
    - Serious infections
    - Mucositis
    - Graft versus host disease (GVHD)



Summary& Questions:

From Step-up to medicine:

	Acute Myelogenic Leukemia (AML)	Acute Lymphocytic Leukemia (ALL)
Acute	• Mostly in adult • Don't response well to treatment • Acute promyelocytic leukemia (APL) characterized by t(15;17) often presents with pancytopenia	• The most common malignancy in children <15 years • Well response to treatment • Poor prognosis when there is CNS involvement.
Clinical features:	• Anemia • Increased risk of bacterial infection • Bleeding • Spleno+hepatomegaly + lymphadenopathy • Bone and joint pain • CNS involvement	
Diagnosis:	**1- Laboratory:** • WBC count is variable (1000-10,000/mm3). High BLAST cells in peripheral blood • Anemia • Thrombocytopenia **2- Bone marrow biopsy**	
Treatment:	**1- Emergencies:** • Blood culture, antibiotics for infections • Blood transfusion for bleeding and anemia **2- Aggressive, combination chemotherapy:** in high doses for several weeks to obtain remission>> then maintenance therapy is used for months or years to prevent recurrence ALL>achieve complete remission AML> require BM transplantation **3- Bone marrow transplantation**	
Chronic	Chronic myelogenic leukemia (CLL)	Chronic lymphocytic leukemia (CLL)
	• Patients are usually older than 40 years of age. • It follows a chronic phase for many years before it transform into acute leukemia in the acute phase • Associated with translocation t(9;22) resulting in Philadelphia chromosome	• Most common leukemia that occurs after age 50 • The cause is unkonown • The least aggressive type of leukemia, patients survive longer than those with CML, or acute leukemias
Clinical features	• Most patients present in the chronic phase (85%), may be asymptomatic at the time of diagnosis> discovered on routine CBC • Fever, night sweat, weight loss • Recurrent infections, easy bruising/bleeding, symptoms of anemia • Spleno+hepatomegaly +lymphadenopathy • in the acute phase (blast crises)	• Asymptomatic, discovered on routine CBC • Generalized painless lymphadenopathy • Splenomegaly • Fatigue, weight loss, pallor, skin rashes, easily bruising, bone tenderness, and/or abdominal pain

Diagnosis	**1- Laboratory findings:** CBC- WBC: 50,000 to 200,000 Small number of blasts Eosinophilia Periphram smear Decreased leukocyte alkaline phosphatase activity Thrombocytosis **2- Bone marrow biopsy**: leukemic cells	**1- Laboratory findings:** CBC- WBC: 50,000 to 200,000 Anemia, thrombocytopenia and neutropenia Peripheral blood smear is often diagnostic Almost all WBCs are MATURE Flow cytometry> clonal population of B cells **2- Bone marrow biopsy:** presence of infiltrating leukemic cells.
Treatment	Oral tyrosine kinase inhibitors (TKI)	**1- Chemotherapy** Given for symptomatic relief and reduction of infection

From Medicine Recall:

How is the diagnosis of leukemia made? Presence of leukemic blasts on peripheral smear, bone marrow aspirate, and biopsy. Auer rods suggest AML.
Immunohistochemicalstains, flow cytometry, and cytogenetics are useful in subtyping leukemias and determining treatment options.
What congenital disorders are associated with an increased incidence of leukemia? Down syndrome, Bloom syndrome, Fanconi anemia, and ataxia telangiectasia
What acquired disorders are associated with an increased incidence of leukemia?Myeloproliferative diseases, myelodysplastic syndromes, and aplastic anemia
What treatments/therapies are associated with an increased risk of leukemia? Chemotherapy and radiation therapy
What is induction chemotherapy? The first cycle of chemotherapy intended to induce remission

