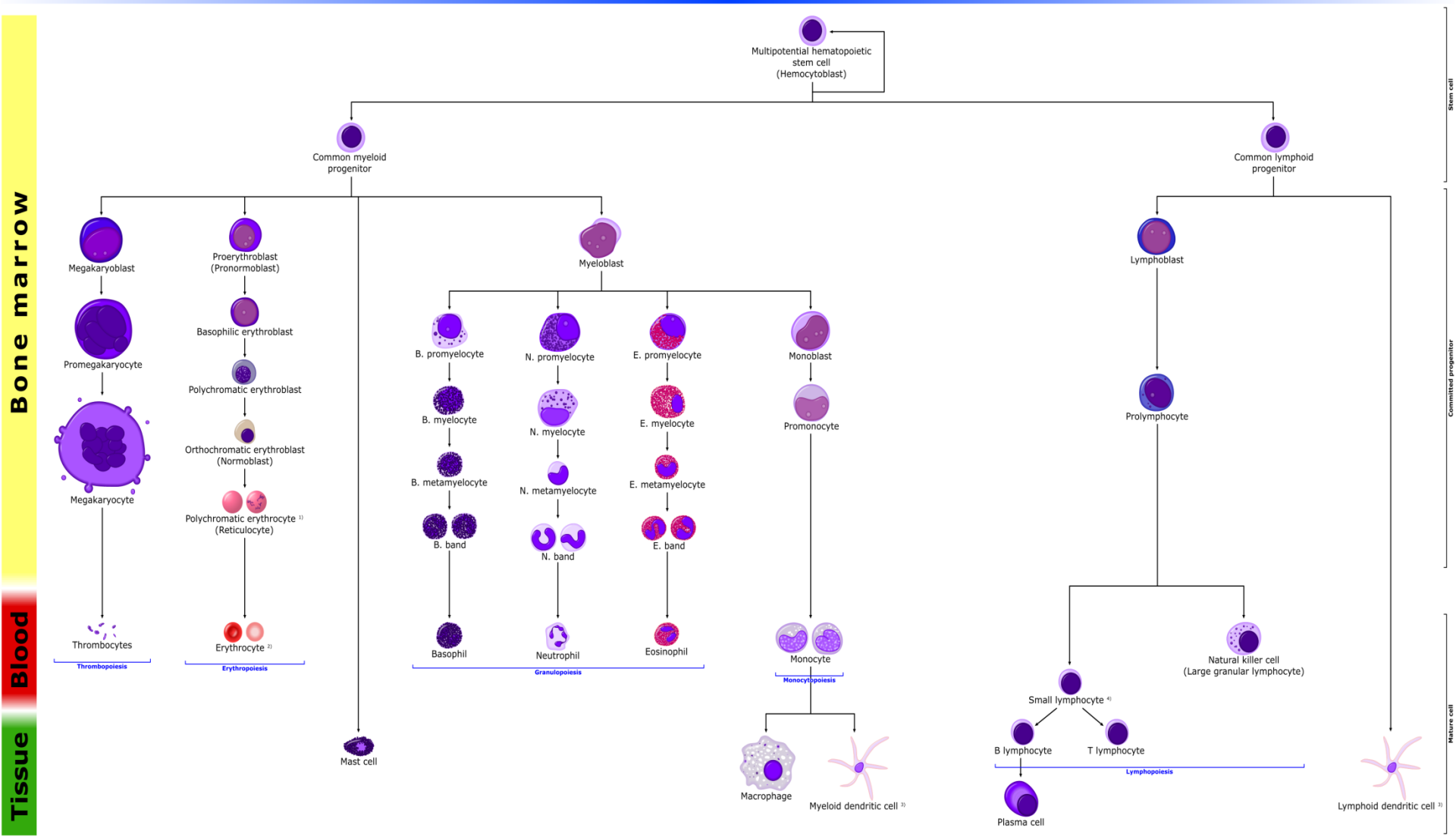
Myeloproliferative disease

Myelodysplastic syndrome

Hematopoiesis in humans



Notes

- Approximate scale information: 10 µm
- The morphological characteristics of the hematopoietic cells are shown as seen in a *Wright's* stain, *May-Giemsa* stain or *May-Grünwald-Giemsa* stain. Alternative names of certain cells are indicated between parentheses.
- Certain cells may have more than one characteristic appearance. In these cases, more than one represenation of the same cell has been included.
- Together, the monocyte and the lymphocytes comprise the *agranulocytes*, as opposed to the *granulocytes* (basophil, neutrophil and eosinophil) that are produced during granulopoiesis.
- *B., N. and E.* stand for *Basophilic, Neutrophilic* and *Eosinophilic*, respectively. As in Basophilic promyelocyte.

1) The polychromatic erythrocyte (reticulocyte) at the right shows its characteristic appearance when stained with methylene blue or Azure B.

2) The erythrocyte at the right is a more accurate representation of what it looks like in reality when viewed through a microscope.

3) Other cells that arise from the monocyte: osteoclast, microglia (central nervous system), Langerhans cell (epidermis), Kupffer cell (liver).

4) For clarity, the T and B lymphocyte are split to better indicate that the plasma cell arises from the B-cell. Note that there is no difference in the appearance of B- and T-cells unless specific staining is applied.

Definition

Malignant transformation of haematopoietic stem cell

- uncontrolled proliferation
- impairment of differentiation

Pluripotent stem cell impairment

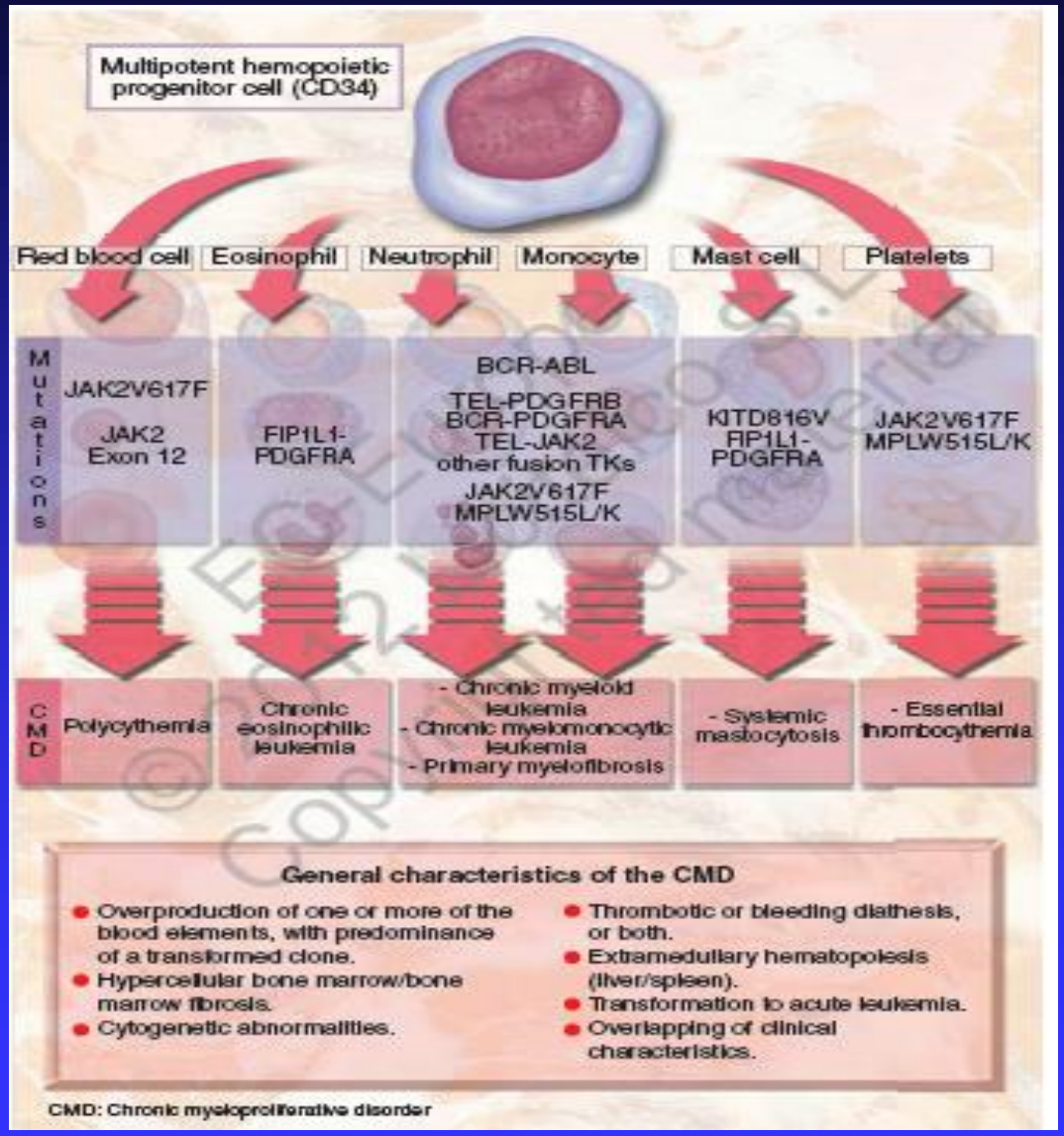
- abnormal proliferation of erythroid, granulopoietic and megakaryopoietic line

