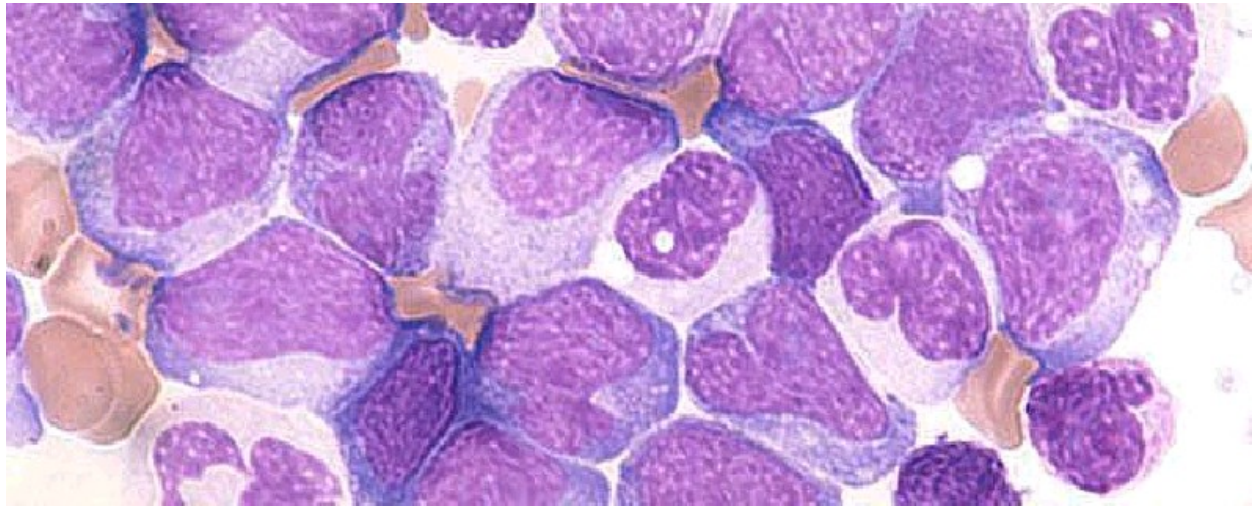
HEMATOLOGIC MALIGNANCIES



Interní hematologická a onkologická klinika
FN Brno a LF MU
Department of Internal Medicine, Hematology and Oncology,
University Hospital Brno
and Masaryk University, School of Medicine

MUNI MED

FAKULTNÍ NEMOCNICE BRNO

the CzEch leukemia study group for life

CEITEC

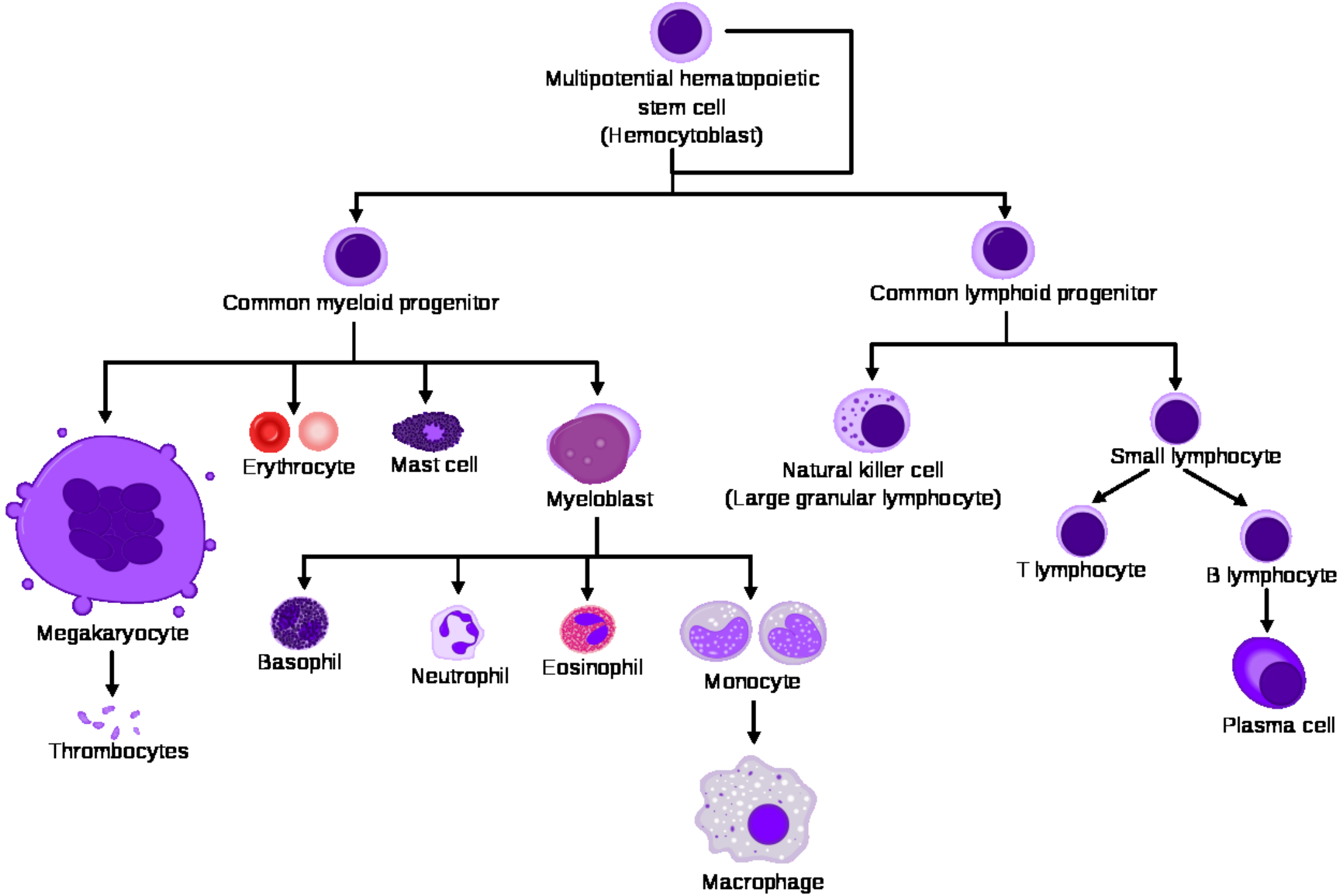
Hematologic malignancies

Origin – hematopoietic cells

According to blood lineage
- lymphoid malignancies
- myeloid malignancies

Diseases

- **Leukemias**
- **Lymphomas a lymphproliferative diseases**
- **Myeloproliferative diseases, myelodysplastic syndromes**


Multipotential hematopoietic
stem cell
(Hemocytoblast)
Common myeloid progenitor
Common lymphoid progenitor
Erythrocyte
Mast cell
Myeloblast
Natural killer cell
(Large granular lymphocyte)
Small lymphocyte
Megakaryocyte
Basophil
Neutrophil
Eosinophil
Monocyte
T lymphocyte
B lymphocyte
Thrombocytes
Plasma cell
Macrophage

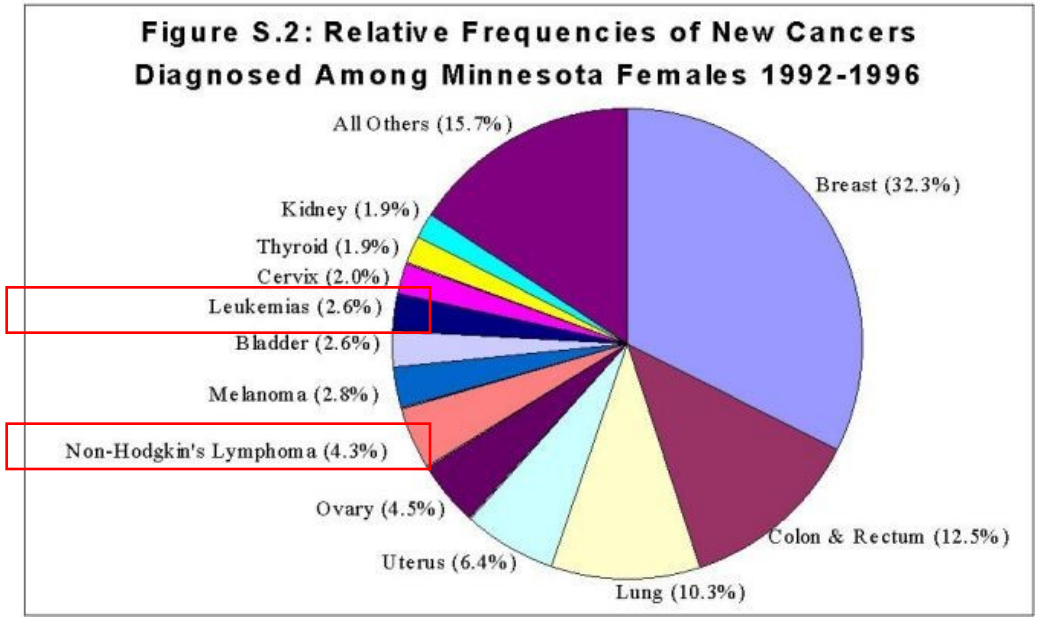


Figure S.2: Relative Frequencies of New Cancers
Diagnosed Among Minnesota Females 1992-1996
All Others (15.7%)
Breast (32.3%)
Kidney (1.9%)
Thyroid (1.9%)
Cervix (2.0%)
Leukemias (2.6%)
Bladder (2.6%)
Melanoma (2.8%)
Non-Hodgkin's Lymphoma (4.3%)
Ovary (4.5%)
Uterus (6.4%)
Lung (10.3%)
Colon & Rectum (12.5%)

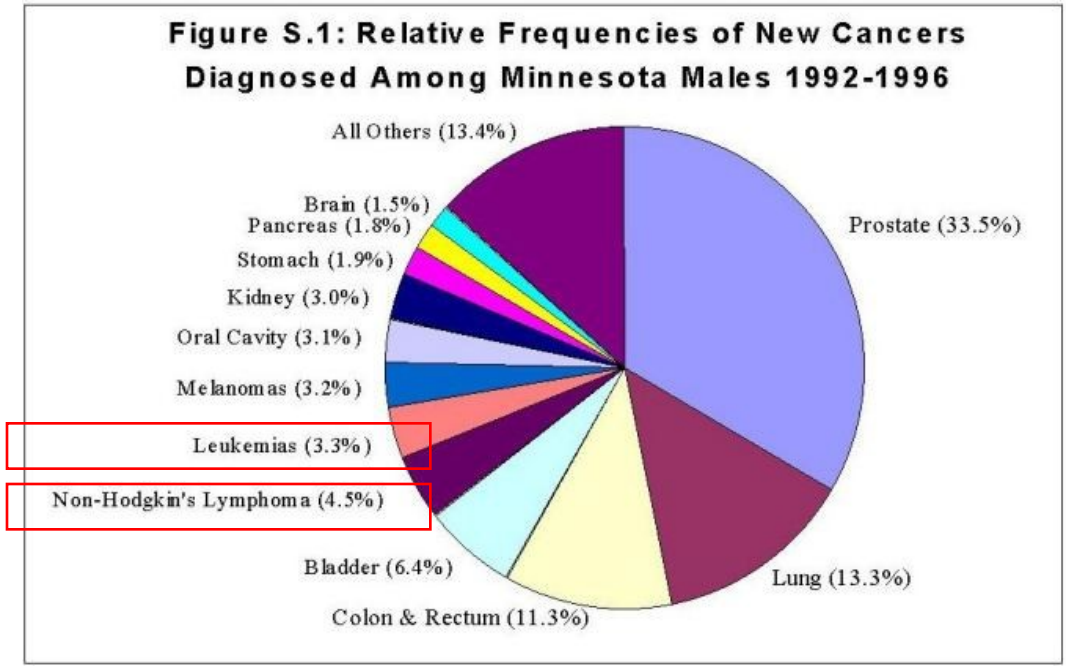


Figure S.1: Relative Frequencies of New Cancers
Diagnosed Among Minnesota Males 1992-1996
All Others (13.4%)
Brain (1.5%)
Pancreas (1.8%)
Stomach (1.9%)
Kidney (3.0%)
Oral Cavity (3.1%)
Melanomas (3.2%)
Leukemias (3.3%)
Non-Hodgkin's Lymphoma (4.5%)
Bladder (6.4%)
Colon & Rectum (11.3%)
Lung (13.3%)
Prostate (33.5%)

Hematologic malignancies

CLONAL
disorders resulting from a mutation of DNA within a pluripotent marrow stem cell or very early progenitor cell.

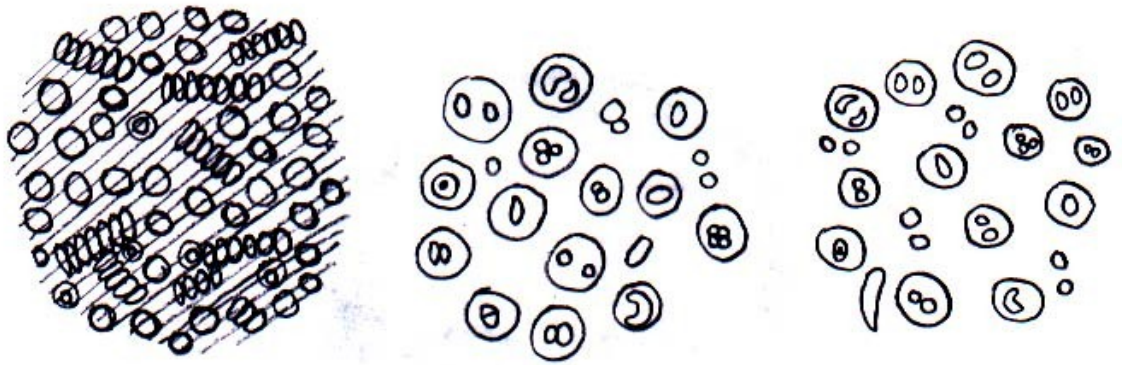
CLONAL POPULATION OF CELLS - cells with growth and/or proliferation advantage over against normal bone marrow cells.

Mutation of DNA can result in the expression of fusion genes that encode fusion proteins that are oncogenic or in the underexpression of genes that encode molecules critical to control of cell growth or progremmed cell death.

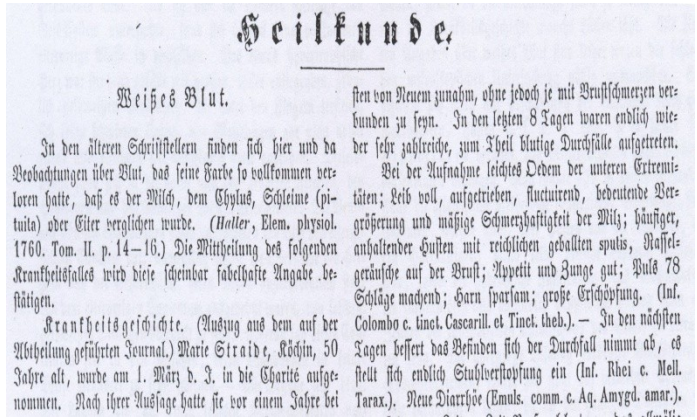
Symptoms

- Similar
- Often non-specific
- Bone marrow involvement (leukemia, myeloproliferative neoplasms)
- Lymphoid tissue involvement (lymphoma)

Leukemia



John Hughes Bennett: Two Cases of Disease and Enlargement of the Spleen, in which death took place from presence of purulent matter in the blood, 1845



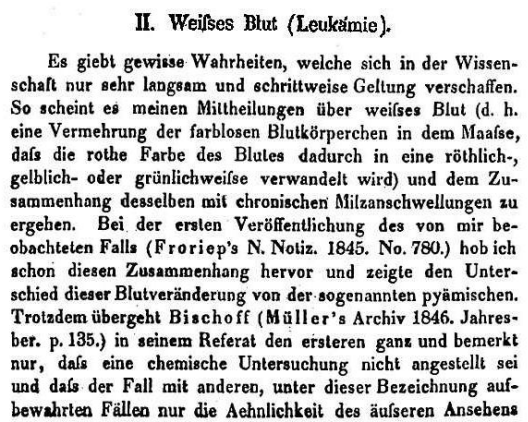
Heilkunde.

Weißes Blut.

In den älteren Schriftstellern finden sich hier und da Beobachtungen über Blut, das seine Farbe so vollkommen verloren hatte, daß es der Milch, dem Chylus, Schleime (pituita) oder Eiter verglichen wurde. (*Haller*, Elem. physiol. 1760. Tom. II. p. 14–16.) Die Mittheilung des folgenden Krankheitsfalles wird diese scheinbar fabelhafte Angabe bestätigen.

Krankheitsgeschichte. (Auszug aus dem auf der Abtheilung geführten Journal.) Marie Straide, Köchin, 50 Jahre alt, wurde am 1. März d. J. in die Charité aufgenommen. Nach ihrer Aussage hatte sie vor einem Jahre bei ften von Neuem zunahm, ohne jedoch je mit Brustschmerzen verbunden zu seyn. In den letzten 8 Tagen waren endlich wieder sehr zahlreiche, zum Theil blutige Durchfälle aufgetreten.

Bei der Aufnahme leichtes Oedem der unteren Extremitäten; Leib voll, aufgetrieben, fluctuirend, bedeutende Vergrößerung und mäßige Schmerzhaftigkeit der Milz; häufiger, anhaltender Husten mit reichlichen geballten sputis, Rasselgeräusche auf der Brust; Appetit und Zunge gut; Puls 78 Schläge machend; Harn sparsam; große Erschöpfung. (Inf. Colombo c. tinct. Cascarill. et Tinct. theb.). — In den nächsten Tagen bessert das Befinden sich der Durchfall nimmt ab, es stellt sich endlich Stuhlverstopfung ein (Inf. Rhei c. Mell. Tarax.). Neue Diarrhöe (Emuls. comm. c. Aq. Amygd. amar.).



II. Weißes Blut (Leukämie).

Es giebt gewisse Wahrheiten, welche sich in der Wissenschaft nur sehr langsam und schrittweise Geltung verschaffen. So scheint es meinen Mittheilungen über weißes Blut (d. h. eine Vermehrung der farblosen Blutkörperchen in dem Maafse, daſs die rothe Farbe des Blutes dadurch in eine röthlich-, gelblich- oder grünlichweiſse verwandelt wird) und dem Zusammenhang desselben mit chronischen Milzanschwellungen zu ergehen. Bei der ersten Veröffentlichung des von mir beobachteten Falls (Froriep's N. Notiz. 1845. No. 780.) hob ich schon diesen Zusammenhang hervor und zeigte den Unterschied dieser Blutveränderung von der sogenannten pyämischen. Trotzdem übergeht Bischoff (Müller's Archiv 1846. Jahresber. p. 135.) in seinem Referat den ersteren ganz und bemerkt nur, daſs eine chemische Untersuchung nicht angestellt sei und daſs der Fall mit anderen, unter dieser Bezeichnung aufbewahrten Fällen nur die Aehnlichkeit des äuſseren Ansehens

Rudolf Virchow: Weisses Blut. Frorieps Notizen, 36, s. 152 – 156, 1845

What leukemias are?

- Very different diseases
- Historical name: accumulation of white blood cells
- Not every accumulation of white blood cells is leukemia
  - Leukemoid reaction
  - Lymphoma leukemization
- Acute or Chronic
- Myeloid or Lymphoid

Common features of leukemia

- White blood cells accumulation
  - precursor cells (myelo-, lympho-) – blasts – acute leukemia
  - myeloid lineage cells – CML
  - mature lymphocytes (CLL)
- Leukocytosis (CML, CLL, AL)
- Normal WBC or leukopenia (AL)
- In almost all cases pathology in differential white blood count
- In all cases bone marrow involvement

LEUKEMIAS

Do you know differences between acute and chronic leukemias?

Briefly:

Acute leukemia - there is defect of proliferation, proliferation of young bone marrow cells (blasts) is increased!

Chronic leukemia - there is defect of apoptosis (programmed cell death), apoptosis of mature cells is decreased, mature cells are accumulated in the body!

CAVE: CL can switch to AL (CML in blast crisis, CLL in Richter s syndrome)

LEUKEMIA INCIDENCE

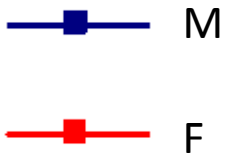
12,7/100 000 M 9,8/100 000 F

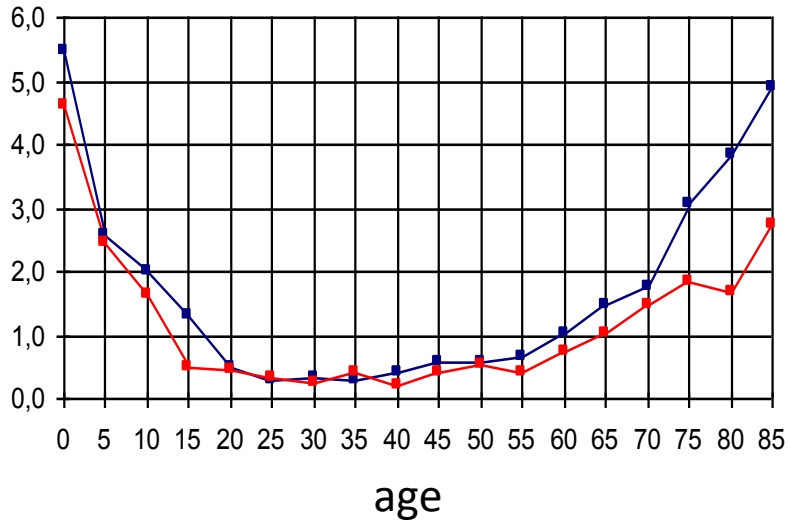
Slightly increasing incidence compared with 90´s (except of CML)

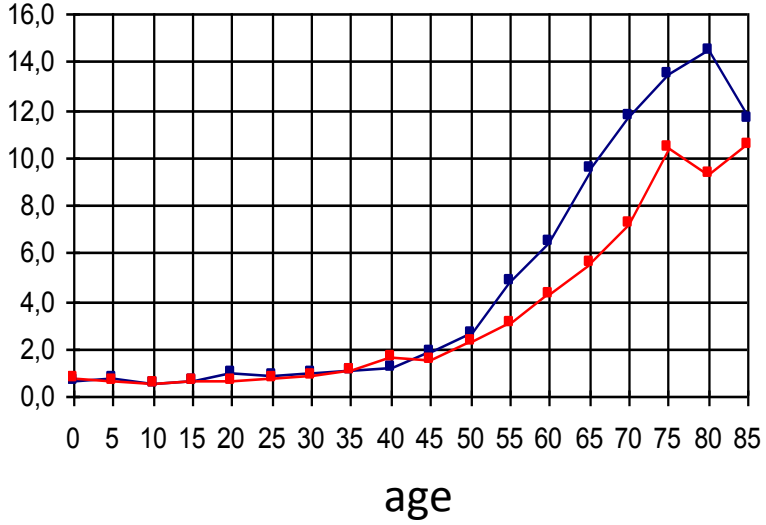
Europe:
40% CLL, 25% AML, 15% CML, 11% ALL,
2% HCL, 7% other

<u>Myelodysplastic syndromes</u>
1 - 2/100 000 (older 10-20/100 000)