ACUTE LYMPHOID LEUKEMIA	ACUTE MYELOID LEUKEMIA
What is the basis for the new World Health Organization (WHO) classification subtypes of ALL? Grouped according to B- or T-cell lineage, and then by cytogenetic subgroups **What physical findings are more common in ALL than AML?**Lymphadenopathy occurs in more than half of patients with ALL and is relatively uncommon in AML. Hepatosplenomegaly occurs in more than two-thirds of patients with ALL. Hepatomegaly is uncommon in AML, although splenomegaly occurs in approximately half of patients with AML. **What are sanctuary sites in ALL?** Common sites of solitary relapse include the CNS and testes. To protect the CNS, all patients with ALL undergo prophylactic cranial irradiation or intrathecal chemotherapy along with induction chemotherapy **How do you differentiate ALL from AML?**Flow cytometry is the gold standard. PAS immunohistochemical stain is positive in ALL. Myeloperoxidase histochemical stain is positive in myeloid leukemias.	**What is the most common type of acute leukemia in adults?**Myeloid leukemia comprises 80% of all cases of acute leukemia in adults. **What is the age distribution for AML?** Most patients are older than 65 years. **Which subtype of AML is commonly associated with DIC?**Promyelocytic (M3)
CHRONIC LYMPHOCYTIC LEUKEMIA	CHRONIC MYELOGENOUS LEUKEMIA
What is CLL? A neoplastic proliferation with accumulation of immune-incompetent lymphocytes within the bone marrow, peripheral blood, and lymphoid organs **What are the morphologic characteristics of CLL?** Small, mature lymphocytes with clumped chromatin and scant cytoplasm	**What cells are characteristically elevated in patients with CML?** All stages of myeloid development may be seen on the smear, from rare myeloblasts to mature, normal-appearing neutrophils, with a spectrum of maturation. **What is the average WBC count and appearance of the peripheral smear in patients with CML?** At the time of

What is the median age of onset of CLL? 65 years
What are the symptoms of CLL? Up to 70% of persons with CLL are asymptomatic at diagnosis. Generalized lymphadenopathy, fever, night sweats, weight loss, easy fatigability, weakness, and increased bleeding are common complaints. Frequent infections and exaggerated responses to insect bites are occasionally noted.
How is the diagnosis of CLL made? An increase in the absolute number (5,000) of lymphocytes in the peripheral blood, which, on the peripheral smear, appear as small, mature lymphocytes. Flow cytometry shows a monoclonal population that coexpresses CD19 and CD5. Frequently, there is lymphadenopathy, splenomegaly, and bone marrow infiltration, making it difficult to distinguish CLL from its lymphomatous counterpart, SLL.
What specific hematologic complication can be associated with CLL? AIHA (autoimmune hemolytic anemia)
What is the treatment for CLL? Observation if the patient is asymptomatic. Oral alkylating agents or fludarabine are commonly used in symptomatic patients.
What are the indications for the treatment for CLL? Some indications include the presence of autoimmune hemolytic anemia, autoimmune thrombocytopenia, bulky lymphadenopathy, progressive hyperlymphocytosis, and frequent bacterial infections, but the exact time to initiate treatment is an area of much debate.

diagnosis, the WBC count is often between 100 and 300 □ 109/L, with a mild normochromic normocytic anemia.
How is a leukemoid reaction distinguished from early CML? The LAP level is very low in CML and elevated in a reactive leukocytosis.
What chromosomal abnormality is highly associated with CML?
The Philadelphia chromosome, a balanced translocation between chromosomes 9 and 22, is usually found. The fusion gene produces a constitutively activated BCR-ABL tyrosine kinase.
What are the symptoms of CML? Lethargy, weight loss, increasing abdominal girth, sweating, and easy bruising and bleeding, as well as symptoms attributable to anemia and splenomegaly
What are the physical findings of CML?
Splenomegaly and hepatomegaly
What are the 3 phases of CML? Chronic phase, accelerated phase, and blast phase
What is the treatment of CML in the chronic phase? Imatinib (Gleevec); second-line agents include dasatinib and nilotinib. Those with an HLA-matched sibling could proceed to allogeneic BMT, the only curative treatment. -Interferon can induce complete remissions and prolong survival, but it has a lower response rate.
What is imatinibmesylate? A tyrosine kinase inhibitor that specifically targets the product of the Philadelphia chromosome, the BCR-ABL tyrosine kinase
How can one measure the response of CML to imatinib?
Hematologic: normalization of CBC
Chromosomal: loss of the Philadelphia chromosome in blood or marrow by karyotype and FISH testing
Molecular: 3-log reduction in the BCR-ABL transcript by PCR testing