Frequently followed by:

- bone marrow fibrosis
- extramedullary haemopoiesis in spleen and liver

Usually more than one line proliferate

Classification of myeloproliferative disorders



Molecular pathogenesis of the MPD

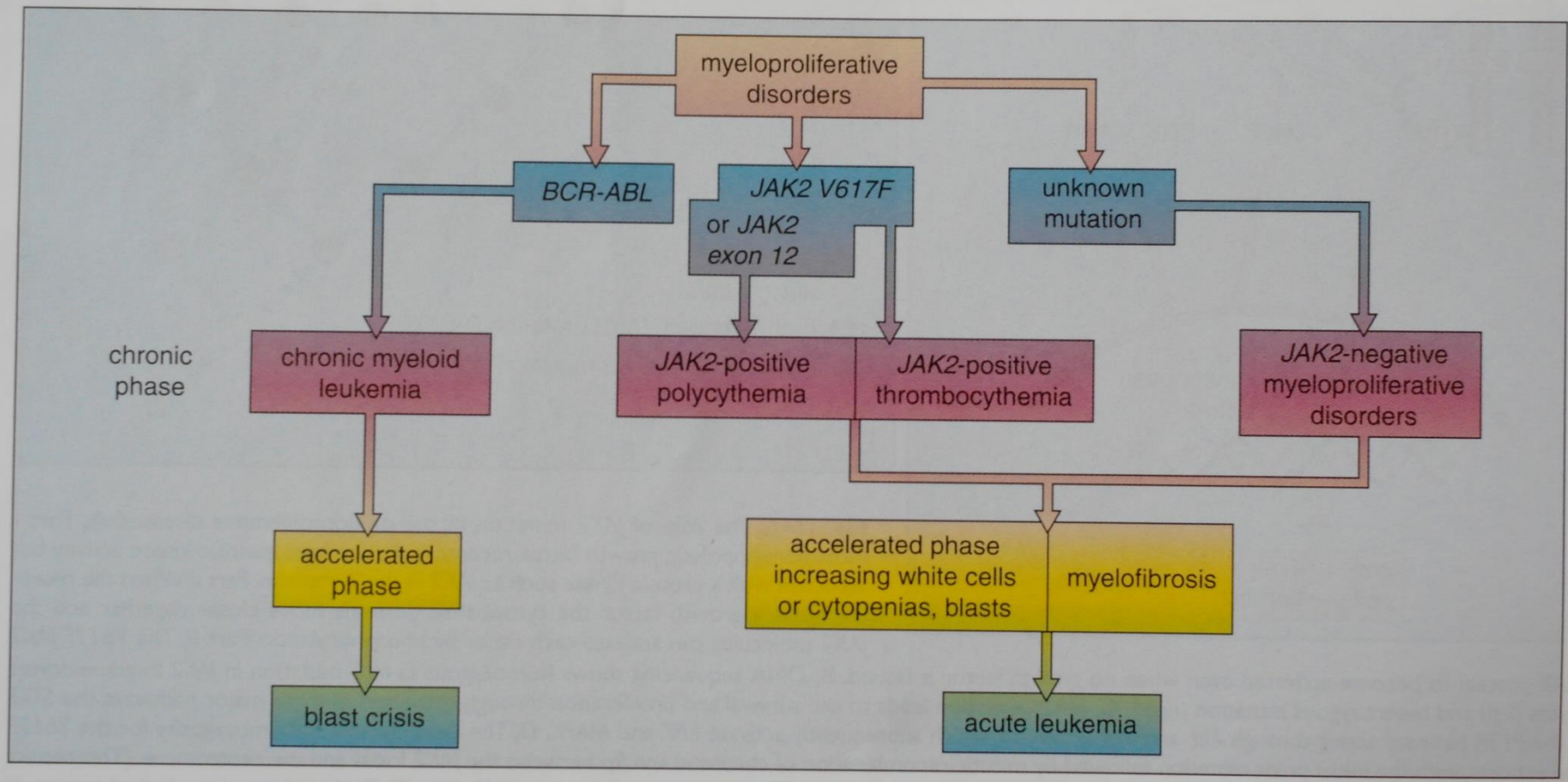


Fig. 15-2. Molecular pathogenesis of the myeloproliferative disorders.

WHO classification of myeloproliferative diseases

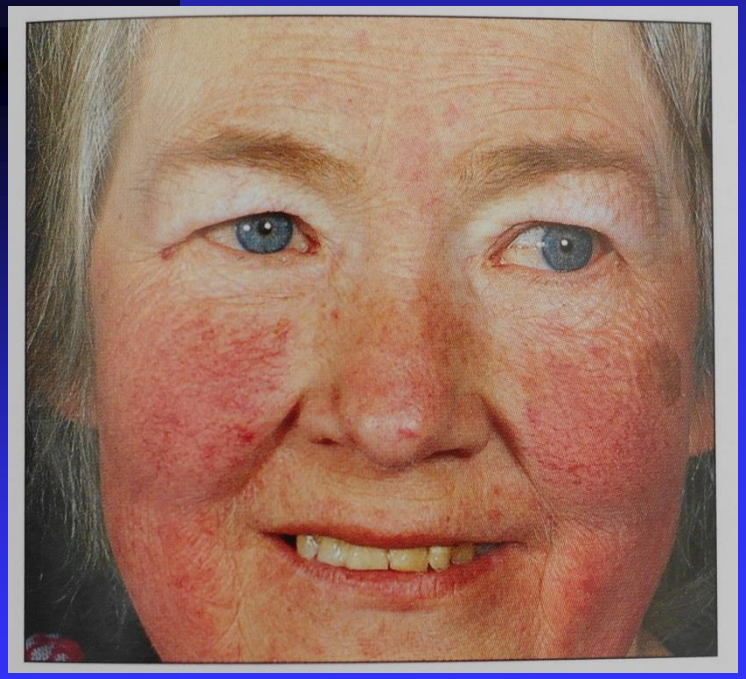
- Chronic myeloid leukaemia
- Chronic neutrophilic leukaemia
- Chronic eosinophilic leukaemia and hypereosinofilic syndrome
- **Polycythaemia vera**
- **Chronic idiopathic myelofibrosis**
- **Essential trombocythaemia**
- Chronic myeloproliferative disease unclassifiable