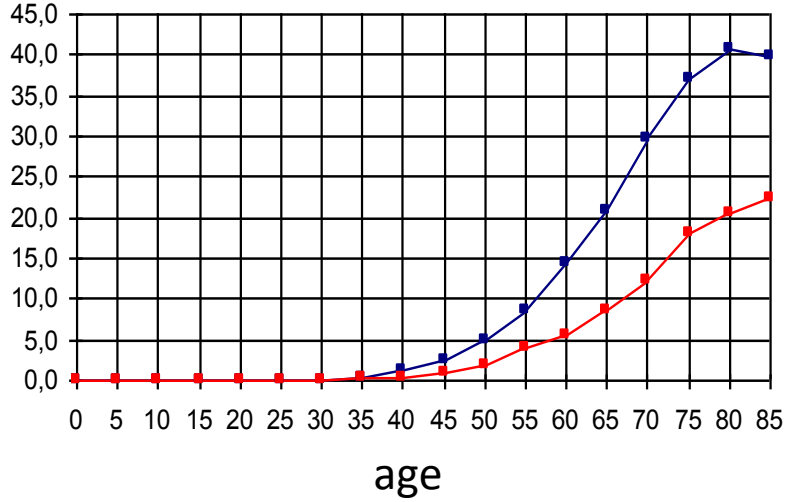
ALL cases /100 000 AML

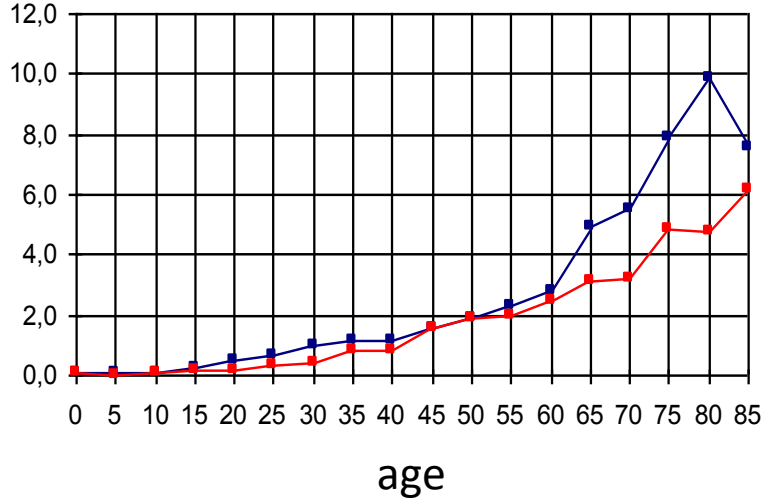




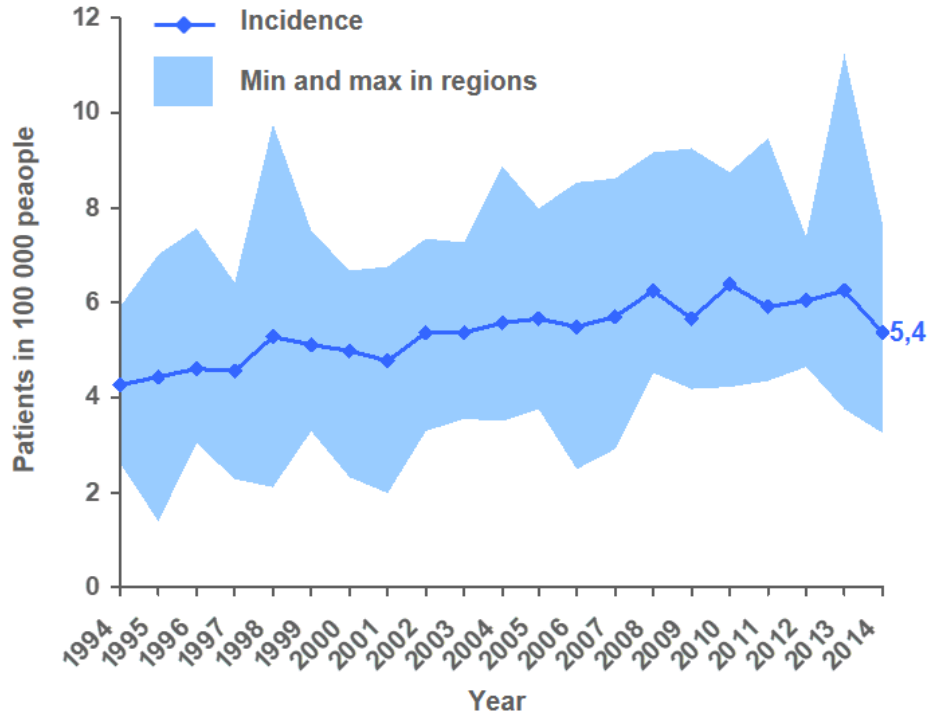


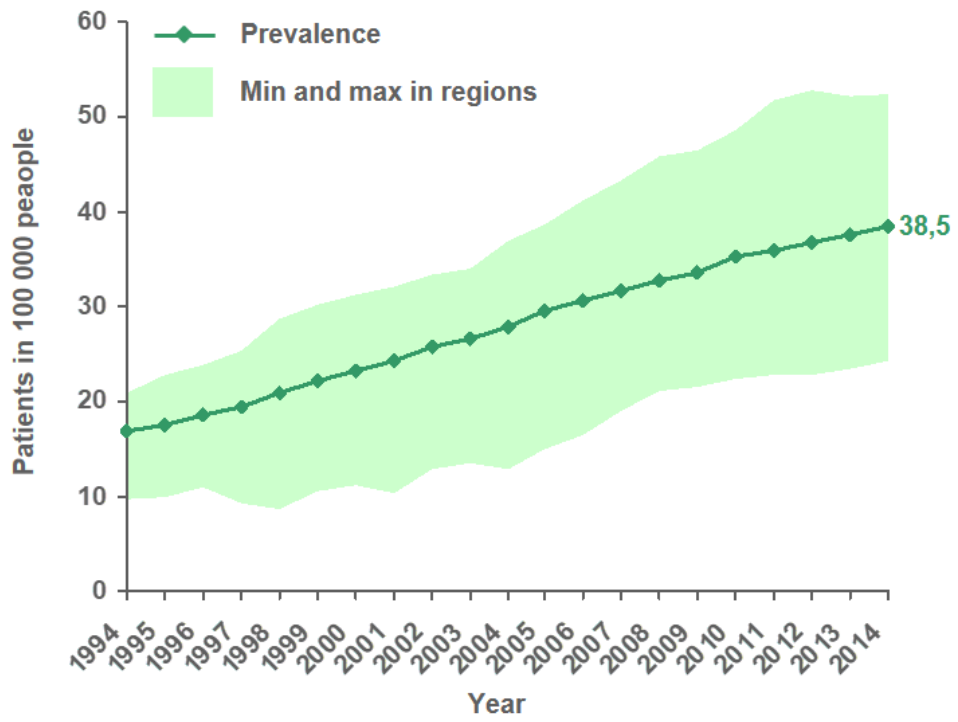
CLL CML





LEUKEMIA INCIDENCE AND PREVALENCE - CLL as example



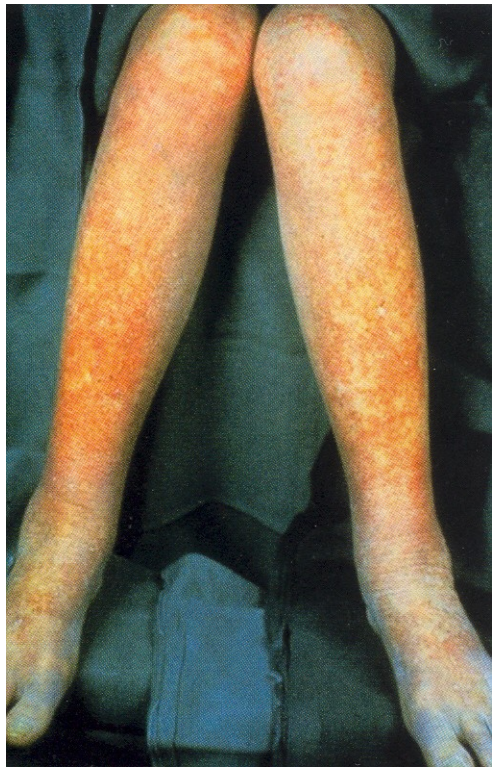


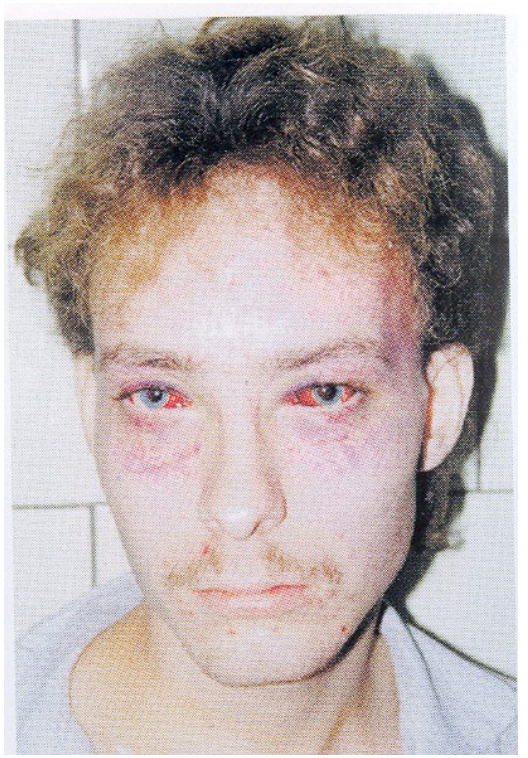
Clinical symptoms of malignant diseases of blood and bone marrow

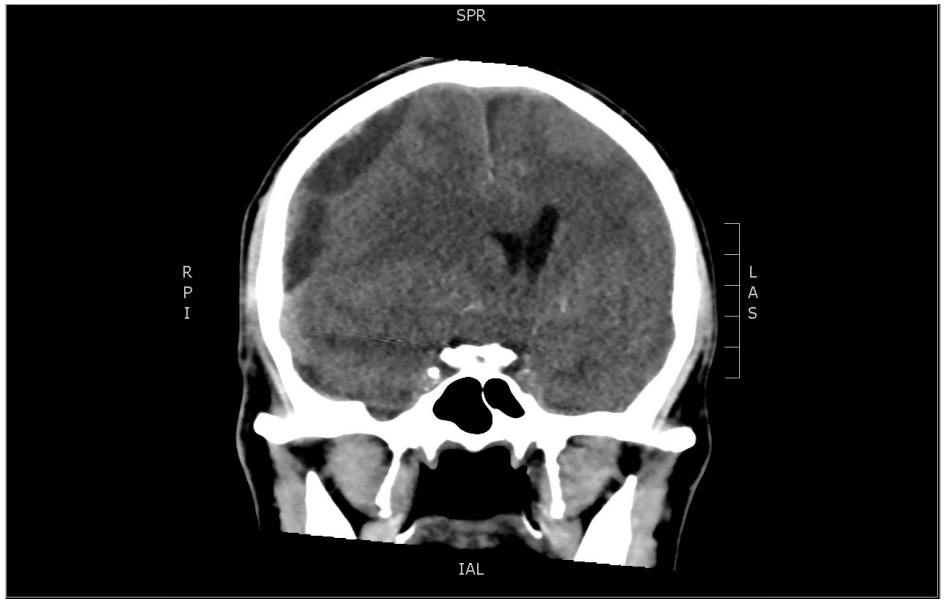
Symptoms affecting patients	Frequency
infection, fever	36 % (all)
bleeding	33 % (APL, AML)
thrombosis, DIC	10 % (APL, ET, PV)
lymph nodes enlargement	57 % (ALL, CLL)
splenomegaly	56 % (CML, CLL, PV, MF)
hepatomegaly	47 % (CML, AML)
mediastinal tumor	14 % (ALL, CLL)
CNS involvemet	7 % (ALL, AML M5)
involvement of another organs	9 % (all)

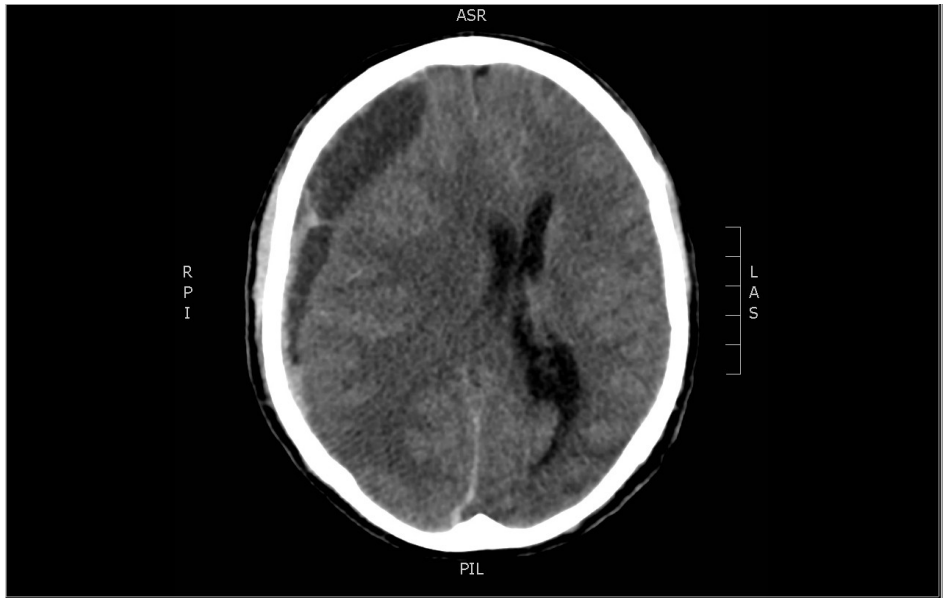
CAVE: All symptoms of hematologic diseases are non-specific!