1- a 65 year old male presents with fever, fatigue, and unintentional 10 kg weight loss over last 3 weeks. On examination he has generalized lymphadenopathy and spleen tip was palpable. CBC shows high WBC 35,000 with 50% neutrophils, 40% mature looking lymphocytes and few myelocytes and metamyleocytes.
What is the most likely underlying diagnosis?
A. Acute lymphoblastic leukemia
B. Chronic lymphocytic leukemia
C. Acute myeloid leukemia
D. Chronic myeloid leukemia

2- young male has bruises, fever, fatigability, blood analysis reveal that he has WBC:20x100(80 blastocyte) increased , platelets decreased with prolong coagulation profile the patient is most likely has:
A. DIC
B. acute leukemia
C. acutehaemolysis
D. Eptsein Barr virus-induced hemolysis

3- 27 years old man with acute leukemia, developed fever while in hospital 4 day after he received chemotherapy. His temperature is 39 c. total white blood cells count is 0.1x109 cells/l (Normal range 4-11 cells x 109/L). The most likely cause of his fever is:
A. Related to leukemia
B. Viral infection
C. Bacterial infection
D. Fungalinfection

4- A 25 year old girl with leukemia, received the first cycle of chemotherapy,one week before she came to the emergency department complaining of fever for about 3 hours . Her temperature is 39C and pulse is 102 beats / minutes the rest of the physical examination is normal.Total WBC are 0.1X 10 cells/L. the patient was admitted .
Which one of the following management should we do?
A. oralciprofloxacine
B. IV pipracillin / tazobactam +amikacin
C. take blood culture and wait for the result
D. Anti pyretic

1:B, 2:B, 3:C, 4:B

CASE1:

A 57-year-old man is referred to a hematologist for a detailed workup of severe anemia. His hemoglobin level is 8.2 mg/dL, but testing reveals no evidence of occult bleeding. For the past several weeks, he has noticed blood on his toothbrush nearly every day and complains that he bruises more easily than before. He has become increasingly fatigued and has cut back on his hours at the waste management company he owns because he often becomes short of breath with mild exertion. In addition, he complains that "I always have a cold; I can't get rid of it." In addition to the low hemoglobin, relevant laboratory results include a WBC count of 3200/mm3, reticulocyte count of 0.8%, and platelet count of 30,000/mm3. His peripheral smear shows myeloblasts and markedly decreased granulocytes.

What is the most likely diagnosis?

Acute myelogenous leukemia (AML). While often idiopathic, AML may be caused by radiation therapy, chemical exposure such as excessive benzene exposure, and alkylating chemotherapeutic agents. AML is a clonal malignancy. It may present de novo or arise out of myelodysplasia or another hematologic malignancy. It is characterized by an increase in blast cells in the bone marrow (> 20%), as well as impaired production of normal RBCs, platelets, and neutrophils. Patients often present with pallor, fatigue, weakness, palpitations, dyspnea on exertion, and other symptoms of anemia. They may also have gingival bleeding, easy bruising, and conjunctival hemorrhages, which indicate thrombocytopenia. In addition, minor infections of
the skin are common. Major infections (pneumonia, meningitis, etc.) are less likely because severe neutropenia may not occur until chemotherapy is instituted.

What is the pathogenesis of this condition?

AML is associated with characteristic somatic mutations or translocations in hematopoietic stem cells. These mutant progenitors do not produce normal red cells, granulocytes, or platelets and have a survival and proliferative advantage over normal cells. Since the growth of normal cell lines is suppressed relative to the mutant cells, patients develop anemia, thrombocytopenia, and neutropenia.