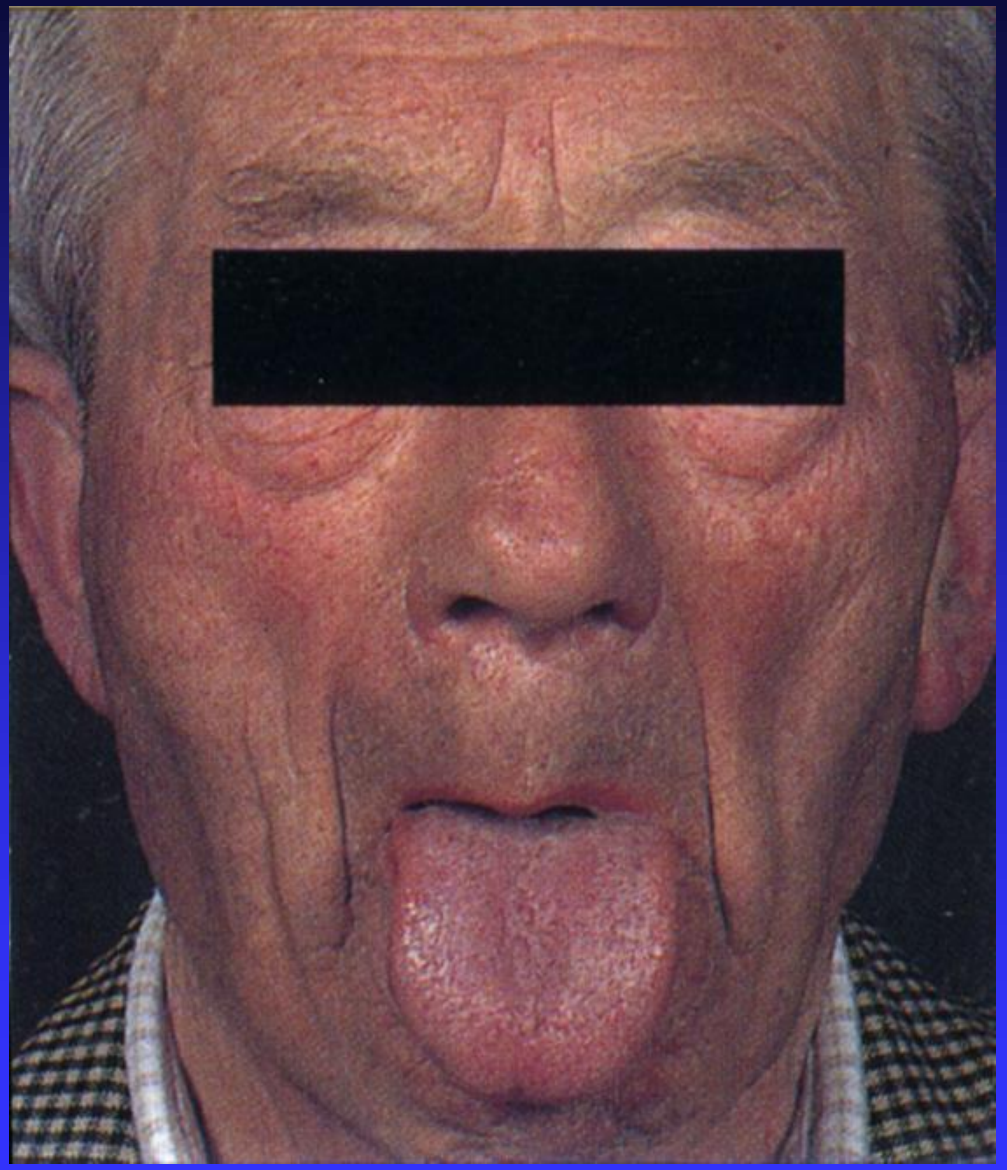
Polycythaemia vera

- primary polycythaemia

- Clonal disease , 95% nucleotid mutation JAK2 V617F
- Enhanced proliferative activity
- Differentiation mostly to erythroid line
- Increased circulating blood volume
- Absolute increase of erytrocytes more than 25% above upper range limit
- Hb 170 g/l, Ery $6x10^{12}$/l, hct 0,55
- all lines impaired
- later marrow fibrosis or acute leukaemia transformation

Facial appearance





Further tests

- Abdominal USG: splenomegaly, no kidney abnormality (x erythropoietin secreting tumor)
- No congenital heart disease (x plasma volume reduction, diuretic therapy)
- Normal blood gases, normal pulmonary function tests (x chronic hypoxia)

Suggested treatment

- Venesection, erytrocytopheresis
- Interferon α
- Hydroxyurea
- Selective JAK2 inhibitors, JAK2+flt3, JAK1/JAK2 TKI and pomalidomide

- Acetylsalicylic acid 75 mg/day

- No iron supplements

Essential myelofibrosis

- chronic idiopatic myelofibrosis, myelosclerosis

- Gradual replacement of haematopoiesis in bone marrow with fibrotic tissue. Platelets growth factor stimulates fibroblasts to collagen production
- Proliferation of haemotopoietic tissue with extramedulary localisation (spleen and liver with organomegaly)
- malignant transformation of stem cell
- in parallel unmatured granolocytes and erytroblasts in the circulation
- 50-60% mutation JAK2 V617F