Bleeding in
acute leukemia



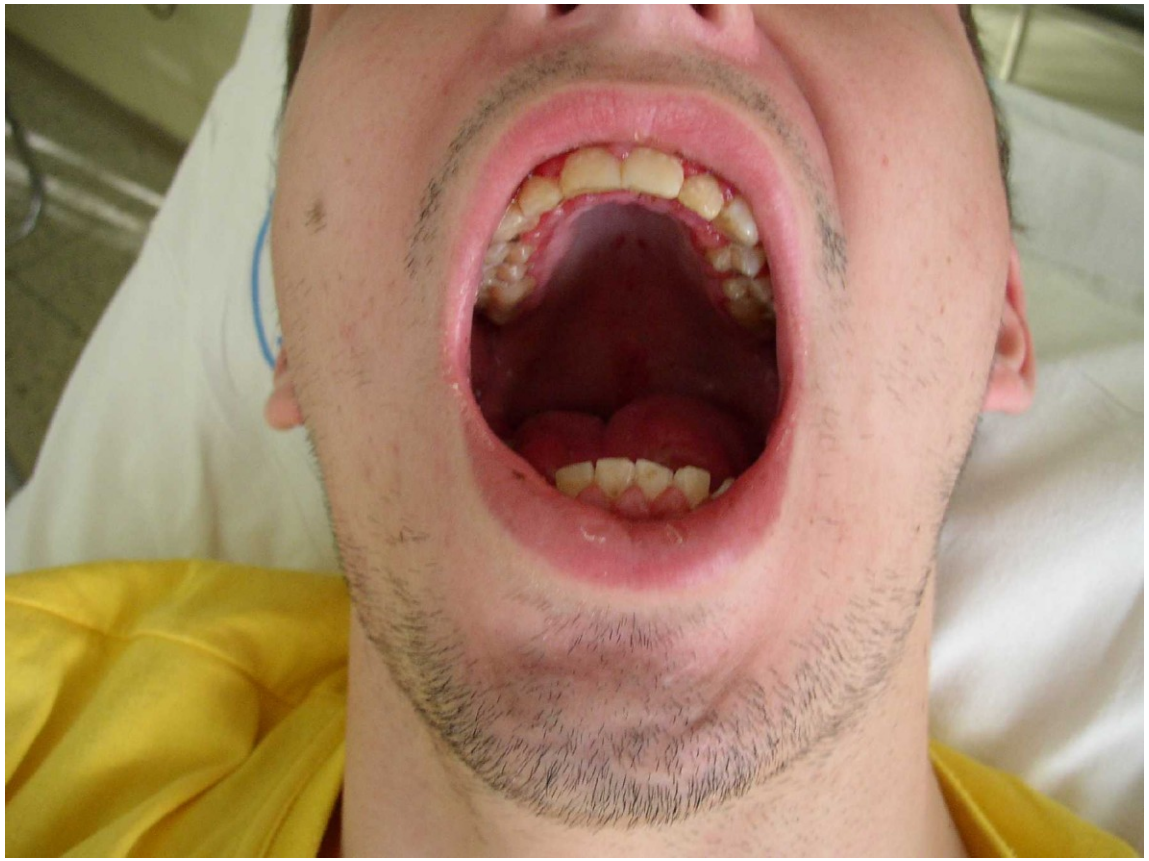




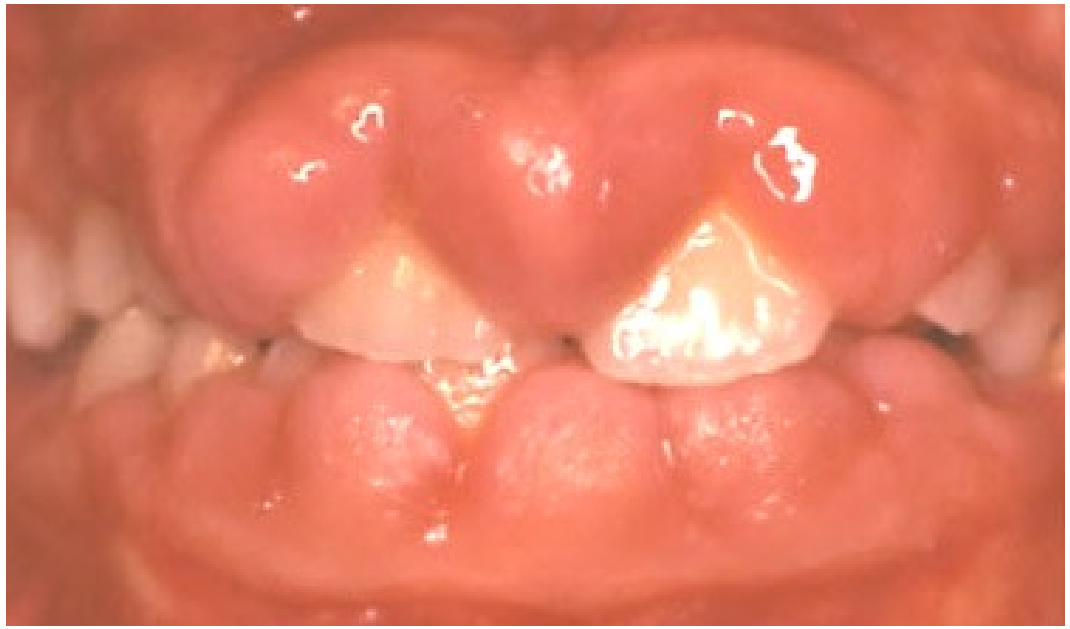


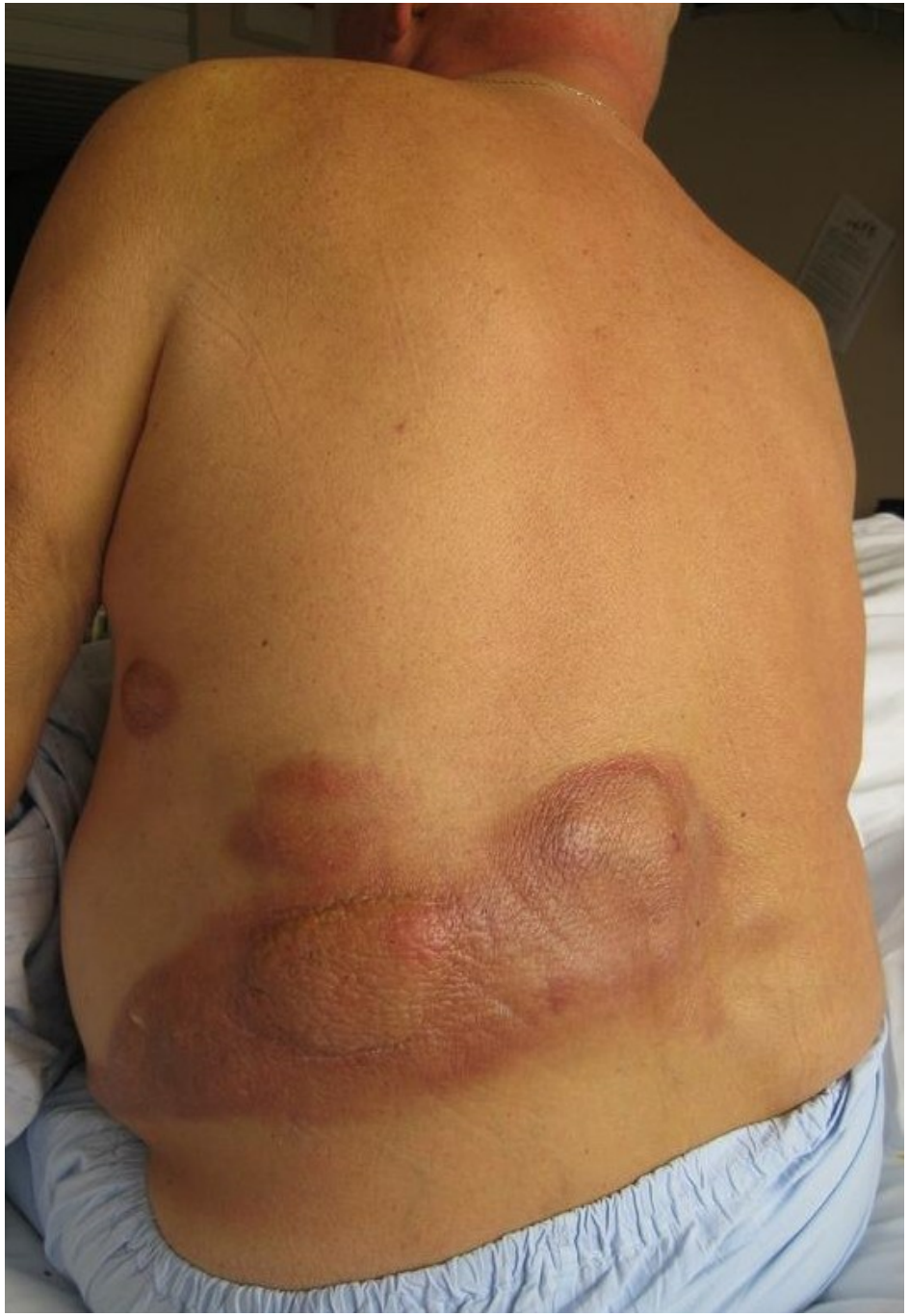
Bleeding in acute leukemia

AML – gum hyperplasia

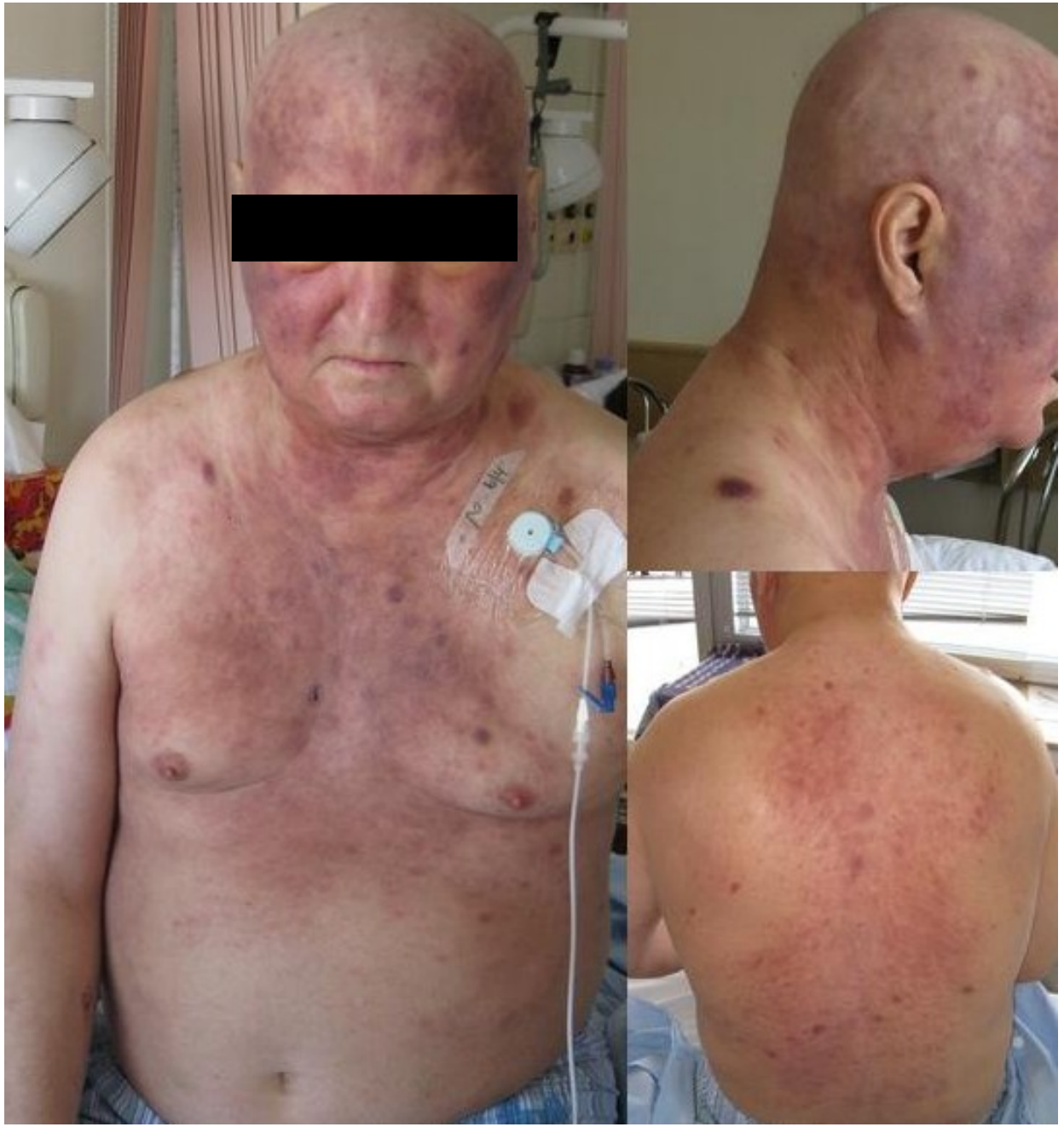


AML – gum hyperplasia

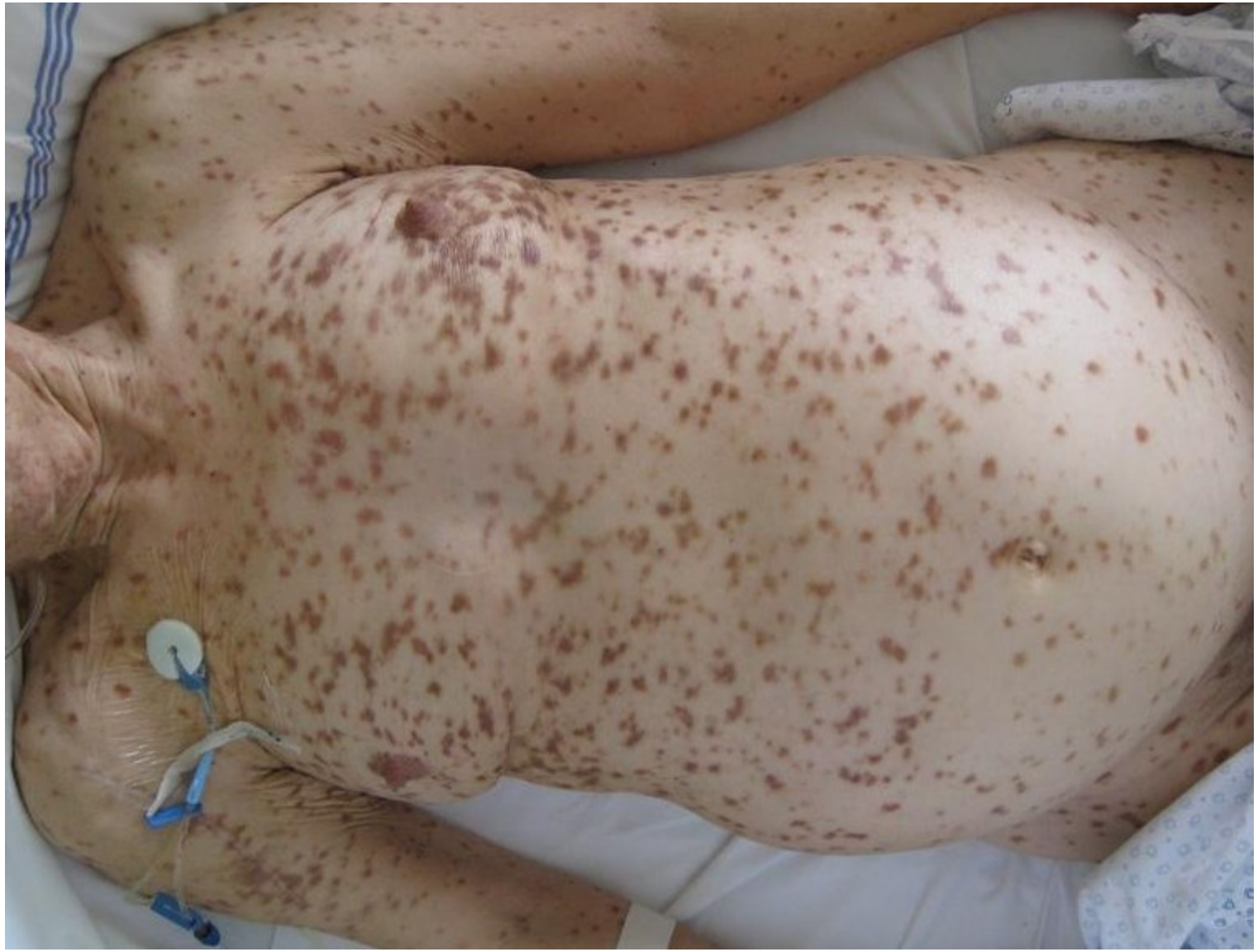




PLL – skin involvement



ALL – skin involvement



Mastocytosis
- *urticatia pigmentosa*

ON SOME

MORBID APPEARANCES

OF

THE ABSORBENT GLANDS

AND

SPLEEN.

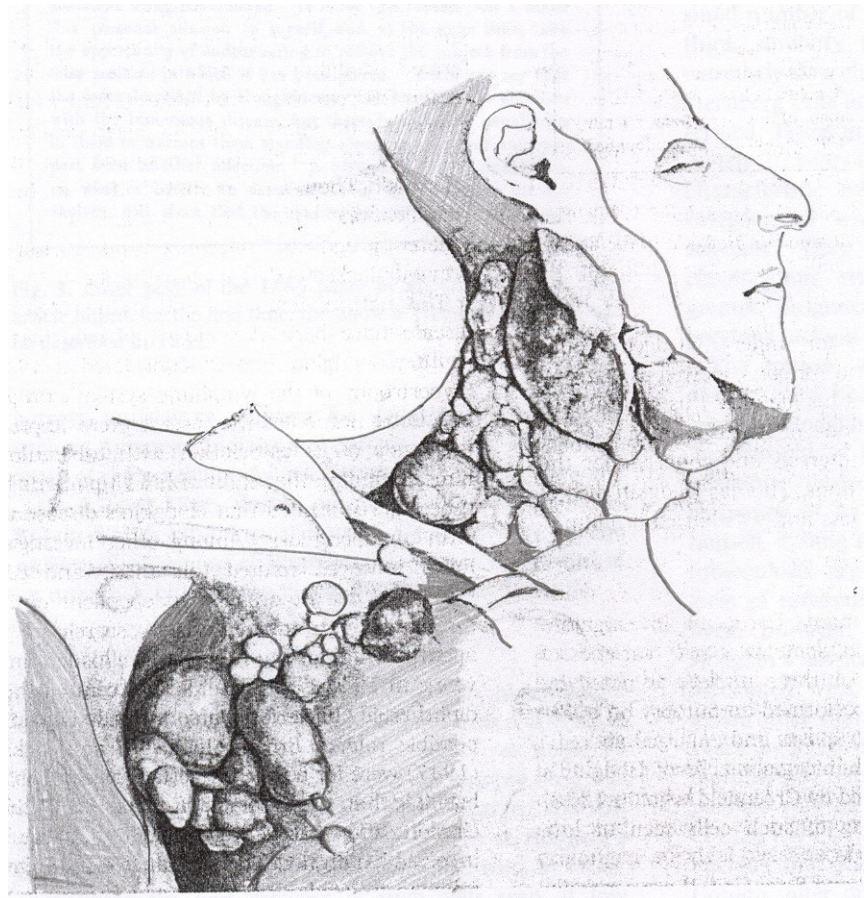
BY DR. HODGKIN.

PRESENTED

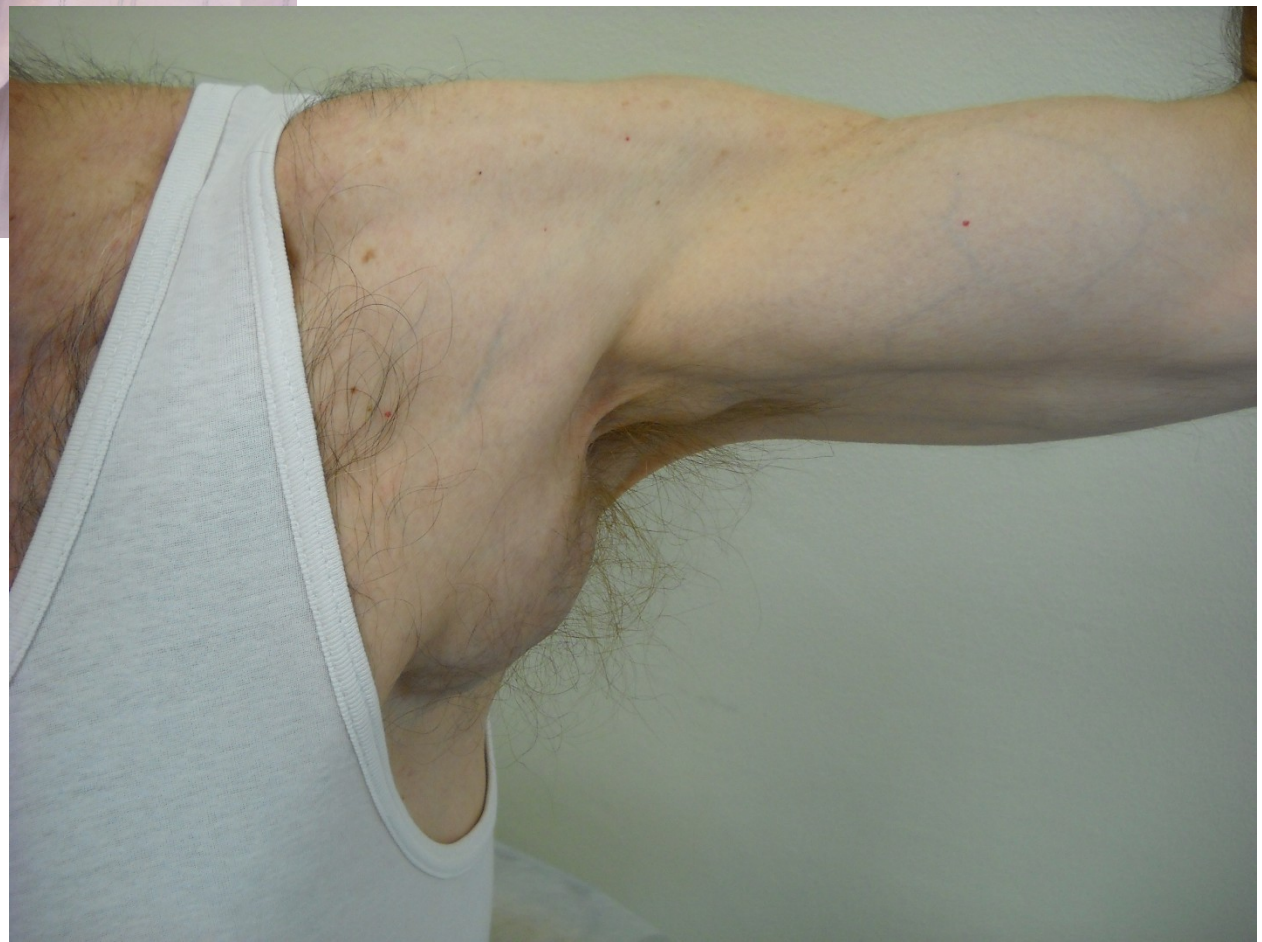
BY DR. R. LEE.

READ JANUARY 10TH AND 24TH, 1832.

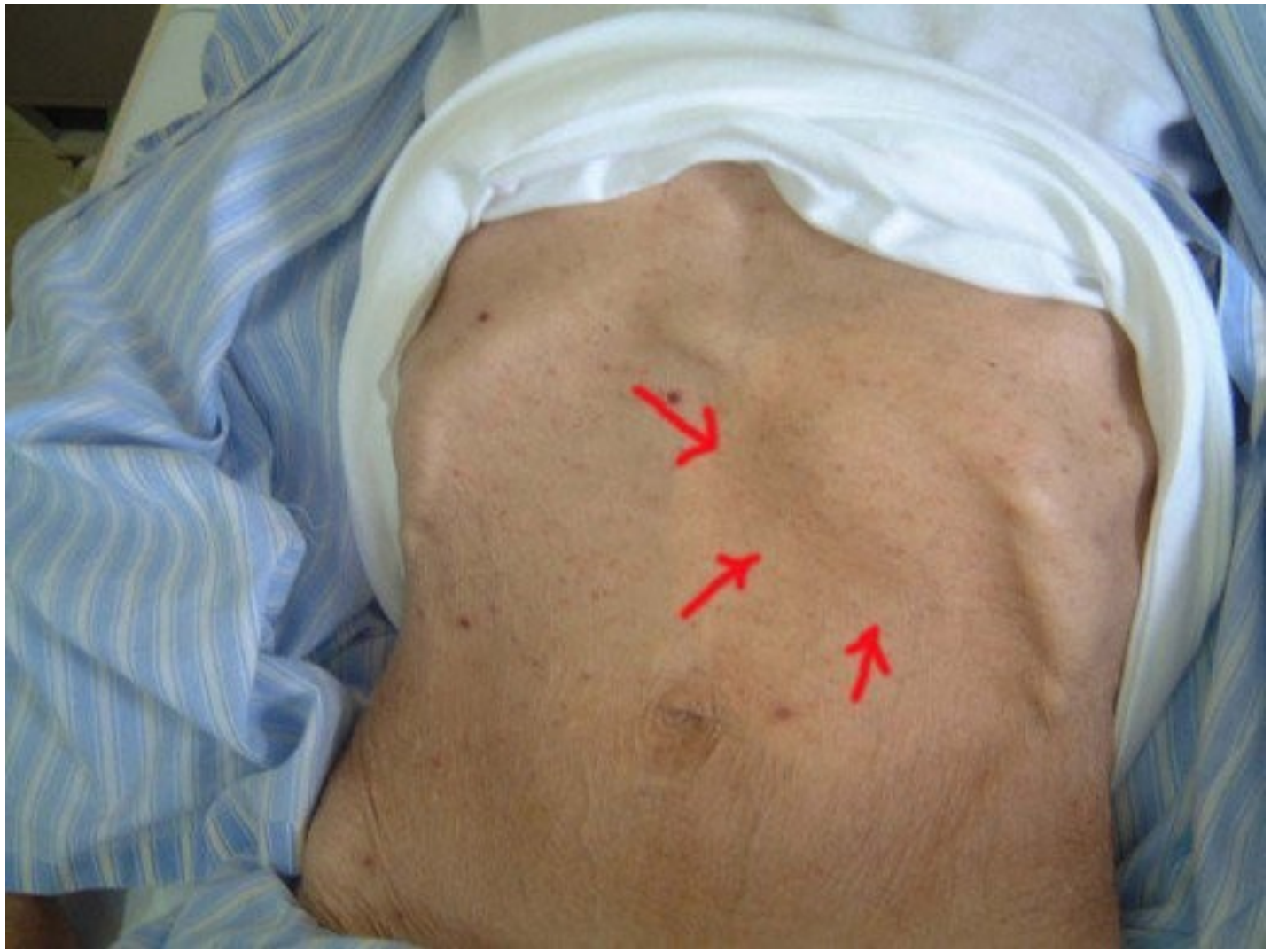
THE morbid alterations of structure which I am about to describe are probably familiar to many

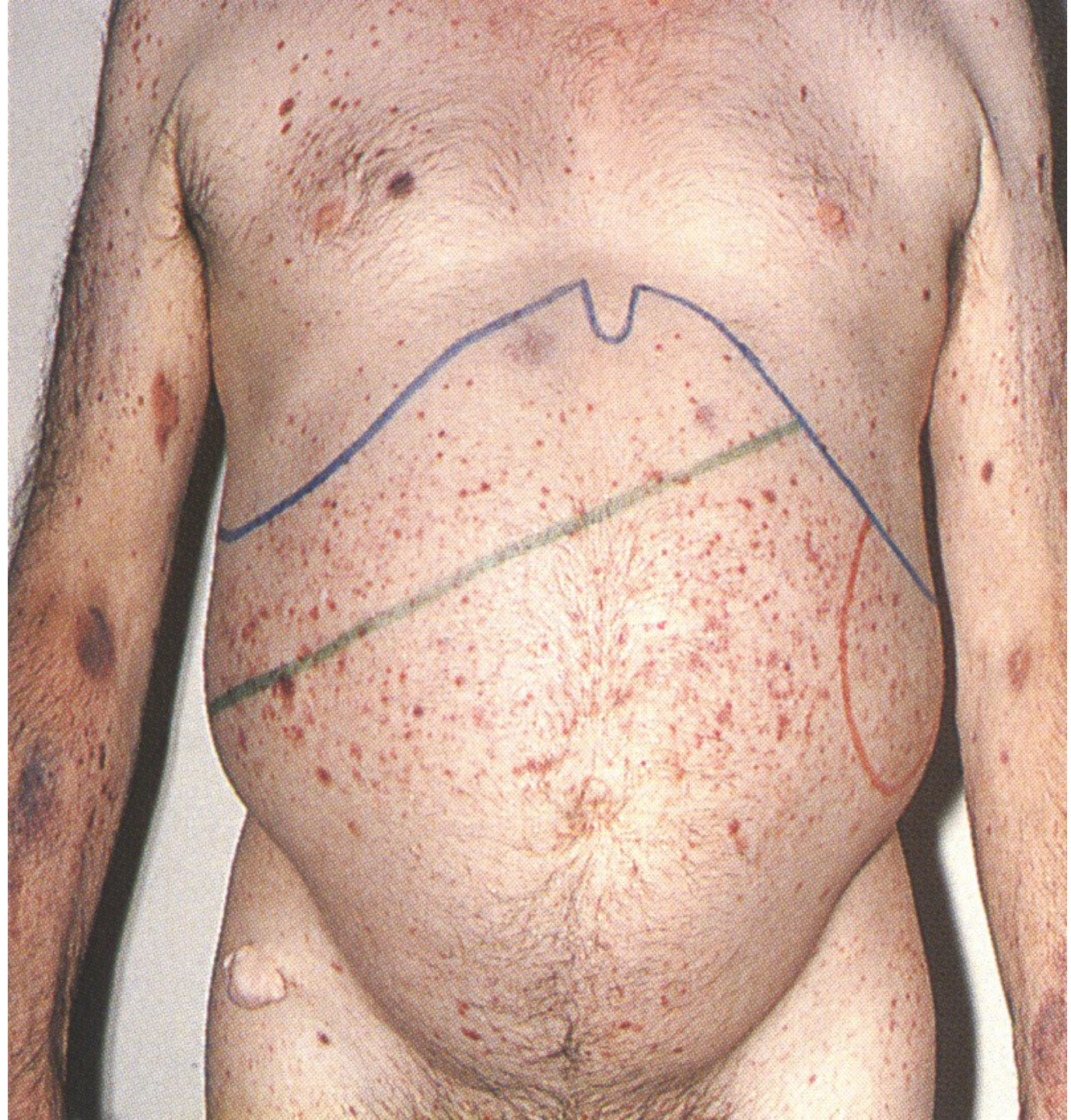


CLL
- lymph nodes



CLL - splenomegaly

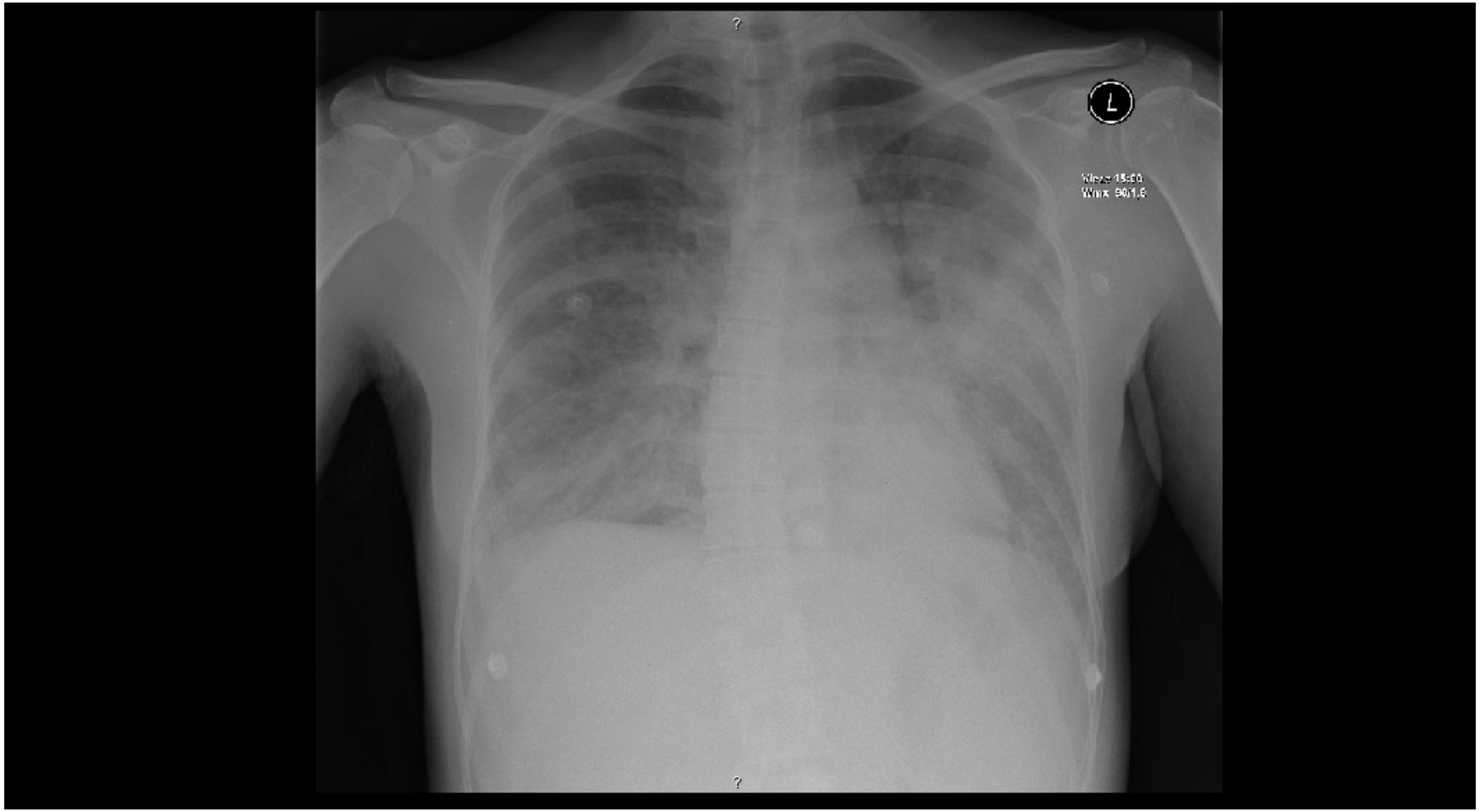




Myelofibrosis - massive splenomegaly

Lekocytes
- leukapheresis bag

Lung infiltration in acute leukemia



Time from first symptoms to final diagnosis

TABLE II. Time from the First Symptoms (Analysis Only Performed Among Patients that Presented Symptoms) and from the First Medical Visit to a Definitive Diagnosis

	ALL	AML	APL	CLL	CML	HCL	Acute leukemias	Chronic leukemias	Total
Time from the first symptoms to a definitive diagnosis[a]									
No. of analyzed pts.	90	305	59	125	68	22	454	215	669
Days—median (range)	25 (3–194)	22 (0–226)	14 (3–90)	27 (3–274)	21 (1–256)	34.5 (4–370)	21 (0–226)	27 (1–370)	22 (0–370)
Days—25–75% interval	14–43	12–36	8–22	14–52	11.5–48	14–77	12–35	13–60	12–42
Time from the first medical visit to a definitive diagnosis									
No. of analyzed pts.	106	366[b]	74	293	123	41	546	457	1003
Days—median (range)	9 (0–108)	7 (0–171)	5.5 (0–71)	12 (0–343)	6 (0–119)	20 (0–355)	7 (0–171)	10 (0–355)	8 (0–355)
Days—25–75% interval	3–16	3–16	2–12	4–22	2–16	8–36	3–15	3–23	3–19

LEUKEMIAS – PREDISPOSING FACTORS

Increased risk of leukemia is in:

Genetic syndromes – M. Down, FA, ataxia telangiectasia, inherited germline mutations (*ETV6, RUNX1, DDX41*…)

Drugs (chemotherapy, alkylating agents)

Radiation (can cause all leukemias except CLL)

Socioeconomic factors

(increased incidence of childhood ALL in industrial countries, probably due to later contact of children with alergens or banal childhood infections)

Viruses (EBV, HTLV I, HIV)

Benzene, toluene, etc.