What tests could be used to confirm the diagnosis?

A CBC and bone marrow biopsy both aid in the diagnosis of AML. Patients are typically anemic due to their inability to produce normal RBCs, as reflected by reticulocyte counts in the range of 0.5–2.0%. The platelet count will be low, often < 50,000/⌈L. The WBC count will generally be < 5000/mm3, while the total neutrophil count will be < 1000/mm3. On a peripheral smear (see Figure 7-1), the major abnormality is seen in the white cells. Some patients will have Auer rods (elliptical cytoplasmic inclusions; Figure 7-2) in their blast cells, although this is more common in acute promyelocytic leukemia. A bone marrow biopsy will contain > 20% blast cells.

What is the most appropriate treatment for this condition?

The goal of treatment is to induce remission. Induction therapy typically combines an anthracycline antibiotic with cytarabine. Complications include hyperleukocytosis and pancytopenia. Red cell transfusions are needed to maintain the hematocrit during treatment. In order to induce sustained remission, either consolidation chemotherapy or allogenic bone marrow transplantation is necessary.

CASE2:

A 4-year-old girl presents to her pediatrician with a 2-week history of cough, nasal congestion, and fatigue. Her mother has brought her in three times in the past month for unremitting cold symptoms; each time, the child has been diagnosed with a viral upper respiratory infection and advised on symptomatic care. She has been sleeping more than usual for the past few days, and she awoke this morning with a new rash. On physical examination, she has a temperature of 37.2° C (99.0° F) and pulse of 140/min. She appears pale, with scattered petechiae across her lower legs, hepatosplenomegaly, and cervical lymphadenopathy.

How should the workup proceed for this patient's condition?

This child presents with symptoms suggesting multiple cytopenias; therefore, the first test should be a complete blood count (CBC) with peripheral blood smear. In this patient, the CBC will show neutropenia, anemia, and thrombocytopenia with lymphoblasts on peripheral smear.

What is the most likely diagnosis?

Acute lymphoblastic leukemia (ALL). ALL is the most common malignancy of childhood. While suggested by the presence of lymphoblasts on peripheral smear, a diagnosis of ALL must be confirmed by bone marrow aspirate and/or biopsy showing > 25% lymphoblasts.

What conditions should be included in the differential diagnosis?

- Chronic viral infection (Epstein-Barr virus [EBV], cytomegalovirus [CMV]).
- Immune thrombocytopenic purpura (ITP).
- Aplastic anemia.
- Autoimmune hemolytic anemia.
- Juvenile rheumatoid arthritis.

What are the stages of treatment, and what chemotherapies are commonly used?

Induction is the first month of therapy and commonly involves oral prednisone or dexamethasone, intramuscular asparaginase, intravenous vincristine, and intrathecal methotrexate; over 95% of patients have minimal residual disease on repeat bone marrow examination after

this stage. Consolidation involves continued systemic therapy with intrathecal and cranial radiation therapy to eradicate CNS "sanctuaries" of disease. A brief intensification of therapy following consolidation has been shown to improve survival in pediatric patients. Finally, maintenance chemotherapy is continued for several years to ensure remission. Hematopoietic stem cell transplantation may be used to treat relapsed patients.

What patient or disease characteristics indicate a negative prognosis?

- Patient age >10 years.
- WBC count at diagnosis >50,000/mm3.
- CNS or testicular involvement.
- Residual disease following induction therapy.
- Chromosomal translocations t(9;22) or t(4;11).

The patient begins induction chemotherapy but develops acute renal failure with a uric acid level of 16 mg/dL. What is the problem, and what is the most appropriate treatment?