- Prefibrotic and fibrotic stadium
- Acute leukaemia transformation

Examination of abdomen

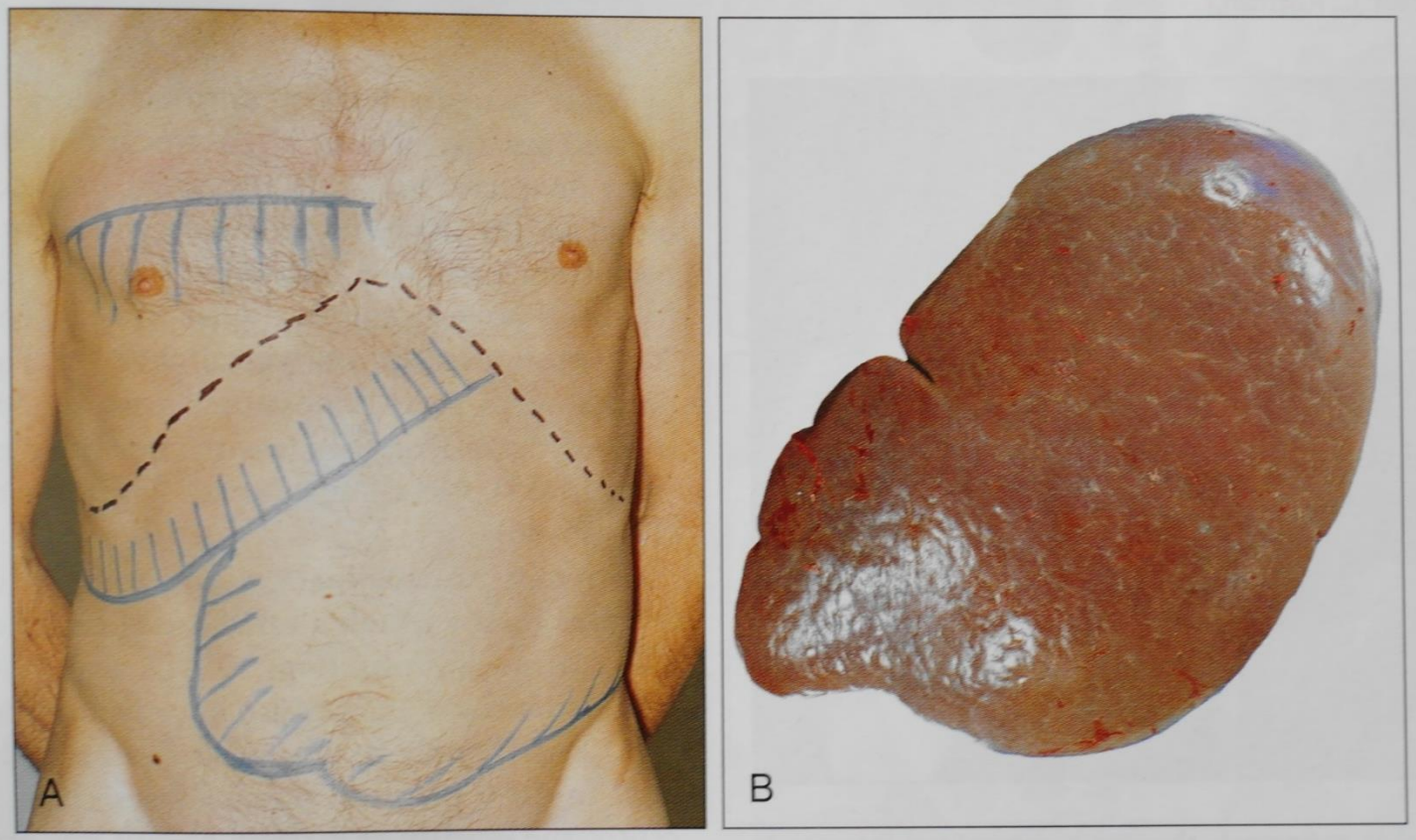
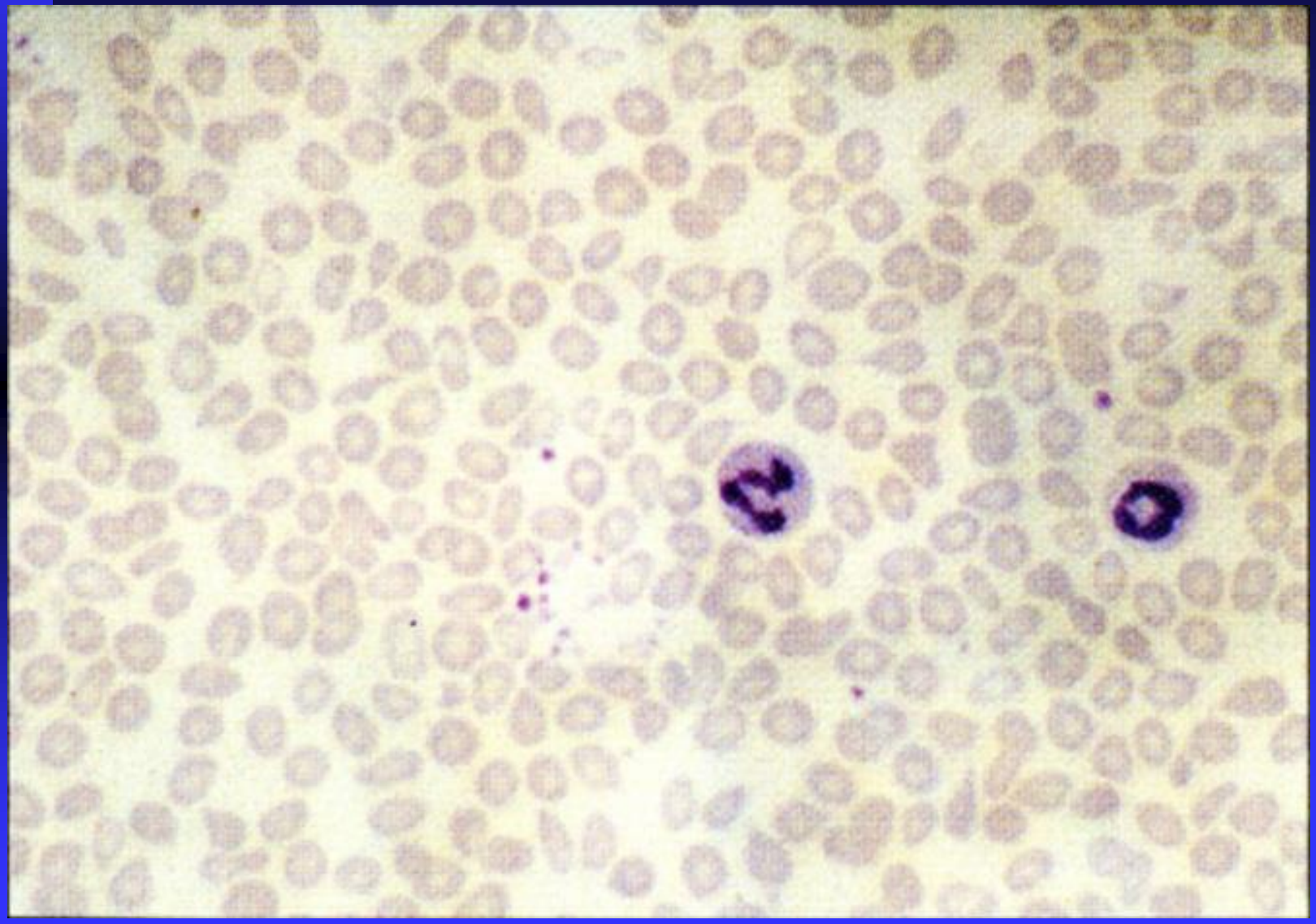
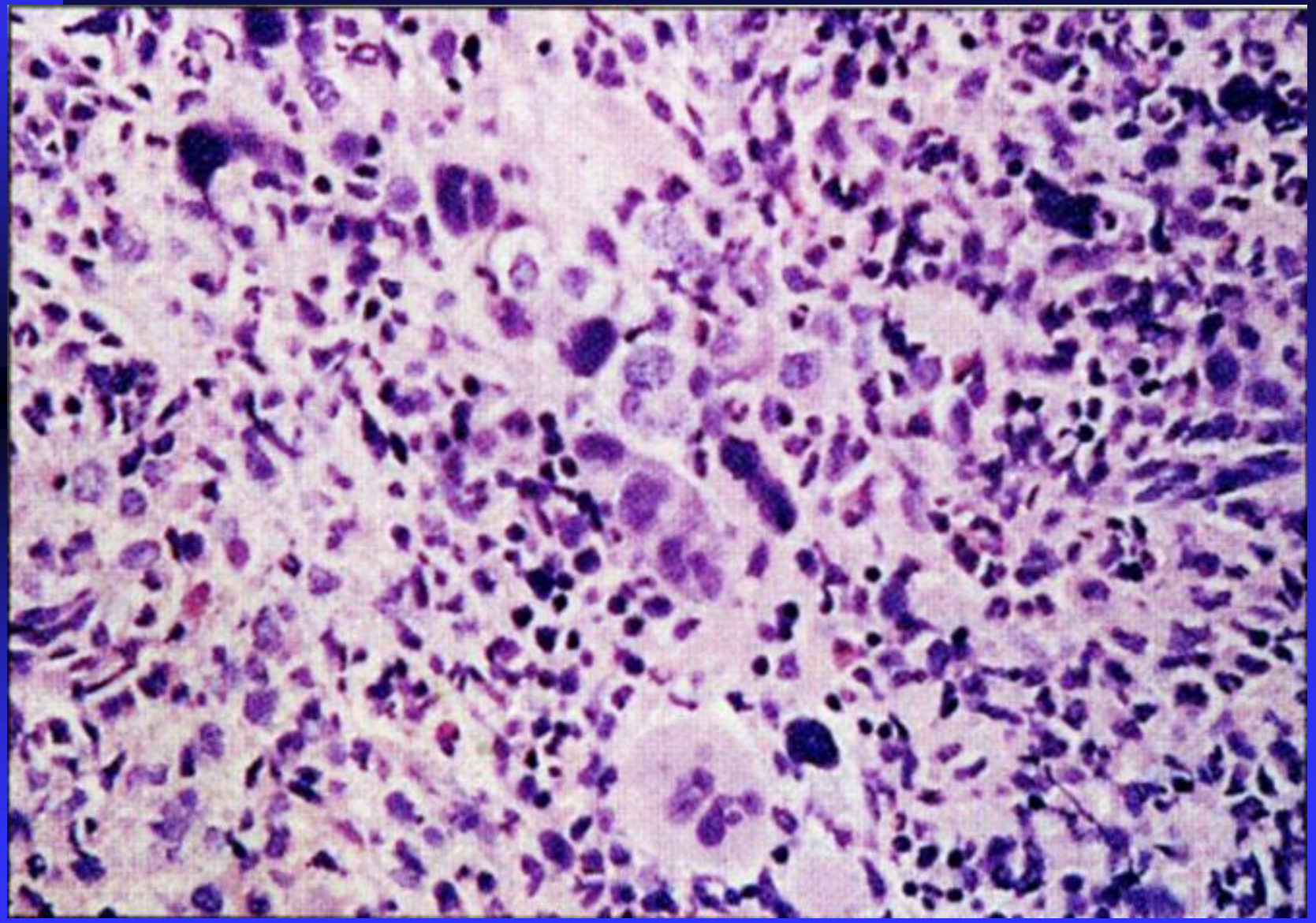


Fig. 15-37. Myelofibrosis. **A,** Splenohepatomegaly; **B,** the patient's spleen shows a well-defined notch in the superior border. The prominent indent in the inferior border was palpable during clinical examination.