LEUKEMIAS – ETIOLOGY

Somatic molecular leasions involving:

Cell proliferation
Cell division
Cell maturation
Apoptosis
Cell self-renewal

LEUKEMIAS – ETIOLOGY

The most important somatic molecular changes in leukemia and myeloproliferative neoplasms:

BCR-ABL
TP53
PML-RARa
JAK2

LEUKEMIAS AND MYELOPROLIFERATIVE DISEASES
Blood and bone marrow features

What can we found in periperal blood (WBC, RBC, platelets)?

- acute leukemia
- chronic leukemia
- myeloproliferative diseases

What can we found in bone marrow?

- acute leukemia
- chronic leukemia
- myeloproliferative diseases

Laboratory diagnostics

Peripheral blood count with differential WBC

Bone marrow

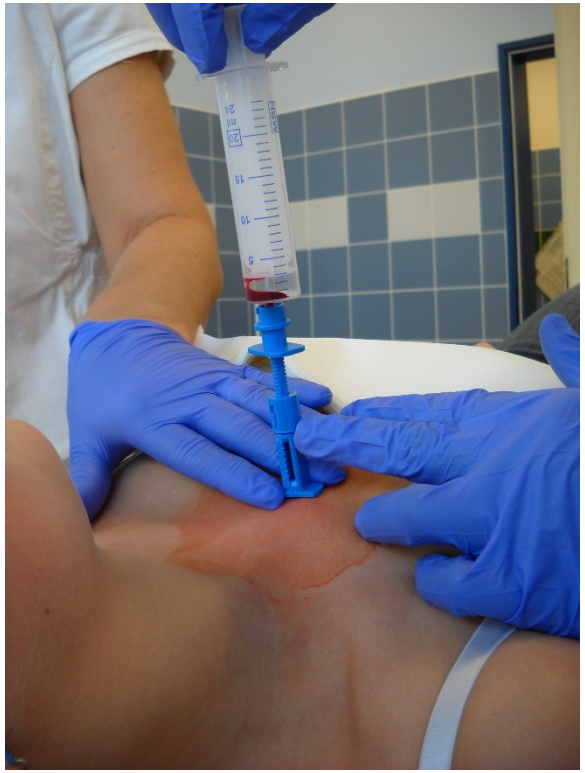
Flow cytometry (analysis of CD antigens)
(ALL, CLL, HCL)
Cytogenetic analysis (CML, AL, MDS, CLL)
Molecular genetic analysis (CML, APL, AL, CLL)
Cytology a cytochemistry
Histology (necessary in myeloproliferative diseases)

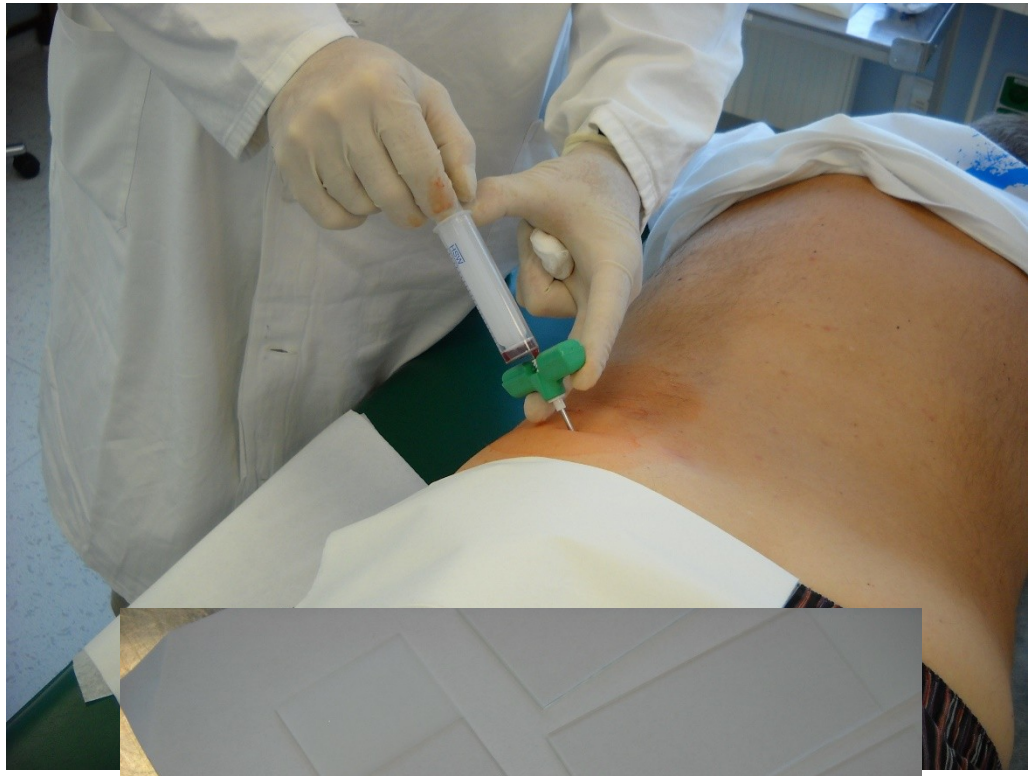
Do you know differences between trephine biopsy and sternal puncture?
Sternal puncure - we can collect only marrow blood. SP fits for diagnostics of leukemias.

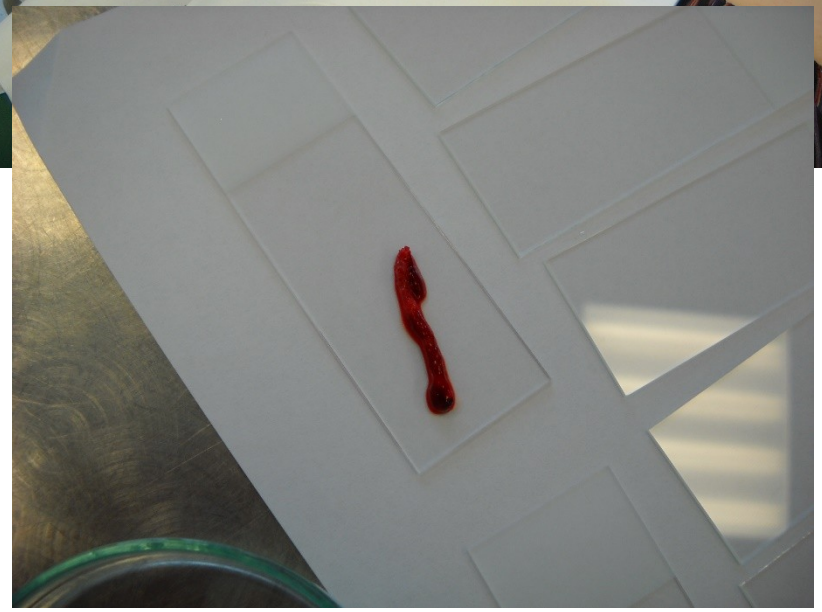
VÝSLEDEK VYŠETŘENÍ Z HEMATOLOGIE

Pacient: [redacted]
Datum a čas odběru: [redacted]

Vyšetření	Hodn.	Výsl.	Jedn.	Meze/koment.
Leukocyty	<.>	8.75	x10 9/l	(4 - 10)
Erytrocyty	<.>	3.80	x10 12/l	(3.8 - 5.4)
Hemoglobin	<L>	115.0	g/l	(120 - 160)
Hematokrit	<L>	0.320	l/l	(0.35 - 0.46)
Střední objem ERY	<L>	83.6	fL	(84 - 96)
Trombocyty	<.>	163.0	x10 9/l	(150 - 350)
Stř. množství HGB v	<.>	30.2	pg	(28 - 34)
Prům. koncentrace HG	<.>	362.0	g/l	(320 - 370)
Šíře distribuce ERY	<.>	13.5	%	(10 - 15.2)
Střední objem trombo	<L>	7.05	fl	(7.8 - 11)
Trombocytový hematok	<L>	1.15	ml/l	(1.21 - 3.5)
Šíře distribuce trom	<.>	17.00	%	(15.5 - 17.1)
Neutrofily %	<L>	19.80	%	(50 - 70)
Lymfocyty %	<.>	22.40	%	(20 - 40)
Monocyty %	<H>	57.00	%	(2 - 12)
Eosinofily %	<.>	0.28	%	(0 - 5)
Basofily %	<.>	0.60	%	(0 - 1)
Neutrofily (absolutn	<L>	1.73	x10 9/l	(2 - 7)
Lymfocyty (absolutní	<.>	1.96	x10 9/l	(0.8 - 4)
Monocyty (absolutní	<H>	4.99	x10 9/l	(0.08 - 1.2)
Eosinofily (absolutn	<.>	0.02	x10 9/l	(0 - 0.5)
Basofily (absolutní	<.>	0.05	x10 9/l	(0.01 - 0.1)
Neutrofily mikroskop	<L>	20.0	%	(50 - 70)
Tyče mikroskopicky	<.>	0.0	%	(0 - 4)
Lymfocyty mikroskopi	<.>	23.0	%	(20 - 40)
Monocyty mikroskopic	<.>	2.0	%	(2 - 12)
Eosinofily mikroskop	<.>	0.0	%	(0 - 5)
Basofily mikroskopic	<.>	0.0	%	(0 - 1)
Metamyelocyty mikros	<H>	1.0	%	(0 - 0)
Myelocyty mikroskopi	<.>	0.0	%	(0 - 0)
Promyelocyty mikrosk	<.>	0.0	%	(0 - 0)
BLASTY mikroskopicky	<H>	54.0	%	(0 - 0)
Prolymfocyty mikrosk	<.>	0.0	%	(0 - 0)
Plazmatické buňky	<.>	0.0	%	(0 - 0)
Nedif.buňky	<.>	0.0	%	(0 - 0)
Nedif.blasty	<.>	0.0	%	(0 - 0)
Normoblasty mikrosko	< >	5.0	/100 bb	
Hodnocení morfologie	< >			
Hodnocení morfologie	< >			hypersegmentace neutrofilů,
Morfologie ERY	< >			:
Morfologie PLT	< >			mírná anizo PLT,
KOMENTAR	< >			Změna oproti předešlému.
Neznamé vyšetření	< >			NRBC/100WBC :0.00







Laboratory diagnostics

Biochemical analysis of blood (elevated LD in myeloproliferative diseases)

Coagulation – DIC, thrombophilia, bleeding
fibrinogen, aPTT, PT, AT III, DD, EGT

Other
(Chest X ray, abdominal sonography,
ECG, heart sonography, serology – CMV…)
- we have to exclude focal infections and to evaluate function of heart, kidneys, liver and lungs (chemotherapy is nephrotoxic, hepatotoxic or cardiotoxic)

CLASSIFICATION OF LEUKEMIA

FAB (1982)

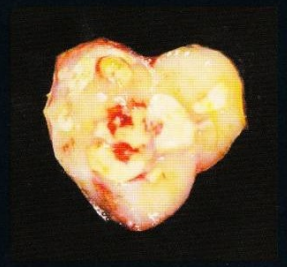
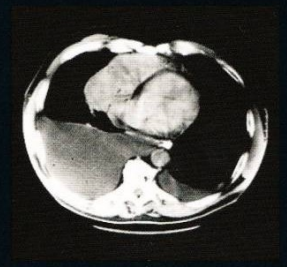
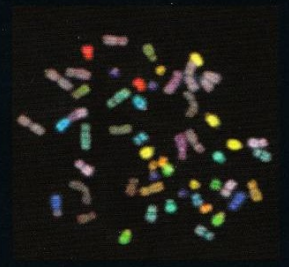
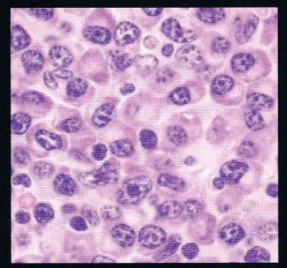
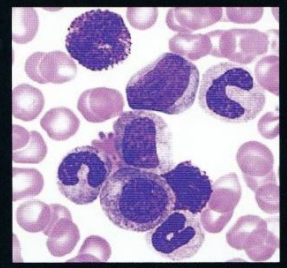
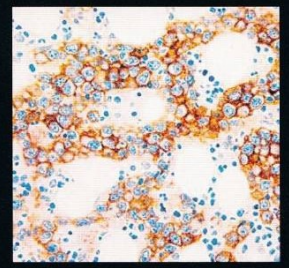
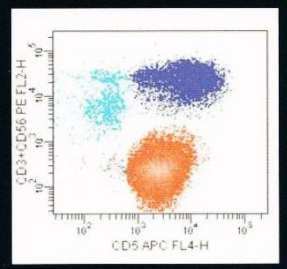
Classification according to morphology of malignant cells

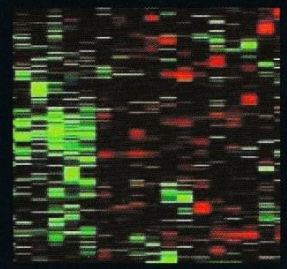
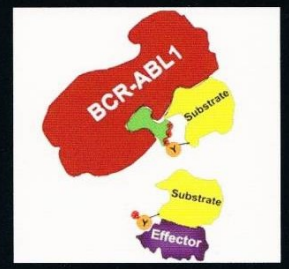
WHO (1999-)

Classification according to morphology, cytogenetic features, flow cytometry, and molecular genetic features

WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues

Edited by Steven H. Swerdlow, Elias Campo, Nancy Lee Harris, Elaine S. Jaffe,
Stefano A. Pileri, Harald Stein, Jürgen Thiele, James W. Vardiman

WHO

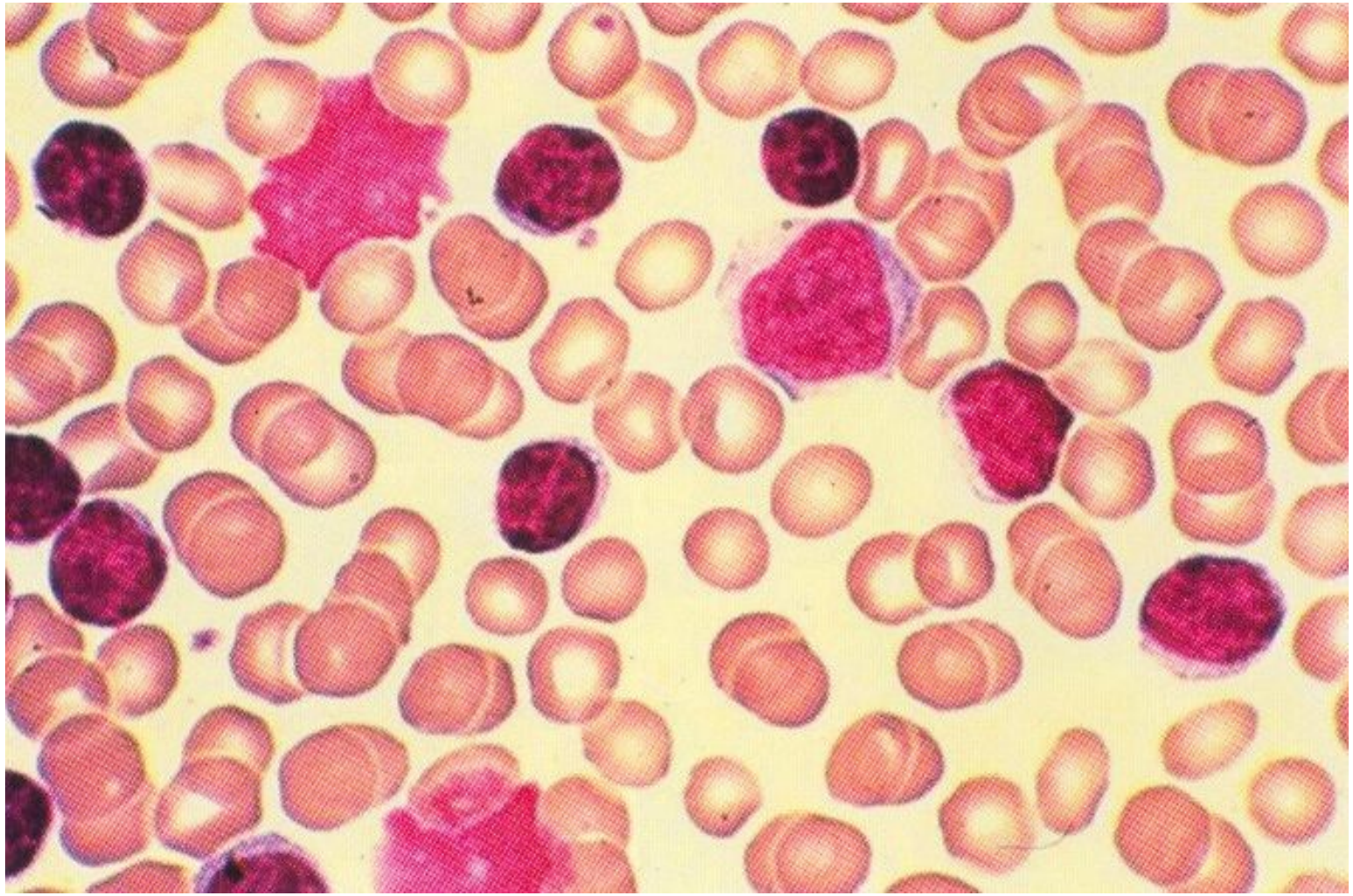
CHRONIC LYMPHOCYTIC LEUKEMIA (CLL)

The most common leukemia of Caucasians. CLL is a disorder characterized by the accumulation of small mature-appearing lymphocytes in the blood, marrow, and lymphoid tissues.

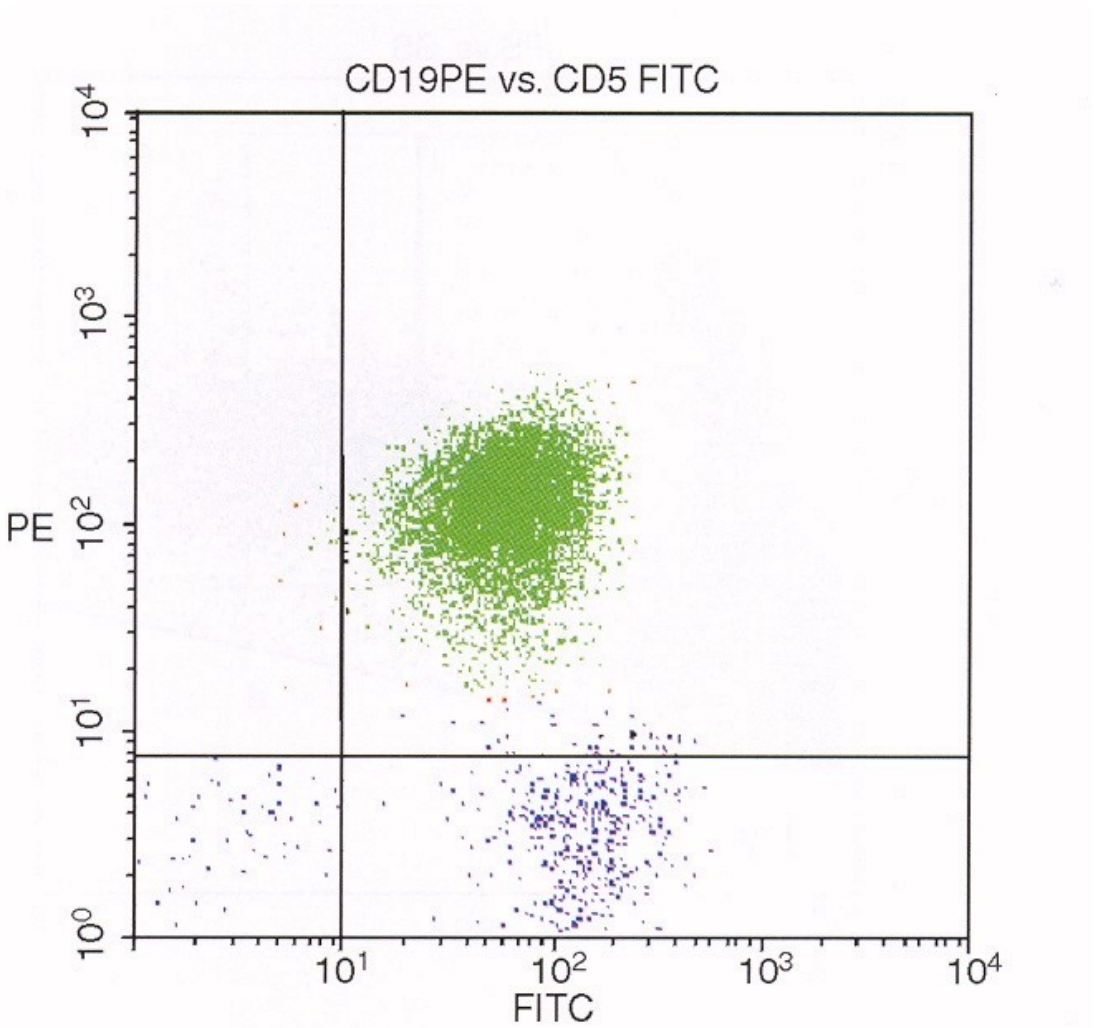
Laboratory and clinical features:
leukocytosis (absolute lymphocytosis), lymphadenopathy, splenomegaly, hepatomegaly, anemia, thrombocytopena, often autoimmune diseases (hemolysis).

Prognosis – different (better in CLL mutated genes for IgH or/and in CLL with del 13q14.

Median survival of CLL patients is 11+ years.