Tumor lysis syndrome typically occurs early in the treatment of patients with a high tumor burden due to the lysis of tumor cells. Laboratory hallmarks include hyperkalemia, hyperphosphatemia, hypocalcemia, and hyperuricemia. Aggressive hydration, alkalinization of urine, and administration of allopurinol (which inhibits uric acid production) are important components of treatment. Hemodialysis may be necessary if renal failure is severe.

CASE3:

A 60-year-old white man presents to his family physician for a full physical examination. His last doctor's visit was 2 years ago when he had the flu. He explains that his health has been "relatively okay" and that he has had many colds but no ongoing medical problems. Upon further probing, he admits that he has been increasingly tired for the past 6 months and that he has lost about 9 kg (20 lb) over the past 2 years. He insists that he is healthy and needed to lose the weight, but he denies any exercise regimen or specific dietary restrictions. His vital signs are within normal limits, and he appears comfortable. Upon examination, he has palpable lymph nodes in his neck and axillae. He denies any pain on palpation; the nodes range from 1.0 cm to 3.0 cm in diameter. His abdominal examination is significant for hepatosplenomegaly. His chemistry test values are within normal limits; his WBC count is 25,200/mm3 with 90% lymphocytes.

What is the most likely diagnosis?

Chronic lymphocytic leukemia (CLL). CLL is a B-cell malignancy that occurs in older patients; the median age at presentation is 65 years. The proliferating B cells cannot effectively recognize antigen, so patients are often immunocompromised and may have coexistent hypogammaglobulinemia. Some patients present with fatigue or lymphadenopathy, while the majority are asymptomatic. Eventually, some patients will develop hepatomegaly or splenomegaly. The disease progresses very slowly in most cases; rarely, patients will develop an aggressive large cell lymphoma (Richter's syndrome). Staging for the nonzero stages of CLL (more than just lymphocytosis) follows the Rai system, which can be remembered using the mnemonic **LOATh**:

I—**L**ymphocytosis + **L**ymphadenopathy
II—**O**rganomegaly (splenomegaly or hepatomegaly)
III—**A**nemia
IV—**Th**rombocytopenia

What is the pathogenesis of this condition?

CLL is a monoclonal proliferation of mature lymphocytes. Despite their morphological maturity, these B cells are unable to produce functional antibodies. This renders patients more susceptible to infection, but patients may be asymptomatic for prolonged periods, given the slow progression of the disease.

What tests could be used to confirm the diagnosis?

A CBC and peripheral blood smear demonstrating isolated lymphocytosis are virtually diagnostic for CLL. The WBC count will usually be > 20,000/mm3, with a total lymphocyte number of > 6,000/mm3. The cells will be small, mature, and structurally indistinguishable from immunocompetent lymphocytes (Figure 7-3). They express both the CD19 surface marker commonly found on B cells, as well as the CD5 marker usually attributed to T cells; this finding in combination with the lymphocytosis is diagnostic for CLL. Cyclin D should be evaluated to rule out mantle cell lymphoma, a common mimic.

What is the most appropriate treatment for this condition?

In the case of indolent CLL, no treatment is required. If patients are symptomatic—fatigued, anemic, thrombocytopenic, or presenting with lymphadenopathy—chemotherapy, often with purine analog-based therapy (i.e., fludarabine) is warranted. If patients have autoimmune hemolytic anemia, they should receive prednisone or a splenectomy; fludarabine may exacerbate warm autoimmune hemolysis. In patients with severe, refractory disease, allogenic bone marrow transplantation or antibody (anti-CD52) may be beneficial; however, the role of allogenic transplantation is limited by the fact that the average patient is over the age of 60.

What is the prognosis for patients with this condition?

The median survival for CLL patients depends on the stage of disease at diagnosis. Patients with stage 0 or stage I disease have a median survival of 10–15 years and may live a normal life for many years. Patients with stage III or stage IV disease have a median survival of at least 2 years; this will hopefully improve as new therapies are developed. Newer cellular markers, such as the presence of somatic mutations in the immunoglobulin domains, ZAP-70, cytogenetics, and CD38 may allow identification of different prognostic categories for this disease.