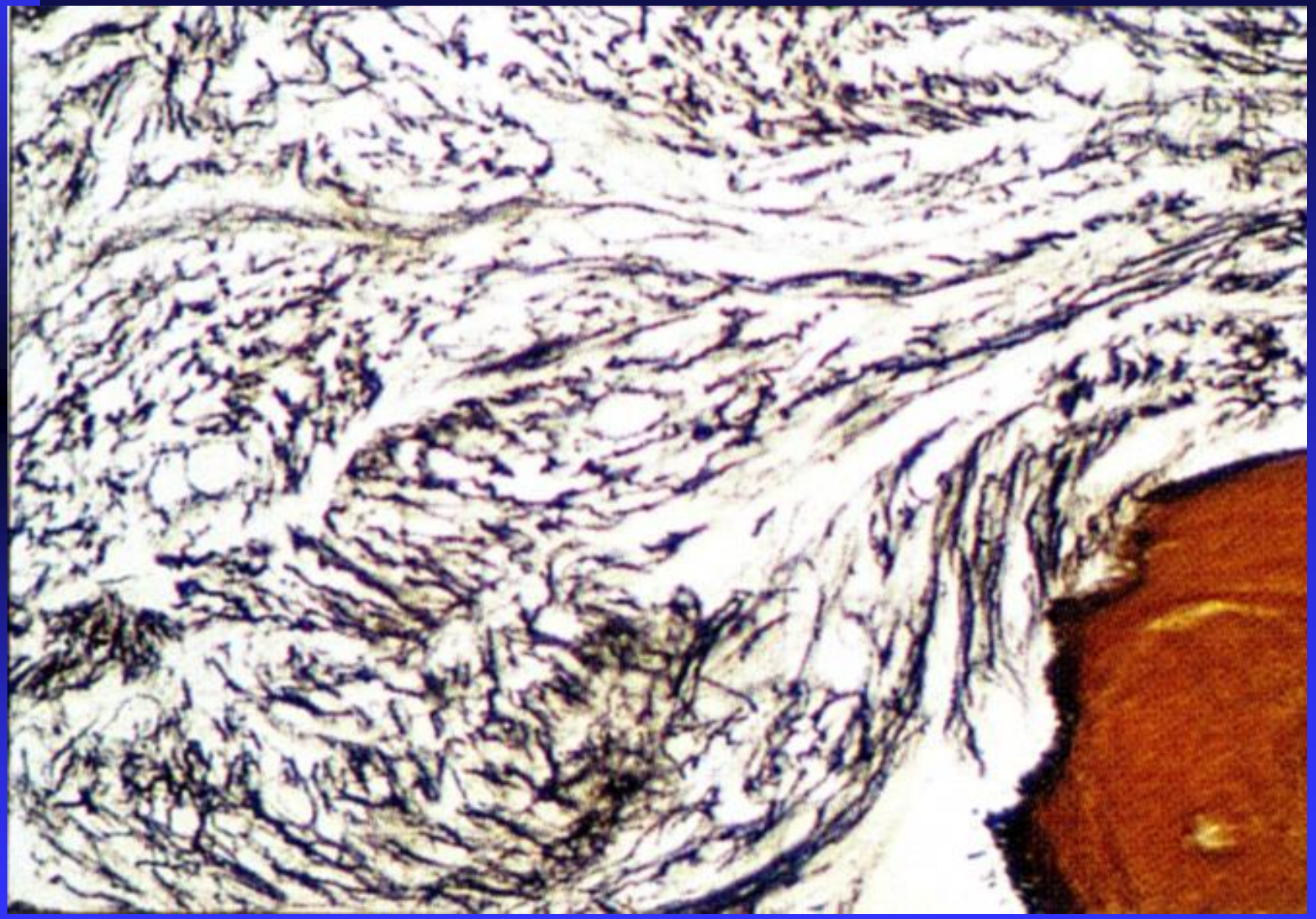
Blood smear



Trephine biopsy



Trephine biopsy



Treatment approach

- No splenectomy if not needed (haemolysis, high abdominal tenderness)
- Supportive care (transfusions with chelation, antiinfective treatment if necessary)
- Hydroxyurea, interferon α (reducing spleen size and secondary symptoms)
- Selective JAK2 inhibitors -JAK1/JAK2 ruxolitinib
- Experimentaly: Thalidomide a Lenalidomide, Pomalidomide – immunomodulatory drugs