Diagnostics based on flow cytometry: CD5+19+20dim+23+FMC7-79b-200+sIg+/-

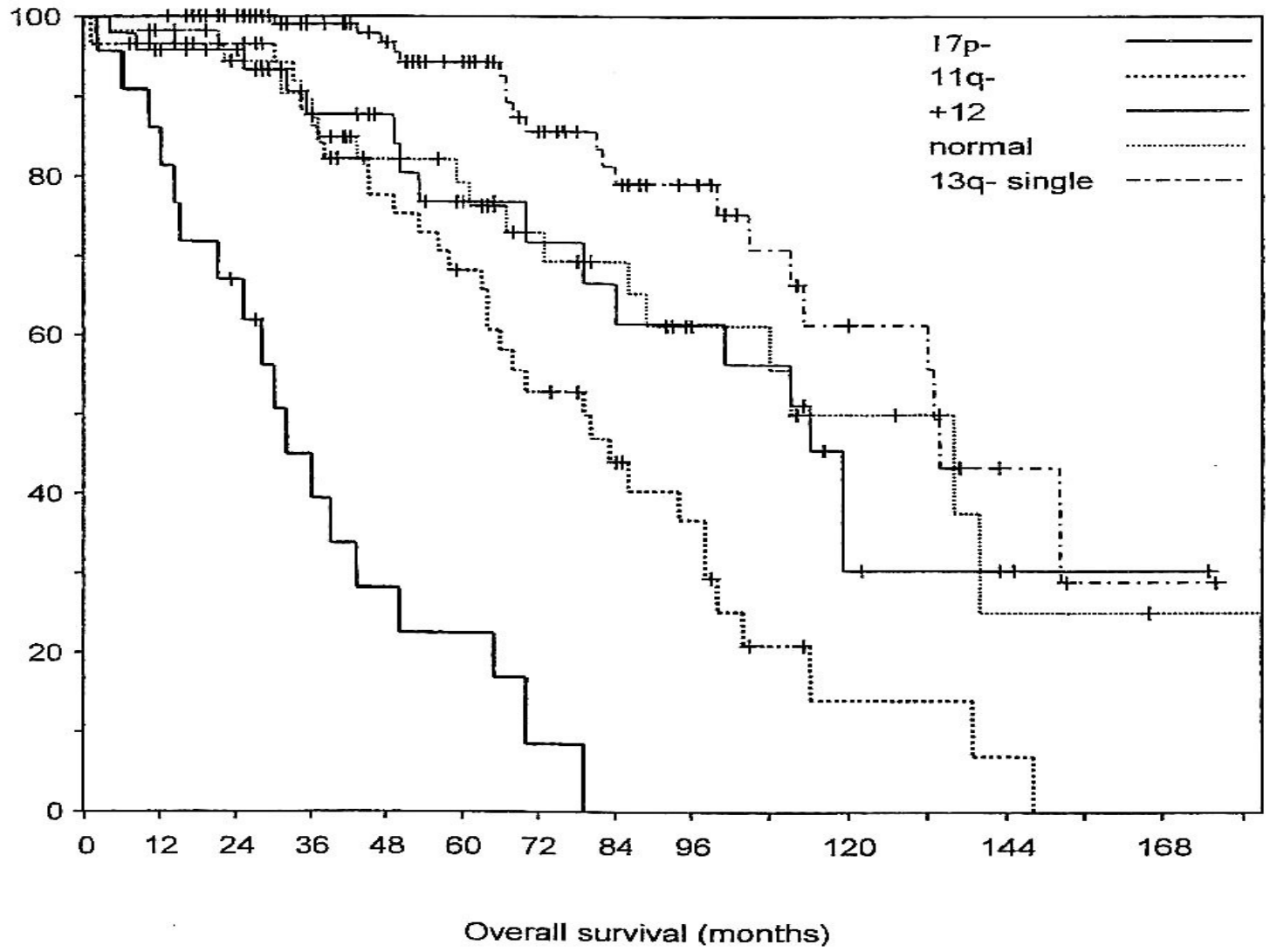


CLL staging

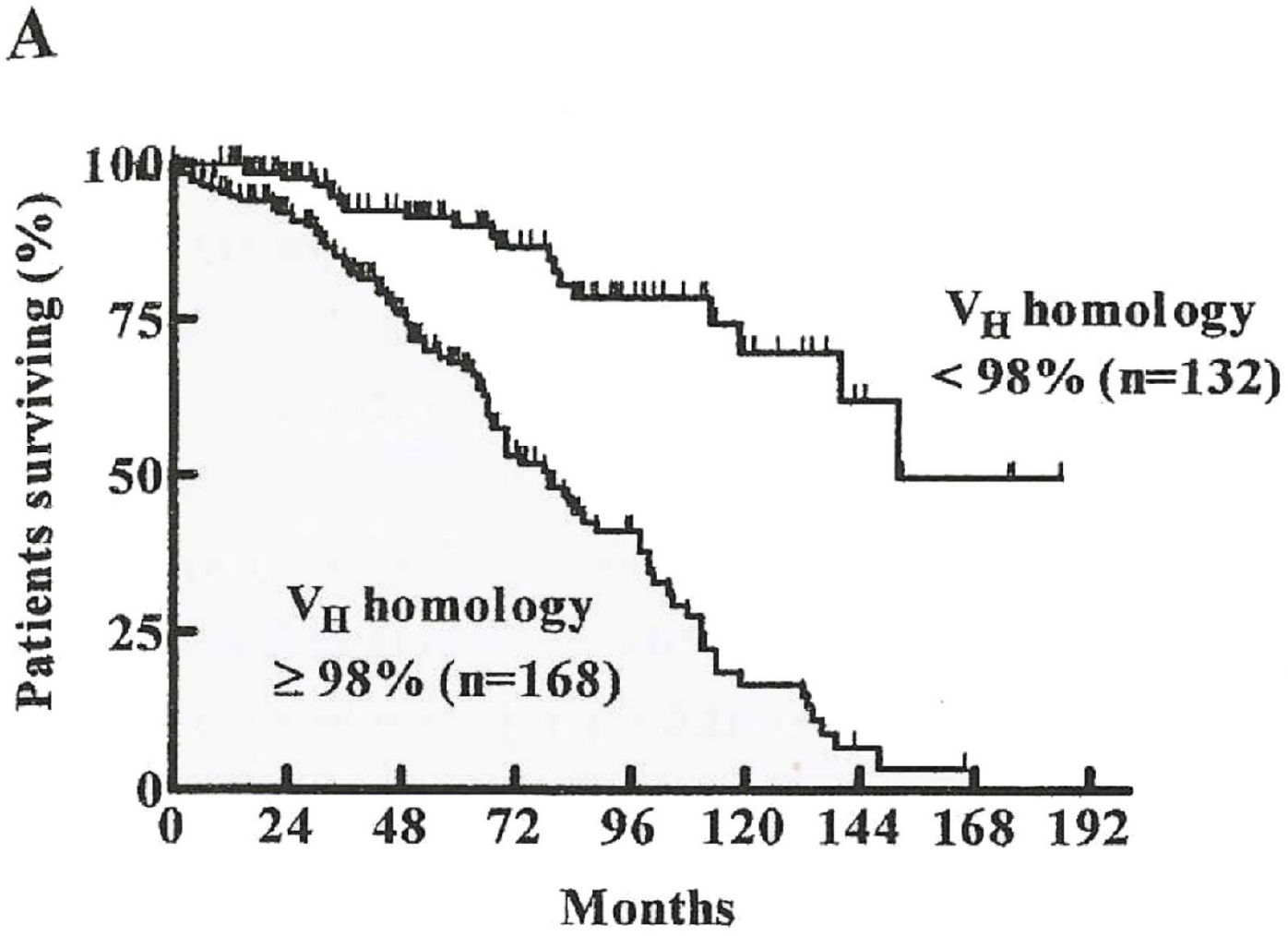
Treatment in stage Rai III or IV patients only (Binet C)

Clinical stage (Rai)	Risk	Median survival
0 (lymphocytosis)	Low	>150 months
I (lymphocytosis + lymphadenopathy)	Intermediate	101
II (lymphocytosis + splenomegaly)	Intermediate	71
III (lymphocytosis + anemia Hb < 110 g/l)	High	19
IV (lymphocytosis + thrombocytopenia < $100x10^9$/L)	High	19
Clinical stage (Binet)		
A (involvement <3 regions)	Low	Not reached
B (involvement ≥ 3 regions)	Intermediate	84
C (anemia and thrombocytopenia)	High	24

CLL prognosis based on cytogenetics



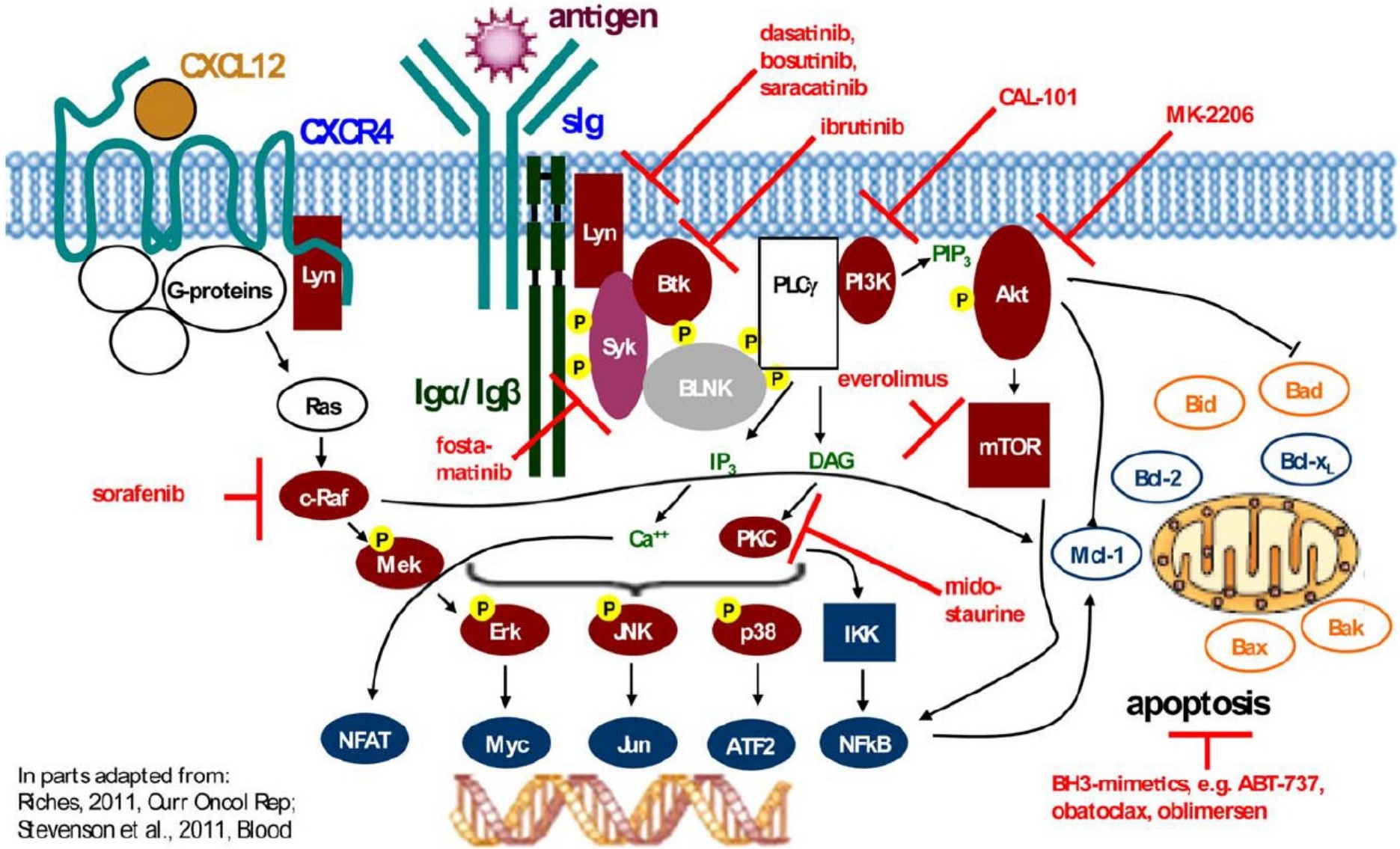
CLL prognosis based on IgHV mutational status



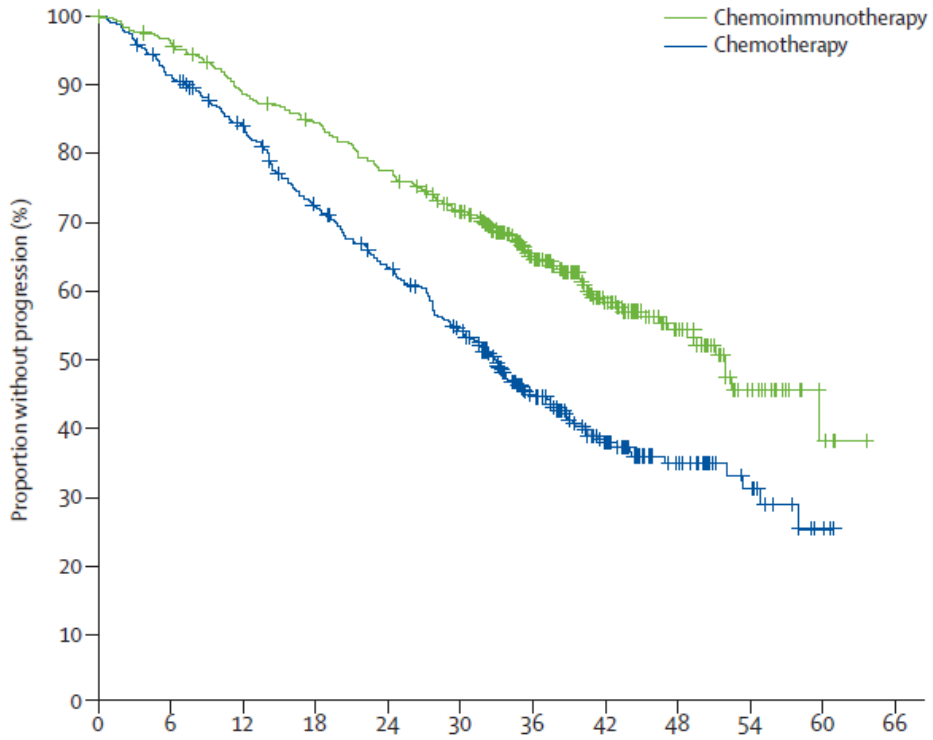
CLL THERAPY

Treatment for advanced stages only:

- fludarabine+cyclophosphamide+rituximab
- bendamustine+rituximab
- chlorambucil + anti CD20 antibody (rituximab, obinutuzumab)
- ibrutinib, idelalisib (BCR inhibitors)
- venetoclax (Bcl2 inhibitor)
- (allogeneic transplant)

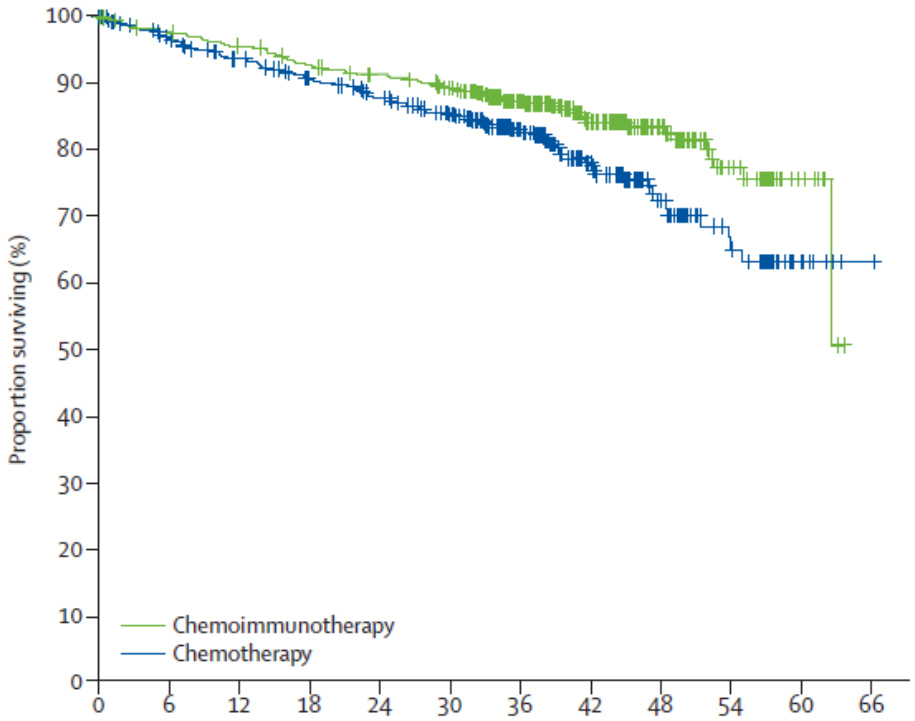

antigen
CXCL12
CXCR4
sIg
dasatinib,
bosutinib,
saracatinib
ibrutinib
CAL-101
MK-2206
Lyn
G-proteins
Lyn
Btk
PLCγ
PI3K
PIP3
Akt
Syk
BLNK
Igα/ Igβ
fosta-
matinib
everolimus
mTOR
Ras
sorafenib
c-Raf
IP3
DAG
Ca++
PKC
mido-
staurine
Mek
Erk
JNK
p38
IKK
NFAT
Myc
Jun
ATF2
NFkB
Bid
Bad
Bcl-2
Bcl-xL
Mcl-1
Bax
Bak
apoptosis
BH3-mimetics, e.g. ABT-737,
obatoclax, oblimersen
In parts adapted from:
Riches, 2011, Curr Oncol Rep;
Stevenson et al., 2011, Blood

CLL – FCR regimen treatment outcome



PFS at 3 years
45% vs. 65%
p < 0,0001

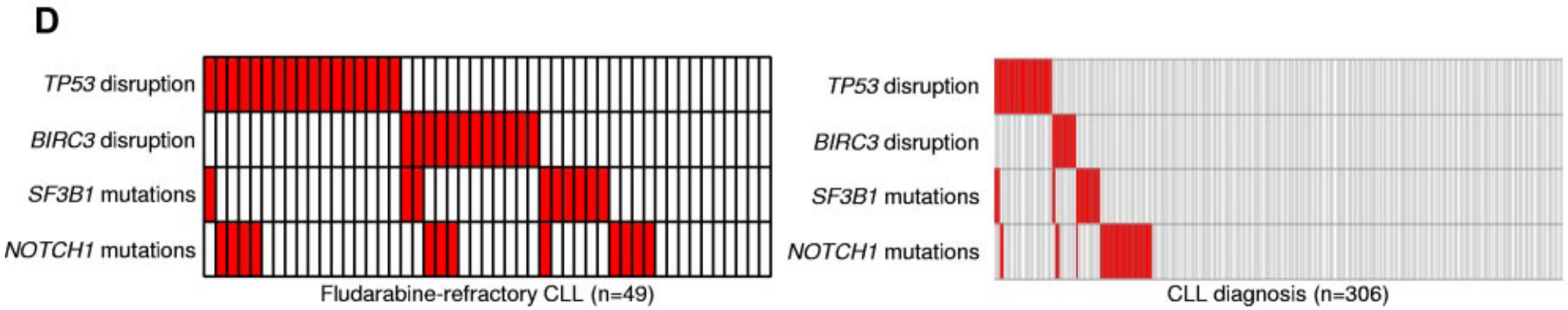
OS at 3 years
83% vs. 87%
p = 0,012



Clonal evolution in CLL – *TP53*

Before treatment

Unfavorable *SFB3, NOTCH1, BIRC3* mutations

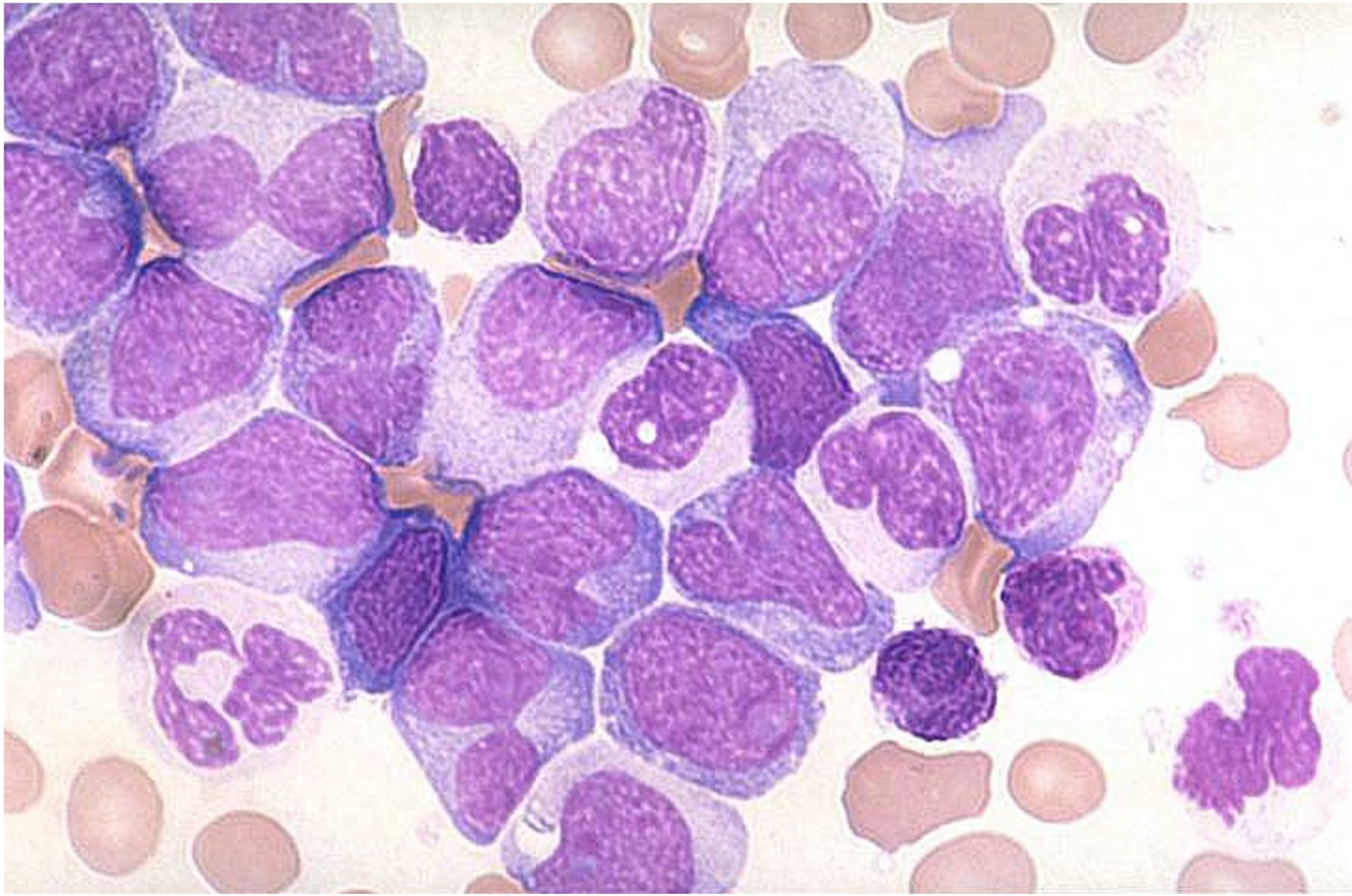


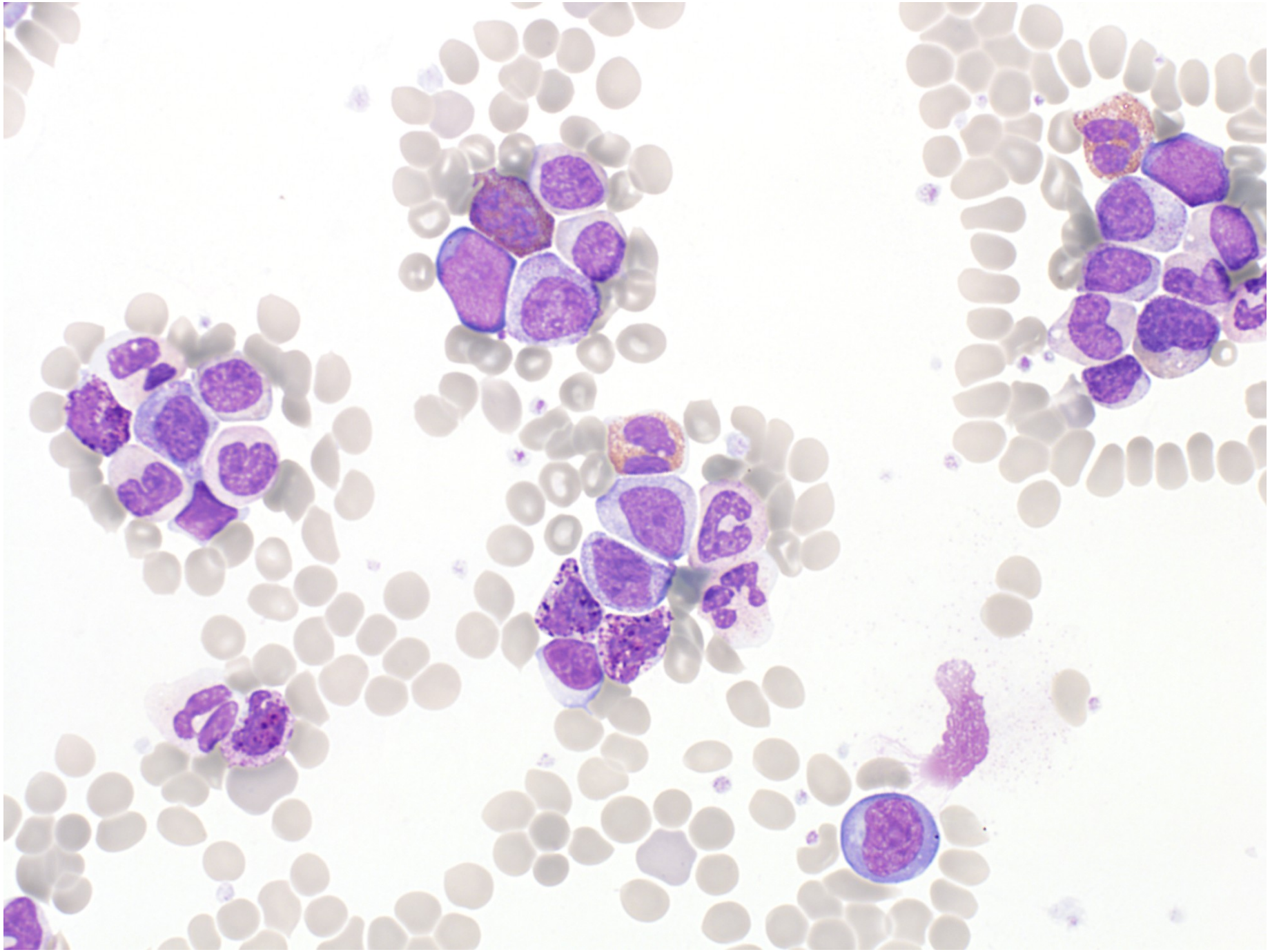
CHRONIC MYELOID LEUKEMIA (CML)