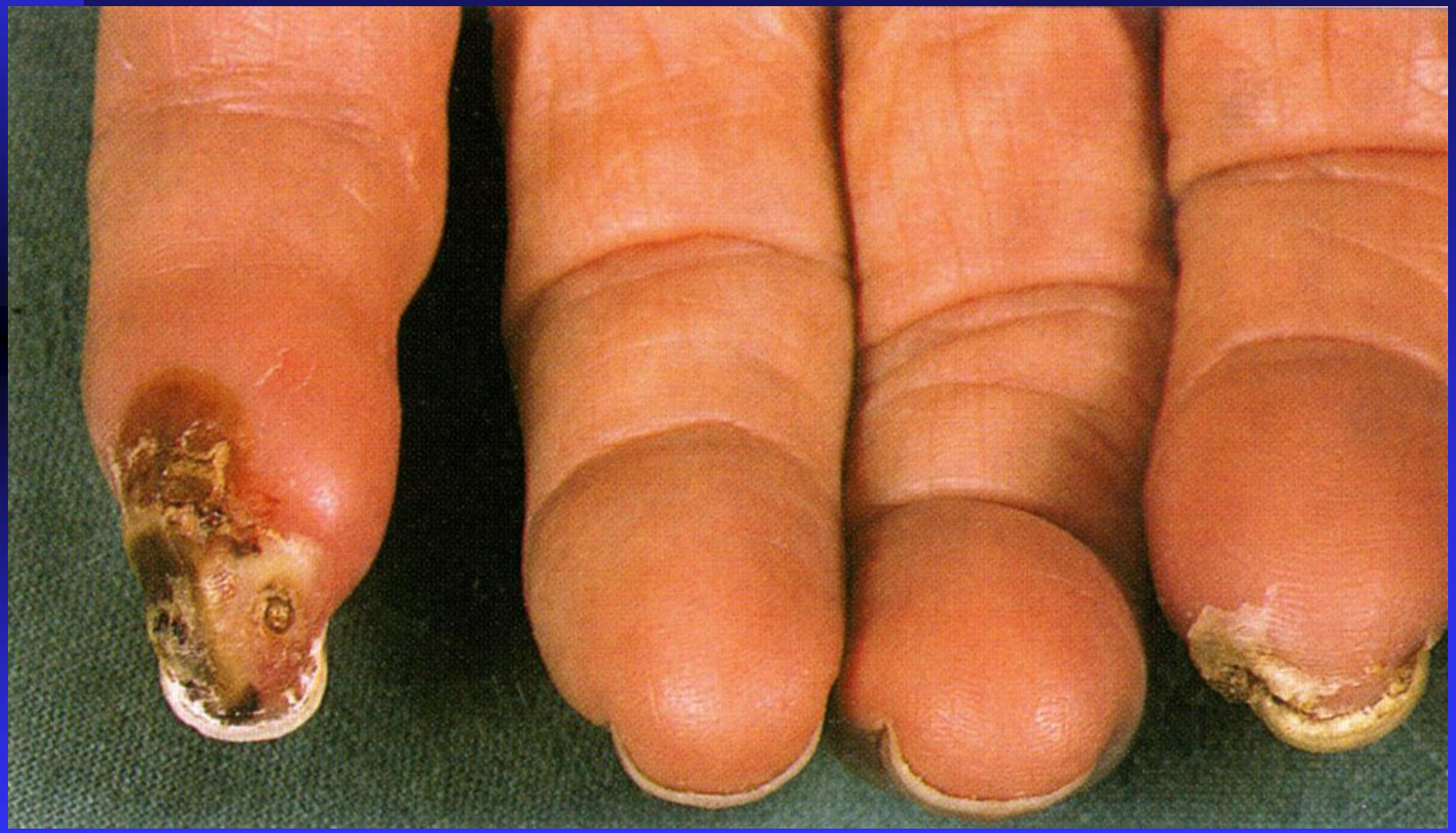
- Allogeneic HSC transplantation

Essential trombocytaemia

- Hyperplasia of megakaryocytic line with platelets elevation (over 1000x 10e9/l, i 3000)
- Platelets are functionally impaired, together with coagulation factors consumption, →
- in parallel bleeding and thrombotic complications

Ethiology:

- enhanced production of thrombopoetin
- defect of TPO receptor



Diferential diagnosis of thrombocythaemia?

- Infection
- Bleeding
- Iron deficiency
- Malignancy
- MDS- 5q-

- Chronic myeloid leukemia (CML, Ph+)
- Polycythaemia vera (PV)
- Prefibrotic stage of idiopatic myelofibrosis (PMF)

Treatment approach

- Acetylsalicylic acid if not bleeding
- Keeping platelets below 1000 x 10e9/l

- Anagrelide
- Interferon
- Hydroxyurea

- Thrombocytopheresis

TABLE I. The 2008 World Health Organization Diagnostic Criteria for PV, ET, and PMF [2]

	PV[a]	ET[a]	PMF[a]
Major criteria	(1) Hgb > 18.5 g/dL (men) > 16.5 g/dL (women) or Hgb > 17 g/dL (men), or > 15 g/dL (women) if associated with a sustained increase of ≥2 g/dL from baseline that can not be attributed to correction of iron deficiency or[c]	(1) Platelet count ≥450 × 10^9/L	(1) Megakaryocyte proliferation and atypia[b] accompanied by either reticulin and/or collagen fibrosis, or in the absence of reticulin fibrosis, the megakaryocyte changes must be accompanied by increased marrow cellularity, granulocytic proliferation and often decreased erythropoiesis (i.e. pre-fibrotic PMF)
	(2) Presence of JAK2V617F or similar mutation	(2) Megakaryocyte proliferation with large and mature morphology. No or little granulocyte or erythroid proliferation	(2) Not meeting WHO criteria for CML, PV, MDS, or other myeloid neoplasm
		(3) Not meeting WHO criteria for CML, PV, PMF, MDS, or other myeloid neoplasm	(3) Demonstration of JAK2V617F or other clonal marker or no evidence of reactive marrow fibrosis
		(4) Demonstration of JAK2V617F or other clonal marker or no evidence of reactive thrombocytosis	
Minor criteria	(1) BM trilineage myeloproliferation (2) Subnormal serum Epo level (3) EEC growth		(1) Leukoerythroblastosis (2) Increased serum LDH (3) Anemia (4) Palpable splenomegaly

Hgb, hemoglobin; Hct, hematocrit; Epo, erythropoietin; EEC, endogenous erythroid colony; WHO, World Health Organization; CML, chronic myelogenous leukemia; MDS, myelodysplastic syndrome; LDH, lactate dehydrogenase.

[a]Diagnosis of PV requires meeting either both major criteria and one minor criterion or the first major criterion and two minor criteria; diagnosis of ET requires meeting all four major criteria; diagnosis of PMF requires meeting all three major criteria and two minor criteria.

[b]Small to large megakaryocytes with aberrant nuclear/cytoplasmic ratio and hyperchromatic and irregularly folded nuclei and dense clustering.

[c]or Hgb or Hct >99th percentile of reference range for age, sex, or altitude of residence or red cell mass >25% above mean normal predicted.

MYELODYSPLASTIC SYNDROME

clonal disorder of hematopoiesis

HIT - leads to harm of stem cell → gene mutation

MUTATION → potential for proliferation of pathological clone of cells

CLONAL GROWTH ADVANTAGE → replacement of healthy marrow by pathological clones

Physiological protection

under standard conditions leads to elimination of atypical cells

Abnormal protective reaction may lead to overproduction of cytokines acting in a process of cellular death – apoptosis – with inadequate reaction of T lymphocytes finally helping the uncontrolled proliferation of pathological clone.

Myelodysplastic syndrome (MDS)

- ✓ ineffective hematopoiesis (peripheral cytopenia)
- ✓ dysplasia of one or more myeloid lineages

- ✓ cytogenetic abnormalities in 40-50% primary MDS
  - monosomy 5, 7, del 5q-, 7q-, trisomy 8

- ✓ older patients: 60 -70 years
- ✓ incidence in adults: 2-5/100 000/year
- ✓ rare in children: 1/200 000/year ?

- ✓ morbidity and mortality cause mainly by:
  infections, bleeding, leukemia transformation

MDS – clinical symptoms

- **anemia** – weakness, dyspnea, cardiac symptoms, collapse
- **neutropenia** – infection, fever
- **thrombocytopenia** – bleeding symptoms

(skin and mucosal, nasal, womb bleeding)

MDS - DIAGNOSTICS

Peripheral blood smear: cytopenia
Marrow aspirate/smear:
- **morphology** - dysplastic changes, % of blasts,…
- **cytochemistry**
- **cytogenetics** - numerical and structural changes
  - chromosomes 5,7,8, chromosomal aberations
- **flow cytometry** - % CD34+ precursors
- **in vitro cultures, molecular biology**

Trephine biopsy - cellularity, fibrosis,…

Biochemistry - iron, ferritin, B12, erythropoetin, bilirubine,
coag tests, CD 55- a CD59- erythrocytes,
tests of hemolysis

Physical examination – to rule-out secondary changes in blood caused by other primary disorder/disease

DIFFERENTIAL DIAGNOSIS in MDS

APLASTIC ANEMIA

PAROXYSMAL NOCTURNAL HEMOGLOBINURIA

ACUTE LEUKEMIA

MEGALOBLASTOID ANEMIA

SIDEROBLASTIC ANEMIA

… temporary dysplastic changes due to infection,…

MDS - prognosis

- …variable
- asymptomatic patients or slowly progressing anemia (transfusion dependence developing years after dg)
- aggressive course with rapidly progressing severe pancytopenia and/or transformation into acute leukemia

FAB classification of MDS

(Bennett et.al, Br J Haematol, 51, p.189, 1982)

FAB subtype	% blasts in BM	% ringed sideroblasts	other criteria
RA refractory anemia	< 5	< 15	< 1 % blasts in PB
RAS refractory anemia with increase of ringed sideroblasts	< 5	>15	< 1 % blasts in PB
RAEB refractory anemia with excess of blasts	5 - 20	different	< 5 % blasts in PB
CMML chronic myelomonocytic leukemia	1 - 20	different	monocytes in PB >1x10^9/L incr. % of monocytes in BM
RAEB-T refractory anemia with excess of blasts in transformation	21 - 30	different	> 5 % blasts in PB and < 30% blasts in BM

WHO CLASSIFICATION - MYELODYSPLASTIC SYNDROME

MYELODYSPLASTIC SYNDROME

REFRACTORY ANEMIA (RA)

REFRACTORY ANEMIA with RINGED SIDEROBLASTS (RARS)