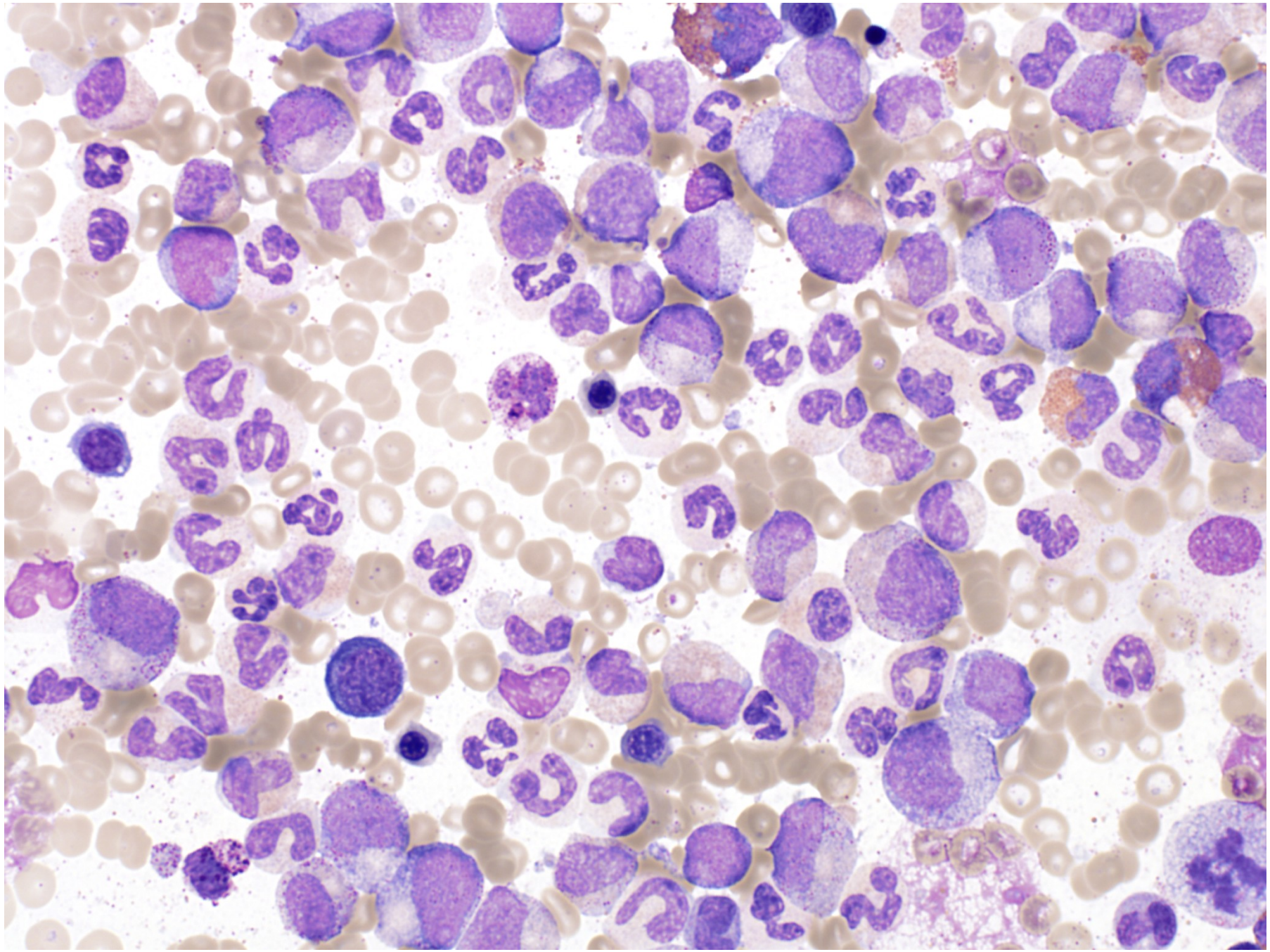
CML is a pluripotent stem cell disease that is characterized by extreme blood granulocytosis, basophilia, often thrombocytosis, anemia, and splenomegaly.

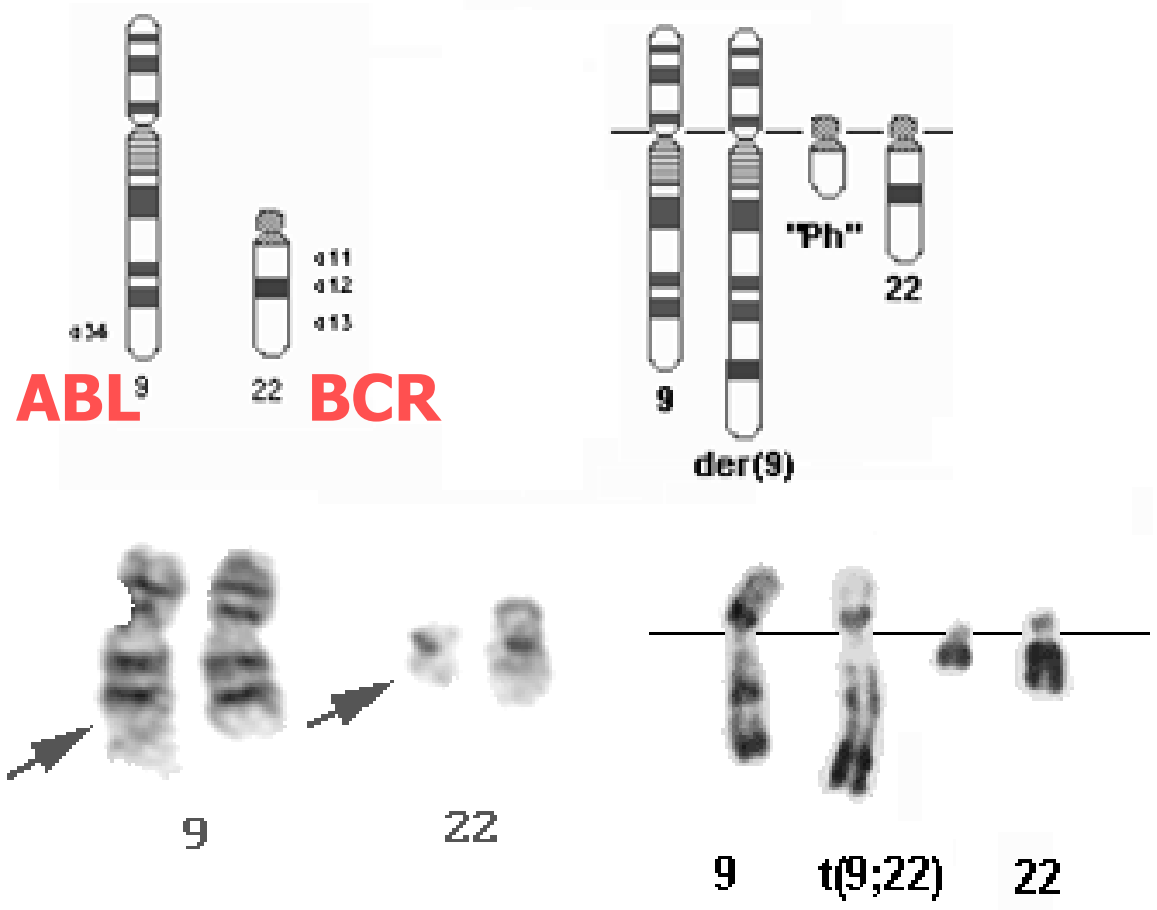
Stages of untreated CML:
chronic phase, accelerated phase (rapid increase of WBC, worsening of thrombocytopenia, new cytogenetic features, resistence to treatment), blast crisis (resembles to acute leukemia)

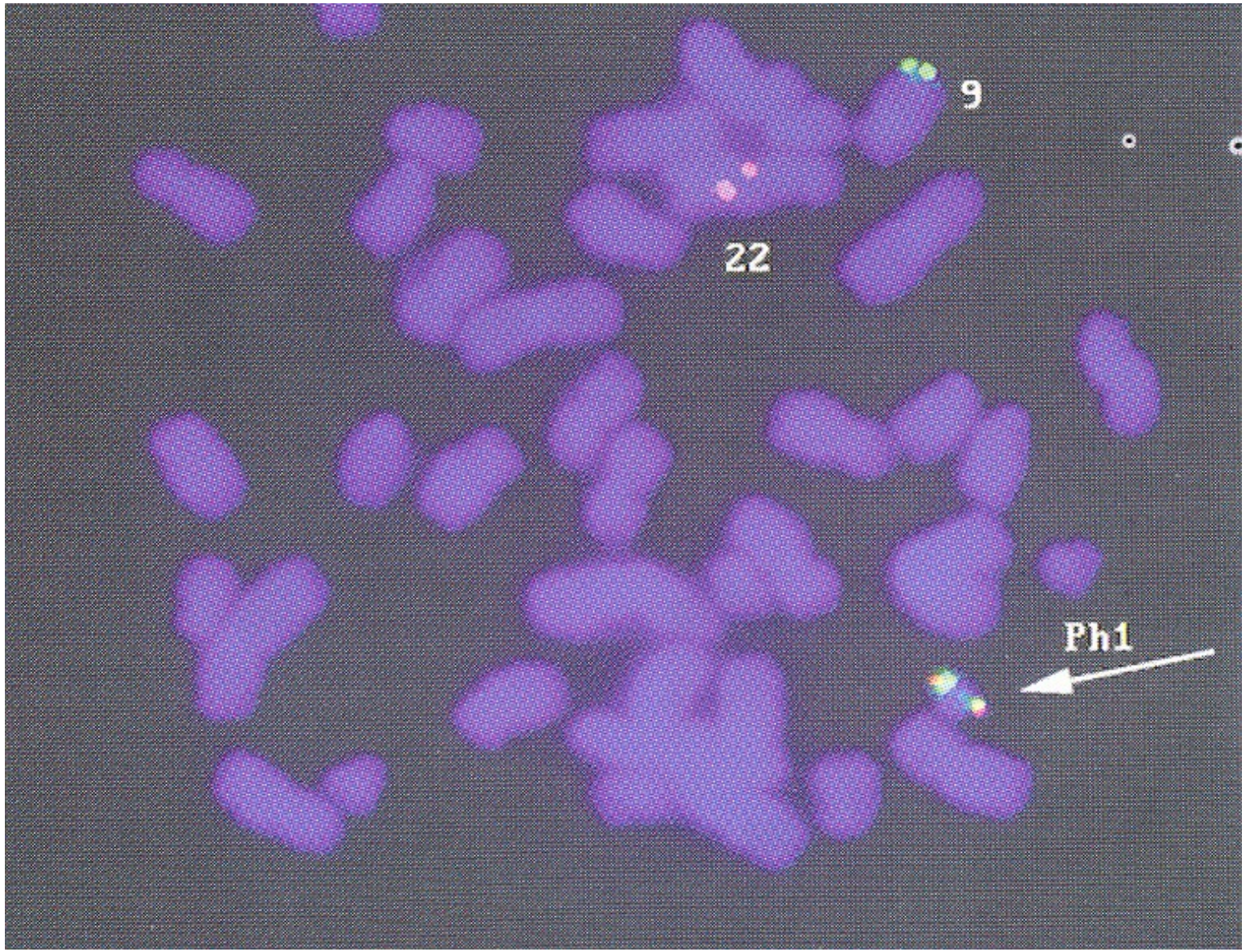
Etiologic role of chromosome discovered in Philadelphia - Ph chromosome








q34
q11
q12
q13
ABL
9
22
BCR
"Ph"
22
9
der(9)
9
22
9
t(9;22)
22


9
22
Ph1

- Ph chromosome arises from t(9;22)

- chimeric gene *BCR-ABL* arises from Ph chromosome

- *BCR-ABL* gene produces BCR-ABL tyrosinkinase

- *BCR-ABL* tyrosinkinase induces defect of apoptosis

There is almost no *BCR-ABL* negative CML!

Minimal residual disease during treatment

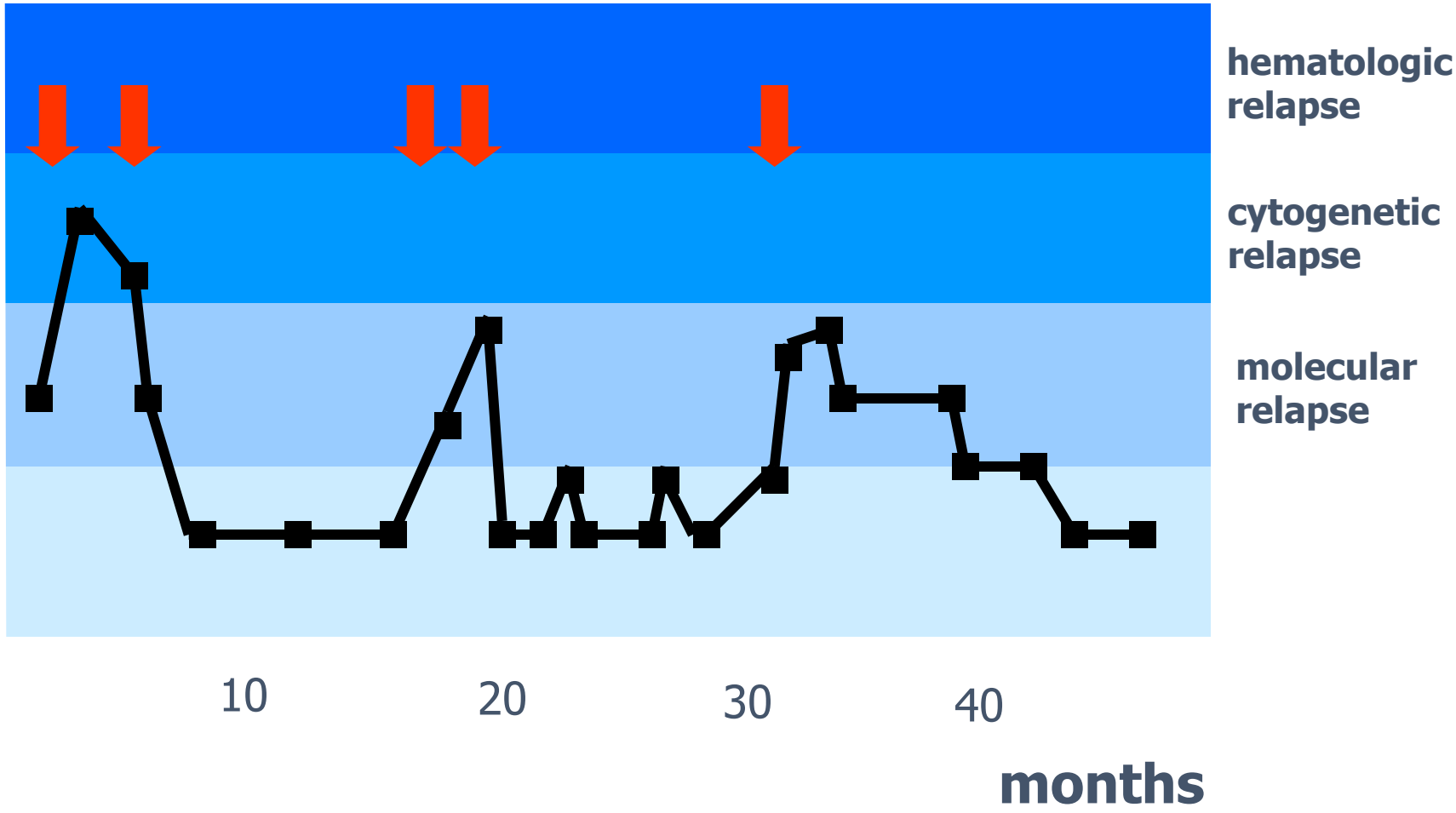
- **Hematologic monitoring**

- **Cytogenetic monitoring**

- **Molecular genetic monitoring (RQ-RT-PCR, digital PCR, NGS)**

Minimal residual disease during therapy

Molecular relapse is better manageable compared with cytogenetic or hematologic relapse



THERAPY

All patients treated!

- **imatinib**
- **nilotinib, dasatinib, bosutinib**
- **ponatinib**
- **interferon**
- **allogeneic transplantation**

II. Zwei Fälle von Leucaemie.

Mitgetheilt

von

Dr. **Lissauer** in Bendorf.

Der in Nr. 31. dieser Wochenschrift von Dr. Valentiner mitgetheilte Fall von Leucaemie, bei welcher zur Coupirung des Fiebers Liq. arsenic. Fowler. angewandt wurde, brachte mir zwei Fälle derselben Krankheit in Erinnerung, die ich kurz nach einander im Landkrankenhause in Cassel zu beobachten Gelegenheit hatte, von welchen bei einem Liq. arsen. Fowler. eine Zeit lang versuchsweise von gutem Erfolge war. Ich theile beide Fälle hier kurz mit, theils als einen kleinen Beitrag zur Kenntniss dieser im Ganzen immer noch selten diagnosticirten Krankheit, theils, um zur weiteren Anwendung obigen Mittels anzuregen.

N. N., 32 Jahre alt, weiblichen Geschlechts, wurde im October v. J. aufgenommen. Sie gab an, früher stets gesund, mit 17 Jahren regelmässig menstruirt gewesen zu sein, und vor ungefähr einem Jahre ein uneheliches Kind geboren zu haben, das bald nach der Geburt gestorben sei. Von ihrem Liebhaber, der ihr die Ehe versprochen, hintergangen, habe sie sich sehr gegrämt und viel Sorgen gemacht. Zugleich will sie seit dieser

---

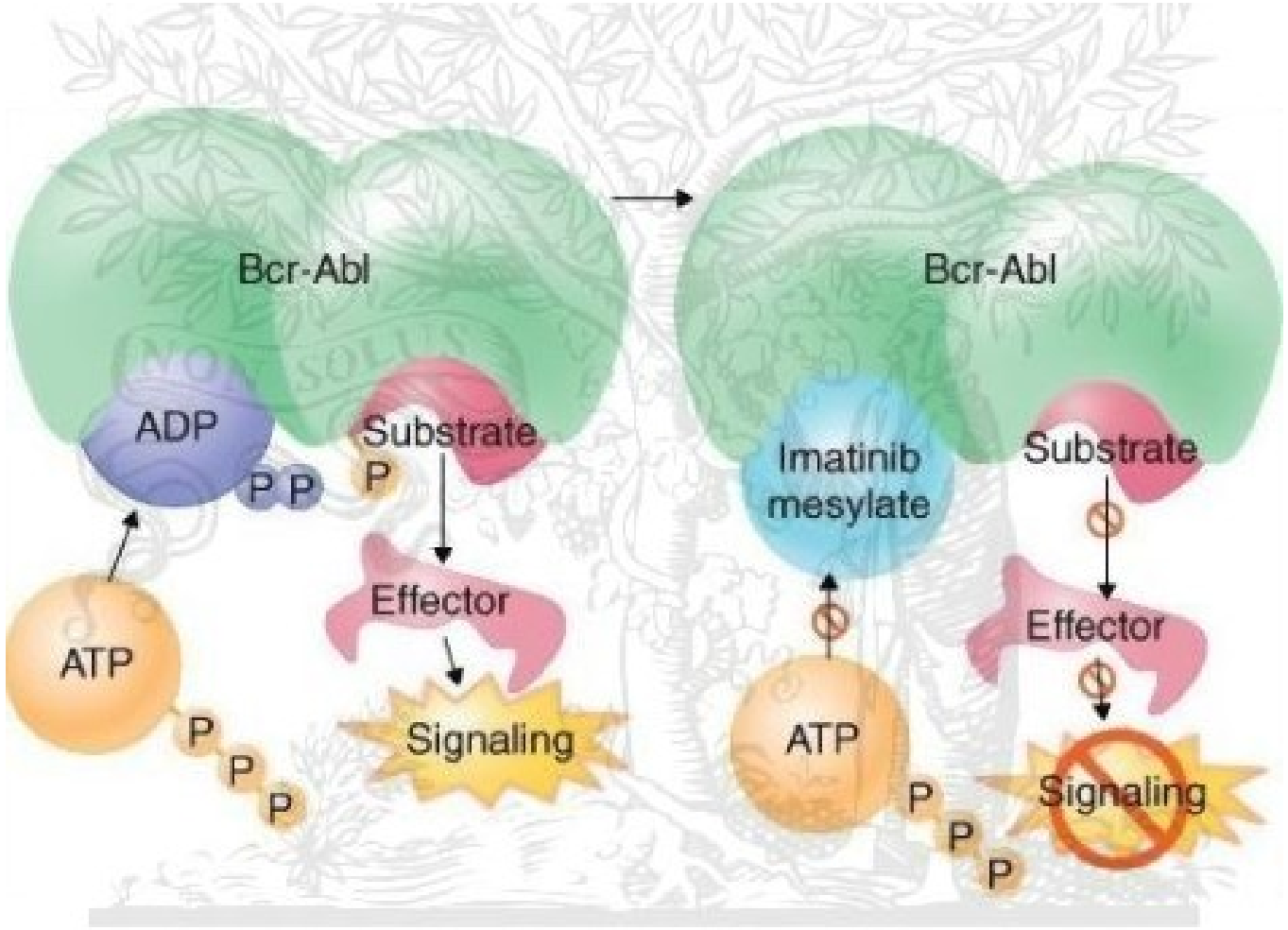
*) Malgaigne l. c. p. 1004. Revue medic. chirurg., 1849, T. V., p. 246.

Asenic trioxide

Lissauer: Zwei Fälle von Leucaemie.