REFRACTORY CYTOPENIA with MULTILINEAGE DYSPLASIA (RCMD)

REFRACTORY ANEMIA with EXCESS of BLASTS (RAEB)

- **RAEB I. (< 10% blasts)**
- **RAEB II. (> 10% blasts)**

5q- SYNDROME

MYELODYSPLASTIC SYNDROME UNCLASSIFIED

MYELODYSPLASTIC / MYELOPROLIFERATIVE SYNDROME

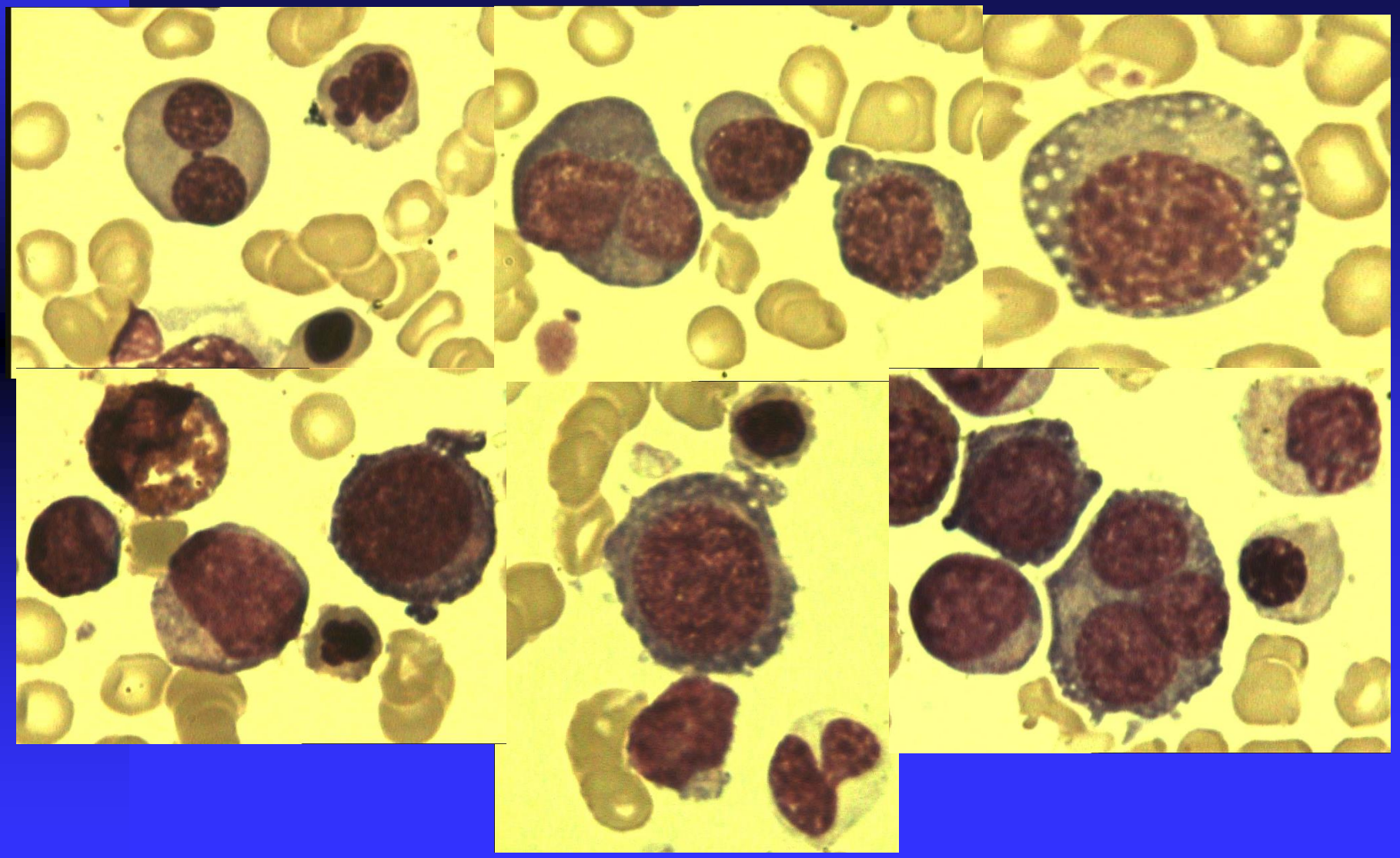
CHRONIC MYELOMONOCYTIC LEUKEMIA (CMML)

ATYPICAL CHRONIC MYELOID LEUKEMIA (aCML)

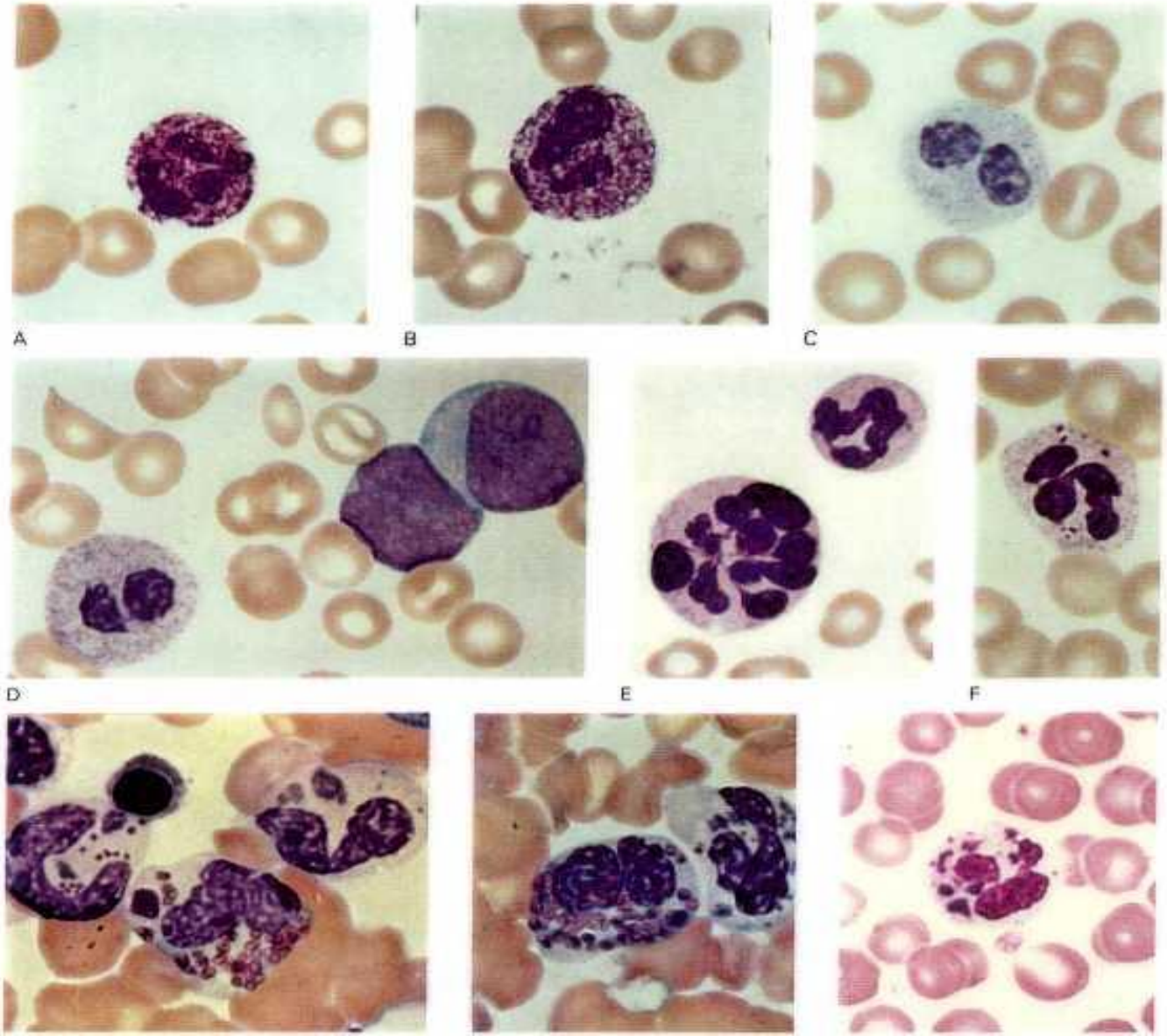
JUVENILE MYELOMONOCYTIC LEUKEMIA (JMML)