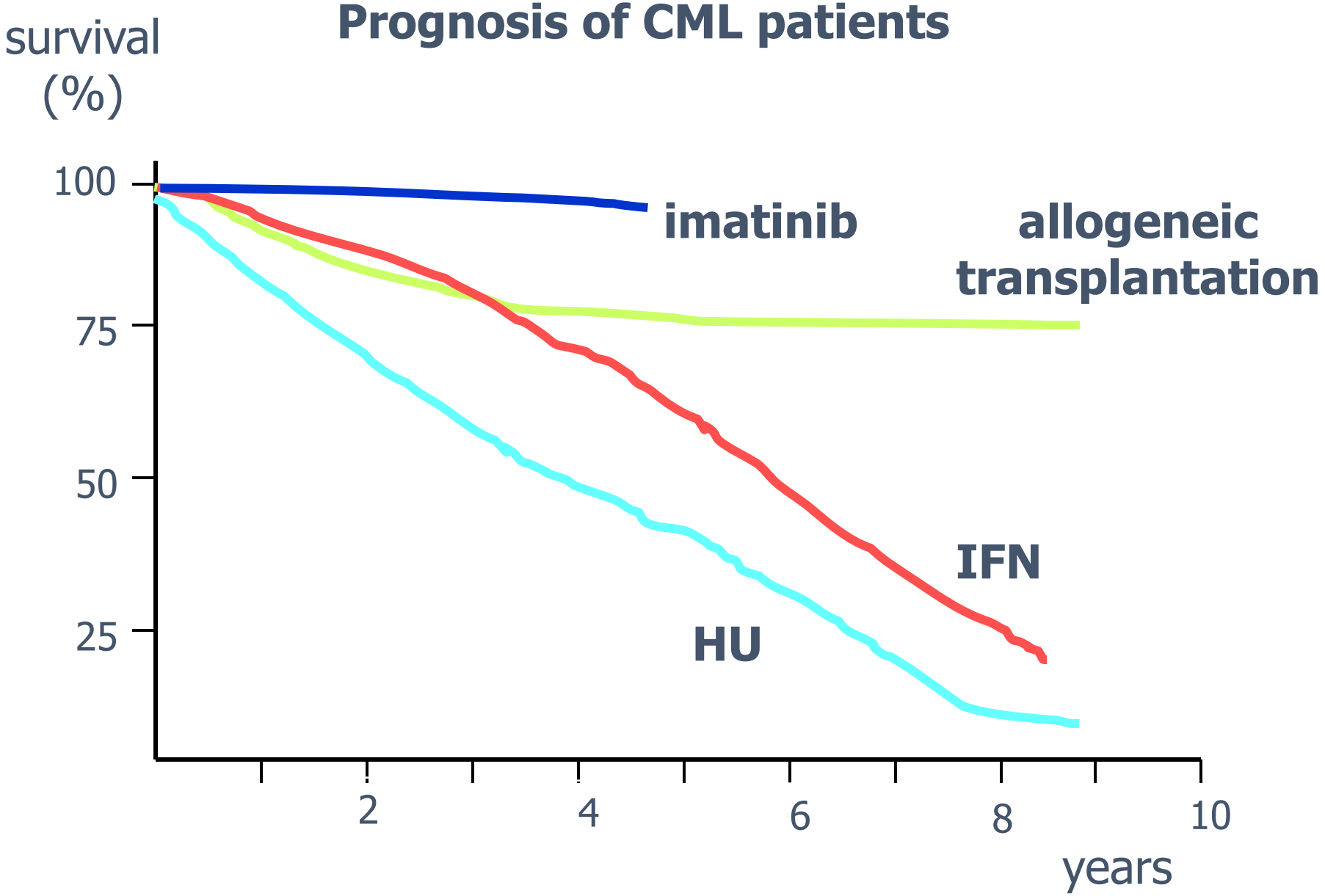
Berlin. Klin. Wochenschrift, 2, 1865, s. 403 - 404

Imatinib mode of action



CP-CML	Léčebná strategie
1. linie:	▪Imatinib 400 mg
2. linie:	
IM-intolerance	▪DASATINIB nebo NILOTINIB
IM-selhání	▪DASATINIB nebo NILOTINIB ▪aloTKB (progrese do AP/BC, T315I)
IM-suboptimální odpověď	▪IM stejná dávka ▪IM navýšení dávky ▪DASATINIB nebo NILOTINIB

Prognosis of CML patients



ACUTE MYELOID LEUKEMIA (AML)

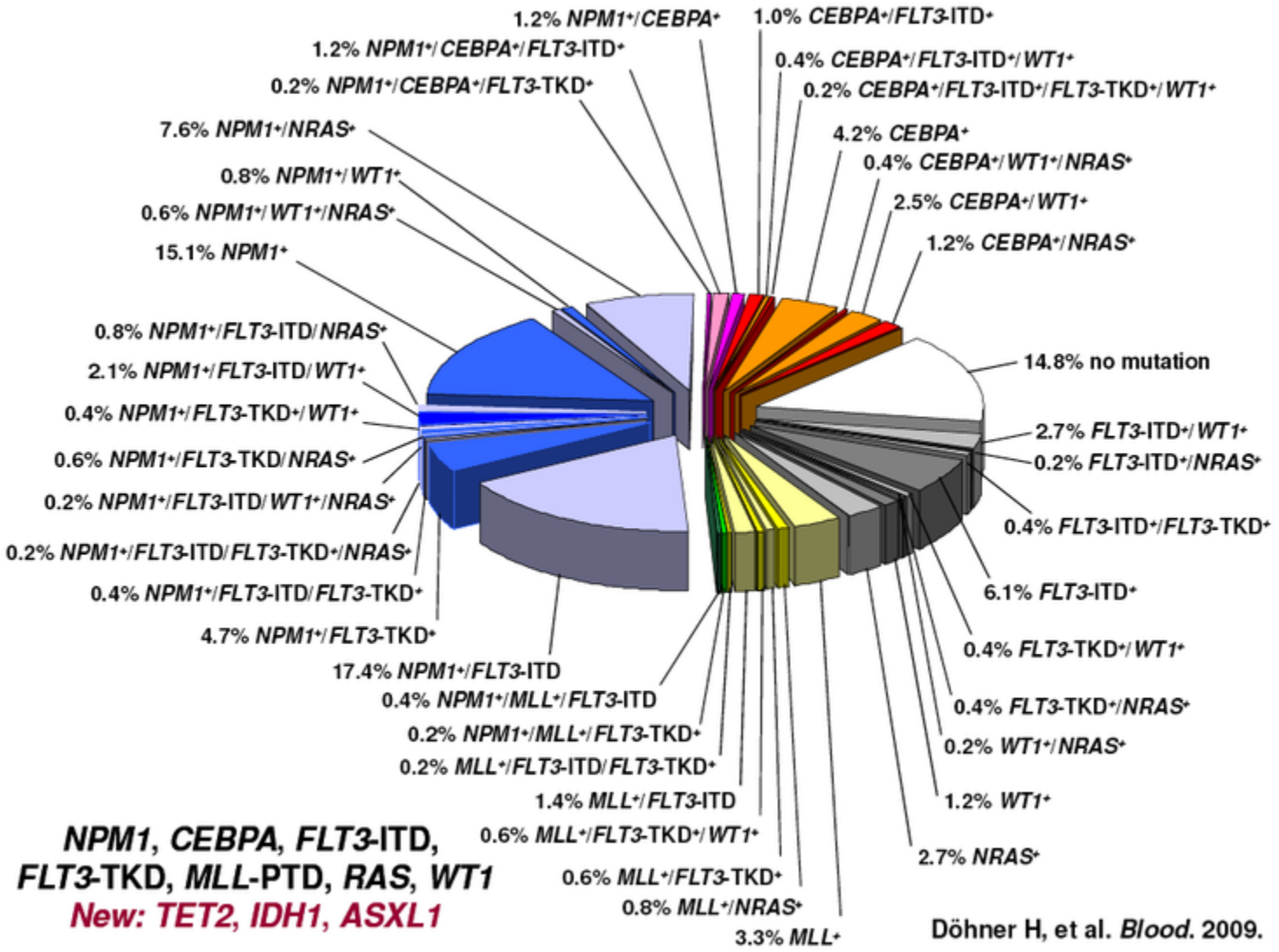
AML is clonal malignant disease that is characterized by the proliferation of abnormal (leukemic) blasts, principially in the marrow, and impaired production of normal blood cells.

Signs and symptoms of AML include pallor, fatigue, weakness, palpitations, bleeding, fever, and dyspnena.

In bone marrow, there is more than 20% of blast cells. (less than 20% - myelodysplastic syndrome)

Median survival of untreated patients is 6 weeks.

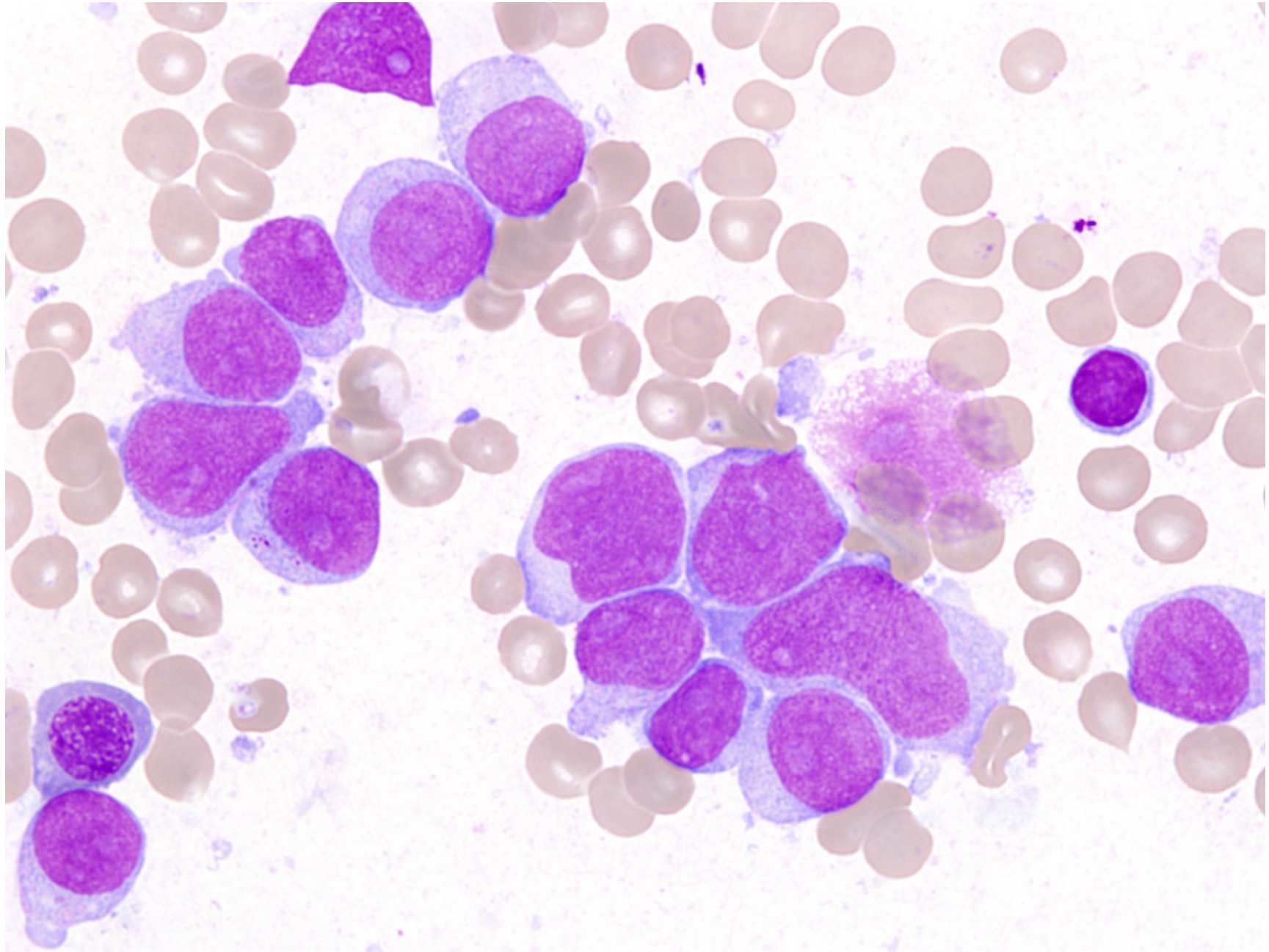
AML – heterogenous disease

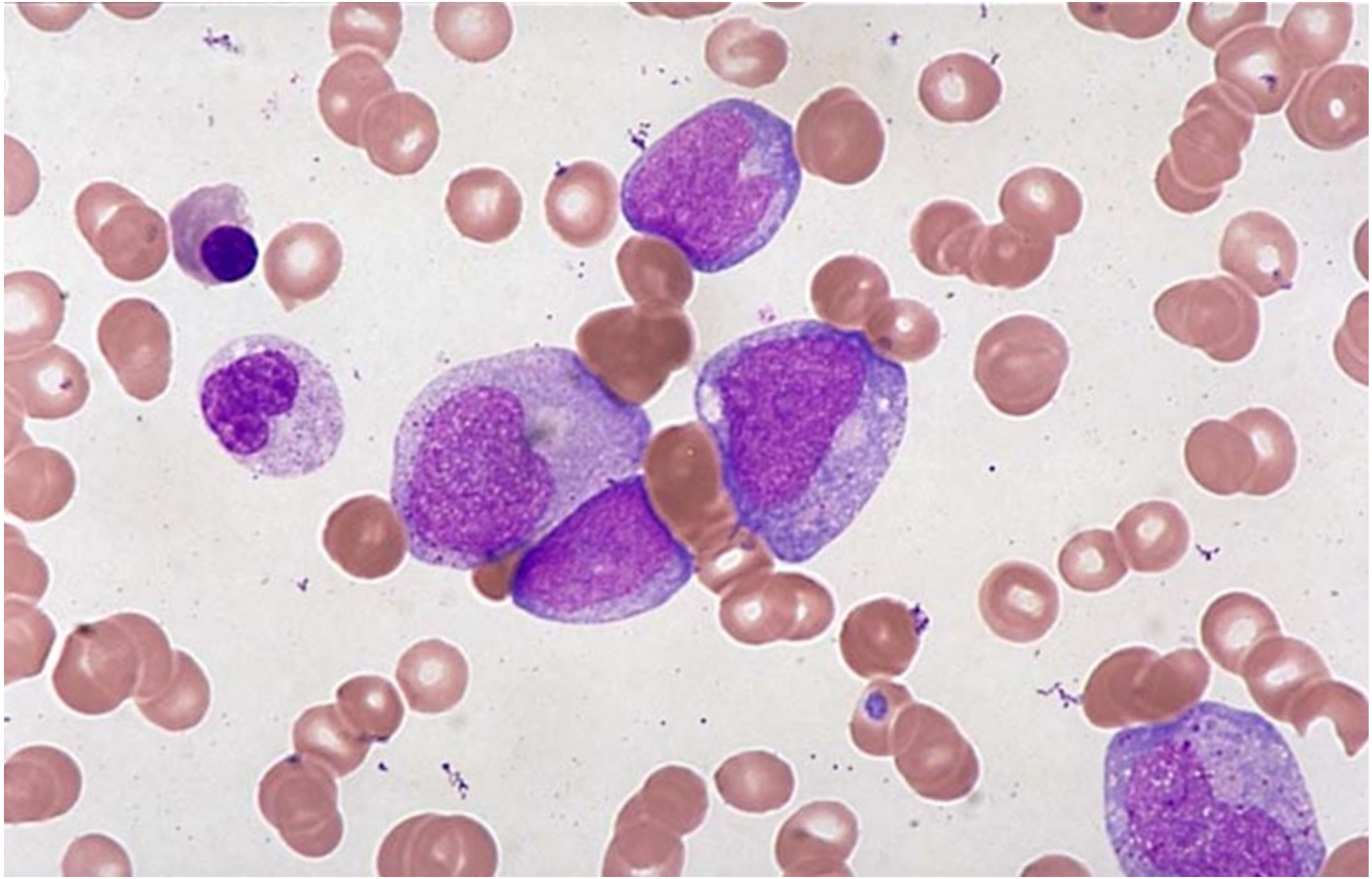


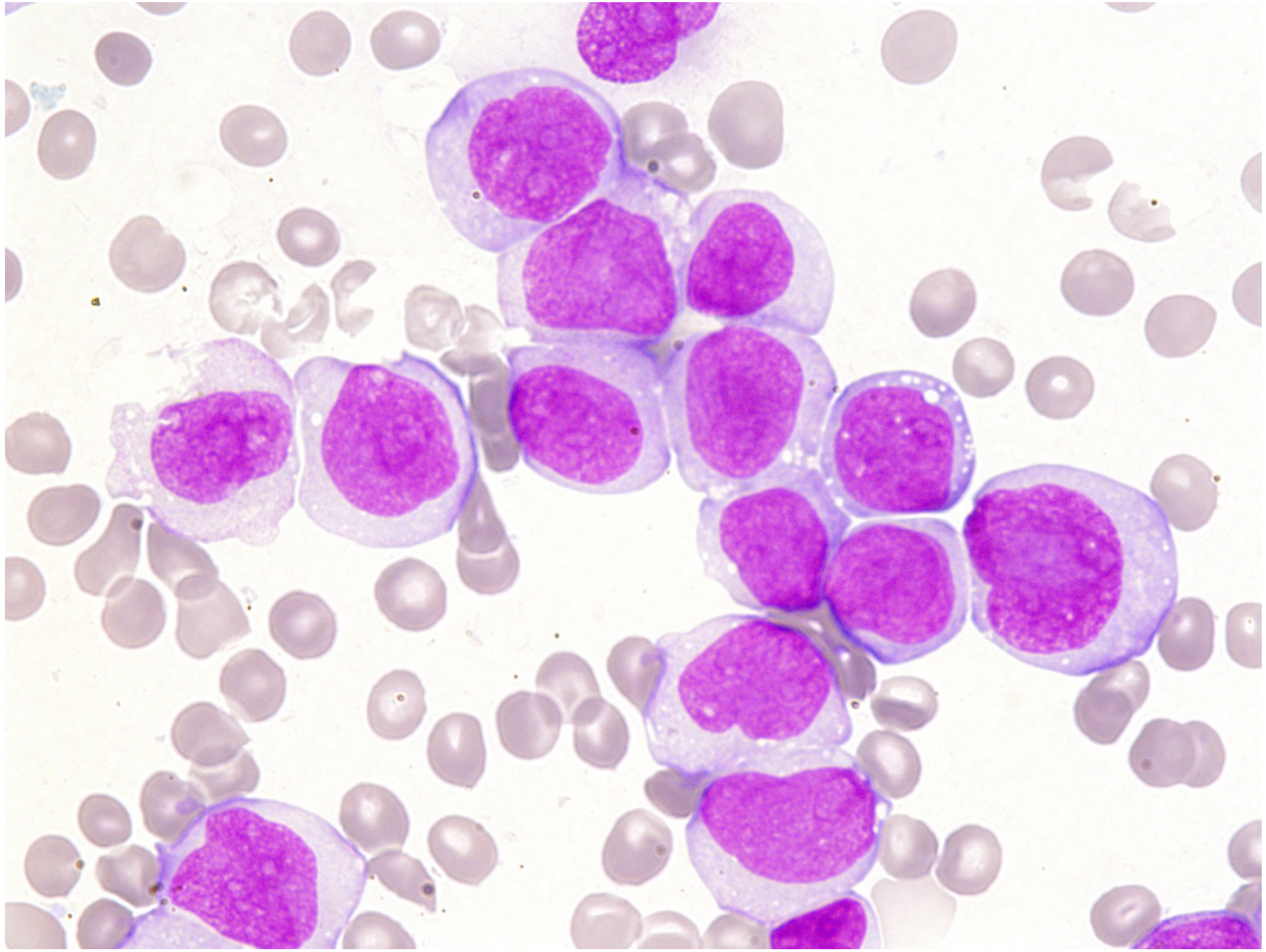
Döhner H, et al. *Blood*. 2009.

Acute myeloid leukemia (AML) and related neoplasms

- AML with recurrent genetic abnormalities
  - AML with t(8;21)(q22;q22.1);*RUNX1-RUNX1T1*
  - AML with inv(16)(p13.1q22) or t(16;16)(p13.1;q22);*CBFB-MYH11*
  - APL with *PML-RARA*
  - AML with t(9;11)(p21.3;q23.3);*MLLT3-KMT2A*
  - AML with t(6;9)(p23;q34.1);*DEK-NUP214*
  - AML with inv(3)(q21.3q26.2) or t(3;3)(q21.3;q26.2); *GATA2, MECOM*
  - AML (megakaryoblastic) with t(1;22)(p13.3;q13.3);*RBM15-MKL1*
  - *Provisional entity: AML with BCR-ABL1*
  - AML with mutated *NPM1*
  - AML with biallelic mutations of *CEBPA*
  - *Provisional entity: AML with mutated RUNX1*
- AML with myelodysplasia-related changes
- Therapy-related myeloid neoplasms
- AML, NOS
  - AML with minimal differentiation
  - AML without maturation
  - AML with maturation
  - Acute myelomonocytic leukemia
  - Acute monoblastic/monocytic leukemia
  - Pure erythroid leukemia
  - Acute megakaryoblastic leukemia
  - Acute basophilic leukemia
  - Acute panmyelosis with myelofibrosis
- Myeloid sarcoma
- Myeloid proliferations related to Down syndrome
  - Transient abnormal myelopoiesis (TAM)
  - Myeloid leukemia associated with Down syndrome

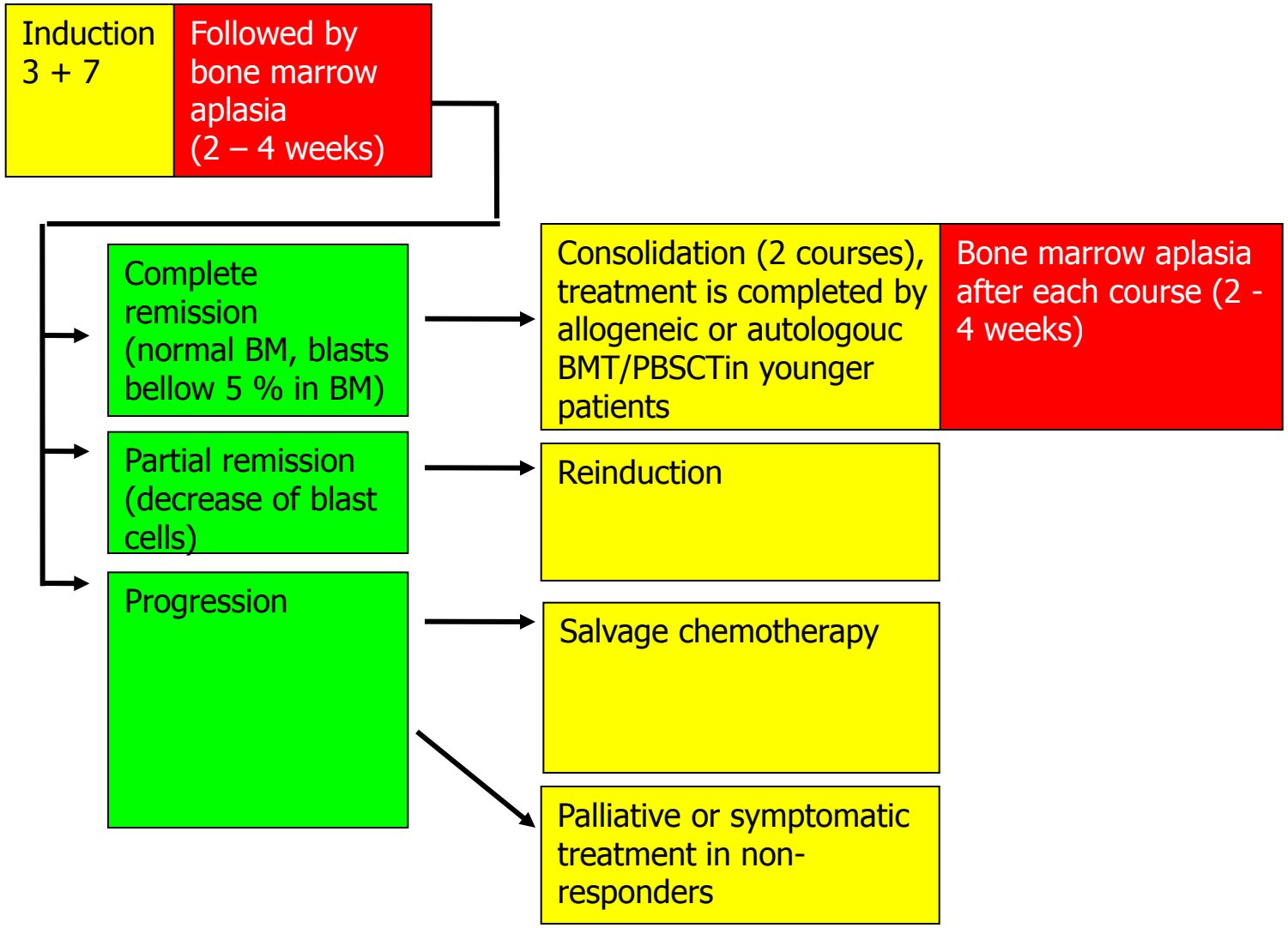






Treatment of AML

Treatment of choice of AML are courses of chemotherapy, the most potent drugs are cytosinarabinoside and anthracyclines.