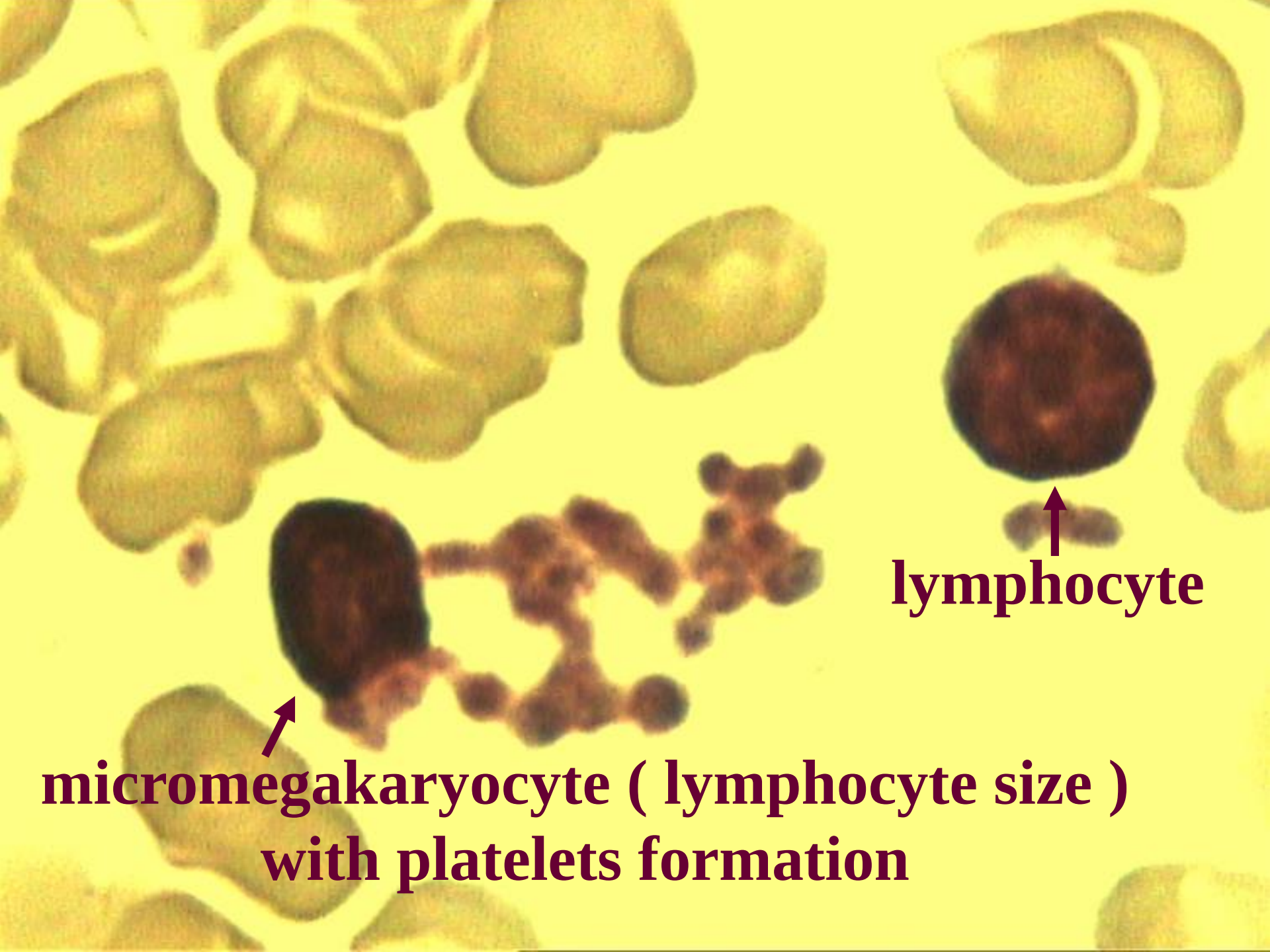
Harris NL, Jaffe ES et al., J Clin Oncol 1999, 17: 3835.

Dyserythropoiesis

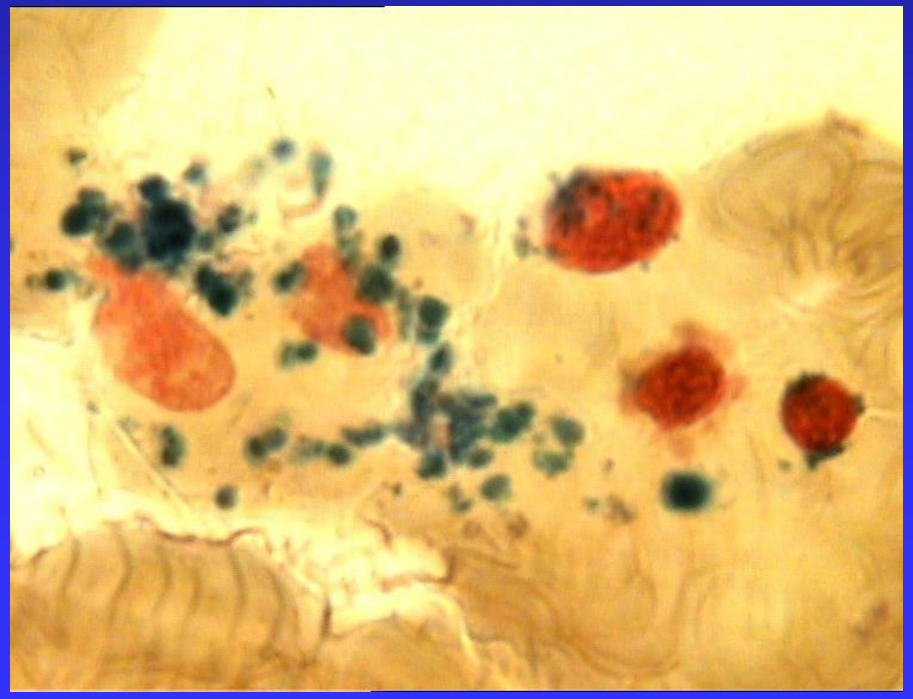


Dysmyelopoiesis




lymphocyte
micromegakaryocyte (lymphocyte size)
with platelets formation

Refractory anemia with ringed sideroblasts - RAS



MDS - therapy

- supportive: transfusions, antibiotics, chelation
- steroids: stimulation of release of blood cells from BM
  inhibition of apoptosis (low efficiency, side effects)
- growth factors (EPO, G-CSF, GM-CSF, IL-10)
  temporary effect, risky in advanced stages ?
- immunosuppressive drugs (CsA, ATG)
- **„low intensity“ chemotherapy** (hydroxyurea, melphalan, low-dose ara-C, VP-16, topotecan …)
- **combined aggressive chemotherapy:** younger patients, advanced stages (anthracyclines, ARA-C) for blast reduction
  limited effect, prolonged marrow aplasia, high toxicity/mortality

MDS - allogeneic HSCT

- **The only curative option in MDS**
- standard part of treatment protocols in younger pts
- the best results in matched donors (MUD, MSD)
- indication in severe cytopenia, transf. dependency, unfavorable karyotype

- **advanced forms of MDS**
- combined chemotherapy before HSCT
  decreased risk of post-Tx relapse, incr.risk of TRM
  myeloablative (MAC) or reduced intensity (RIC)

MDS-EB, BuCyMel^{BMBS}

Survival, Relapse and Transplant-Related Mortality