Treatment of AML

Novel drugs for AML:

Midostaurin

Venetoclax

Gilteritinib

Acute promyelocytic leukemia (APL, AML M3)

APL is variant of AML (constitutes about 5-10% of AML in central Europe, about 25% of AML southern Europe, and 50 % of AML in eastern Asia).

There are prominent hemorrhagic complications (DIC, melena, hematuria, pulmonary bleeding, CNS bleeding)

Prognosis of APL was very poor 30 years ago (almost all patients died).

Nowadays, DFS is 80%.

Acute promyelocytic leukemia (APL, AML M3)

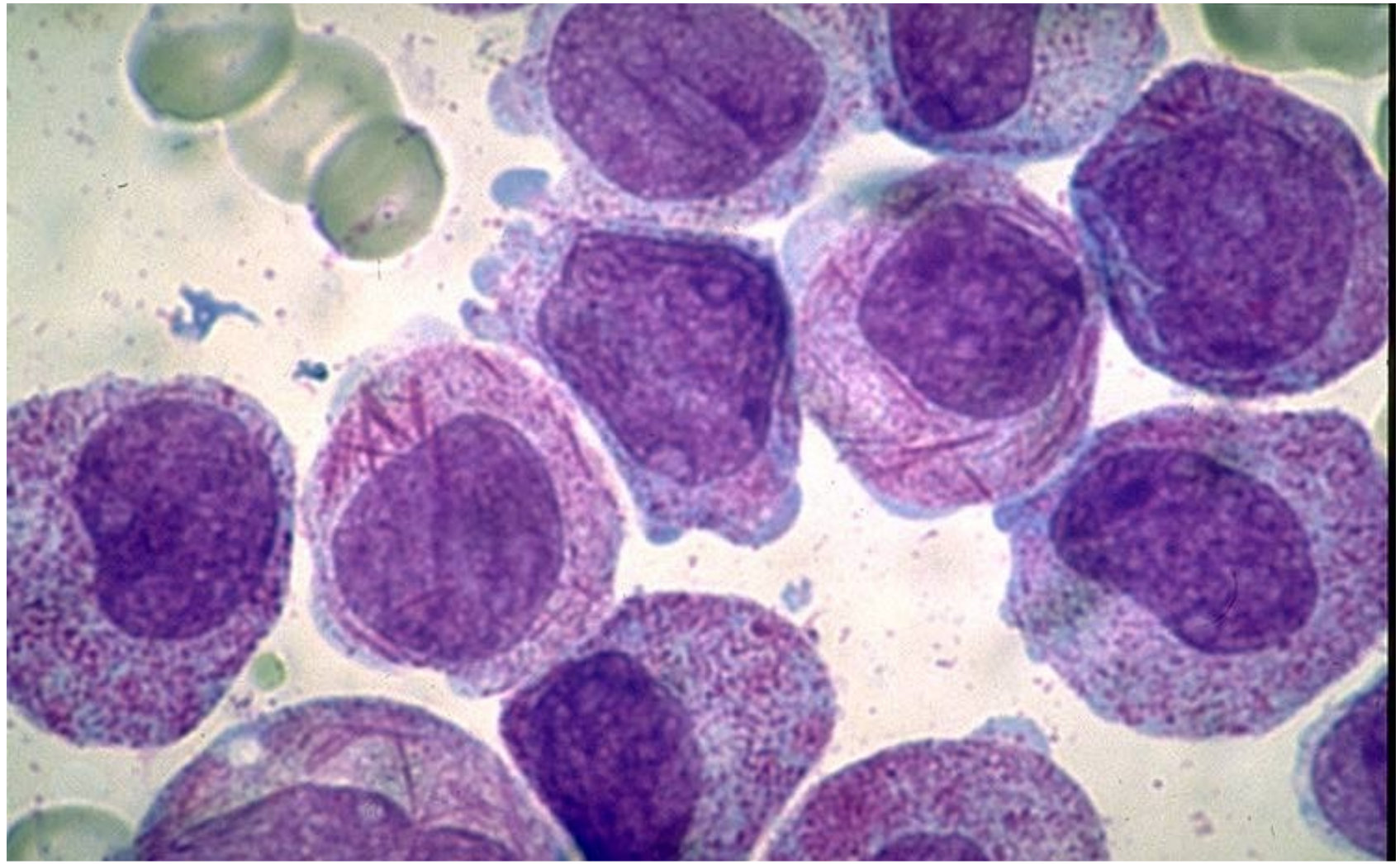
Promyelocytes are granular cells. In granula are coagulopathy-inducing factors (tissue factor…).

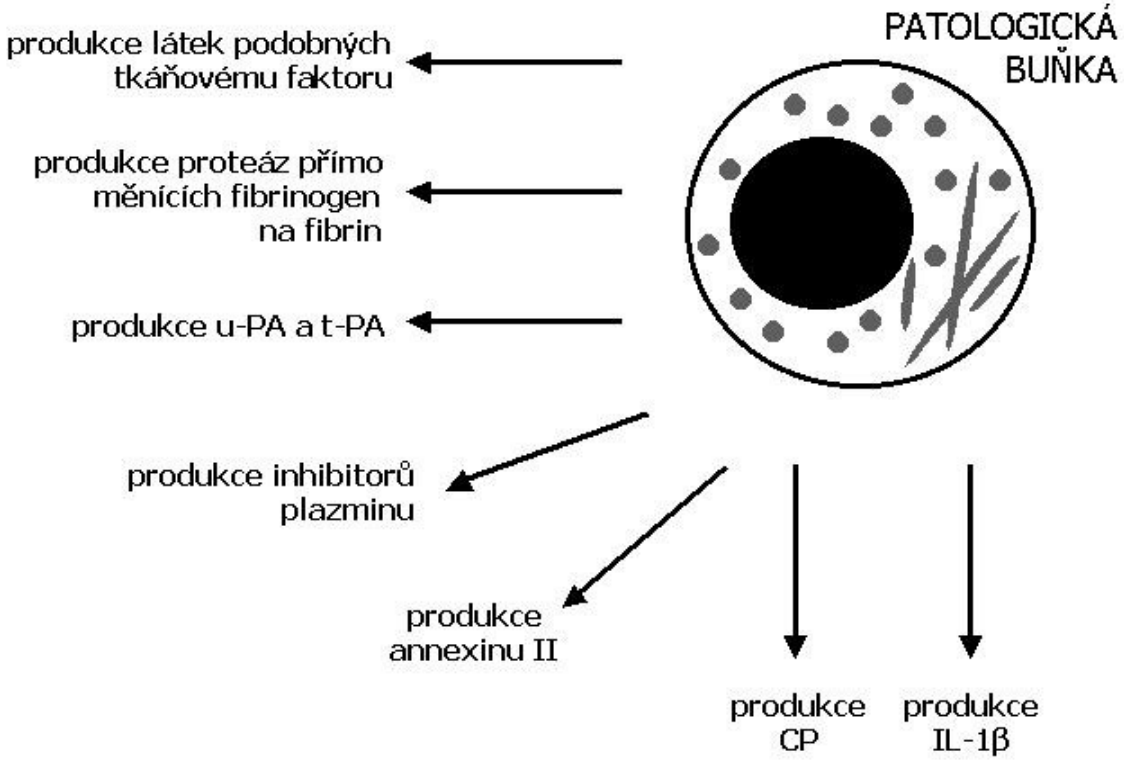
Majority of APL is characterized by t(15;17).
A translocation between chromosome 17 and 15 results in chimeric fusion gene *PML-RARα*.

PML-RARα gene produces PML-RARα abnormal receproor for retinoids. (Retinoids are necessary for normal bone marrow cells differentiation). In cells with t(15;17) normal differentiation is stopped.

We can restore differentiation by means of ATRA + ATO or chemo.

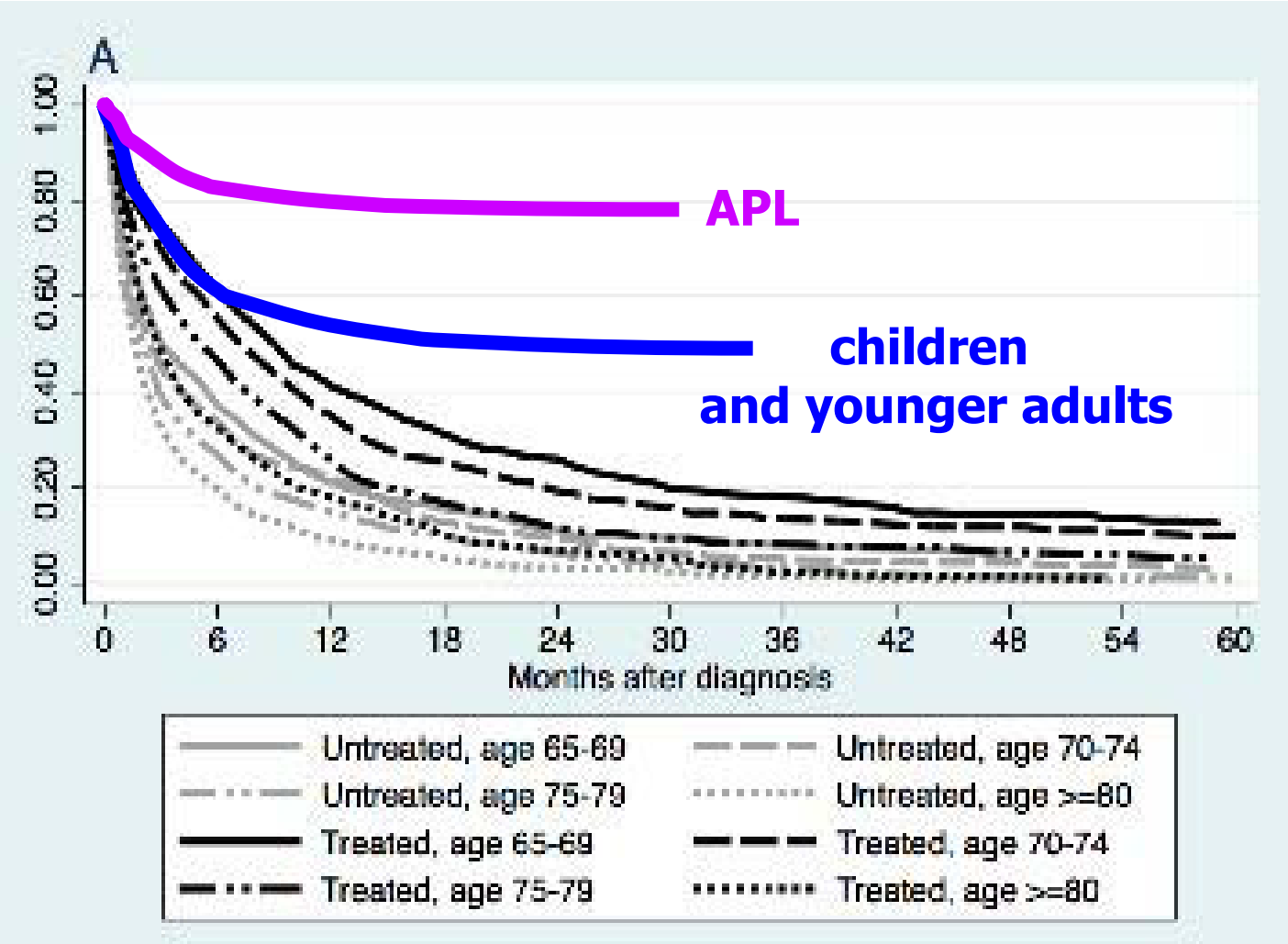
Chemotherapy or arsenic trioxide + ATRA is treatment of choice for APL!



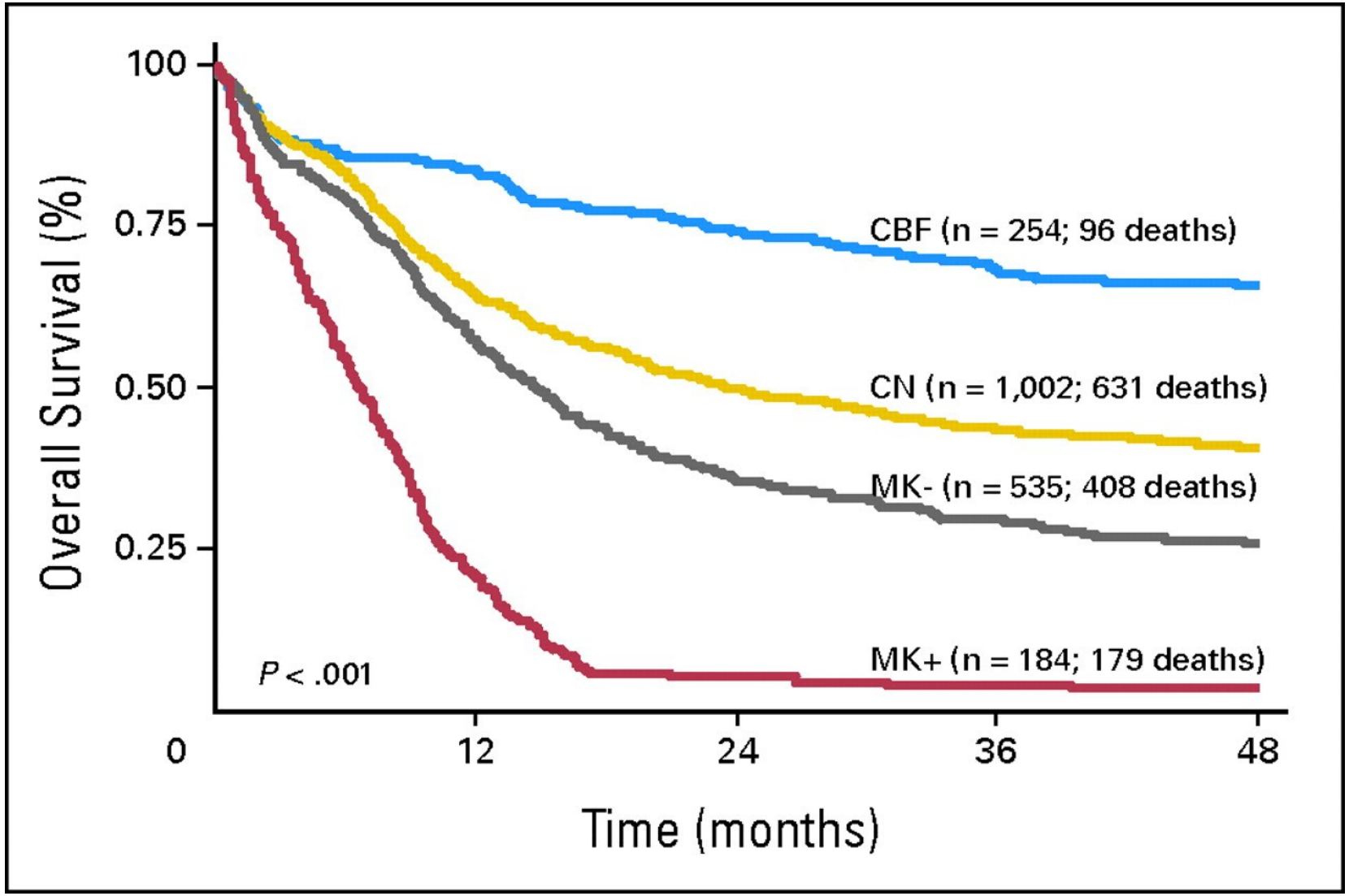


Bleeding diathesis in APL:
CP – *cancer procoagulant*, IL-1β – interleukin 1β, t-PA – tissue plasminogen activator, u-PA – urokinase.

Survival of AML patients



Survival of AML patients



good risk
standard risk
poor risk

t(8;21)
inv(16)

normal karyotype

komplex. karyotype - 7

ACUTE LYMPHOBLASTIC LEUKEMIA (ALL)

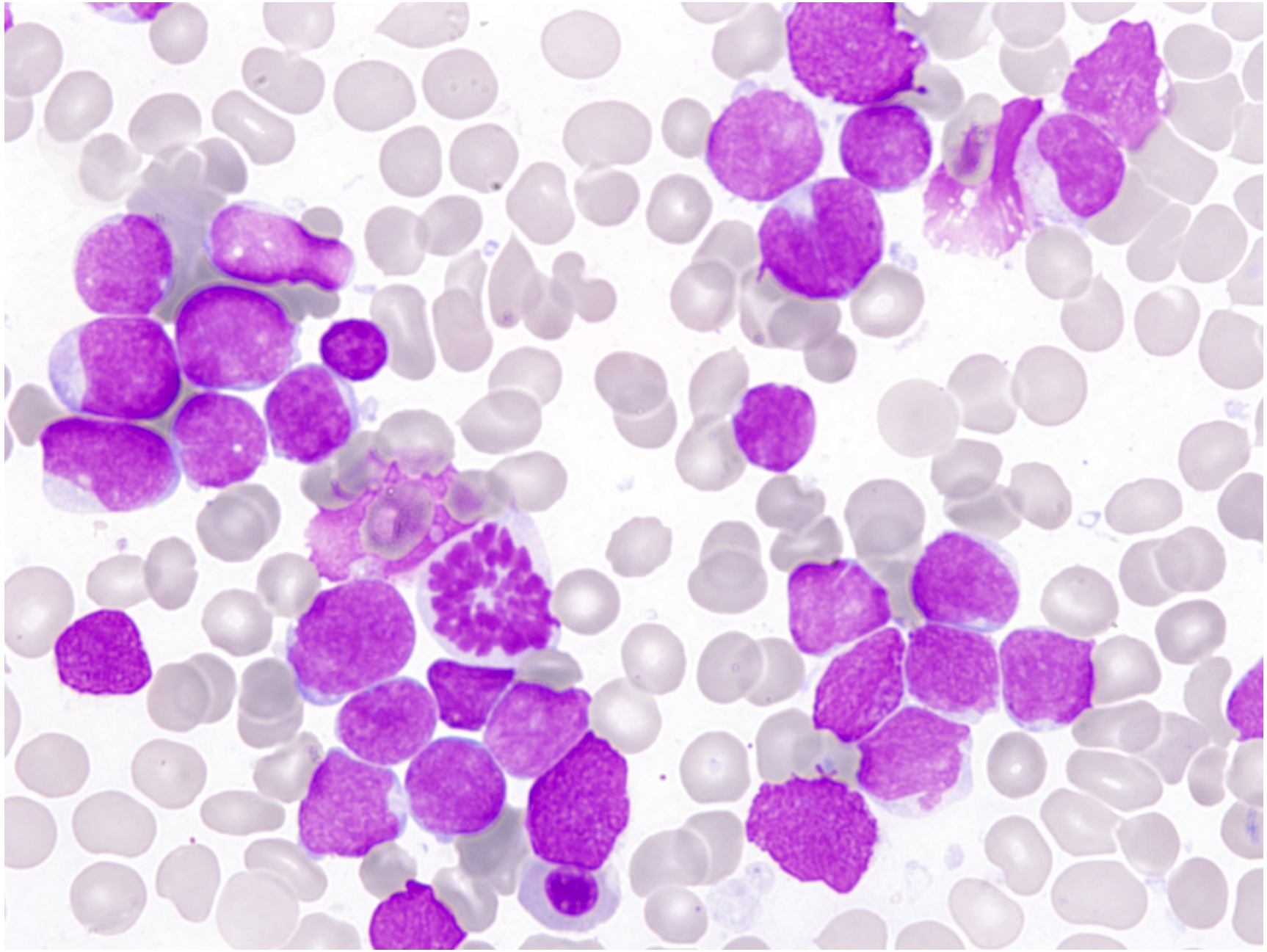
The most common leukemia in childhood.

In children - very good prognosis.
In adults - poorer prognosis.

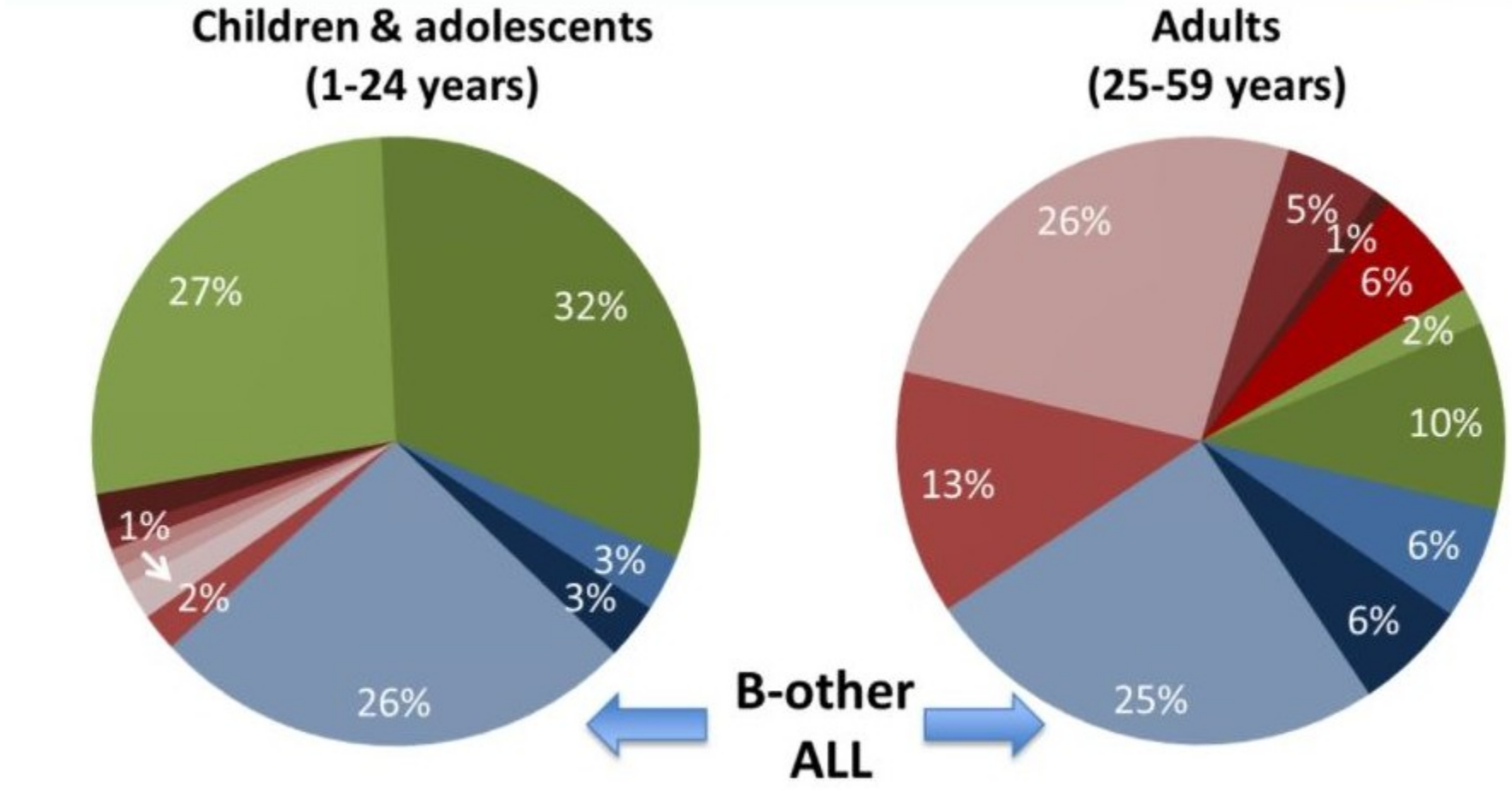
ALL is a neoplastic disease resulting from somatic mutation in a single lymphoid progenitor cell.

B precursor ALL vs. T precurosr ALL

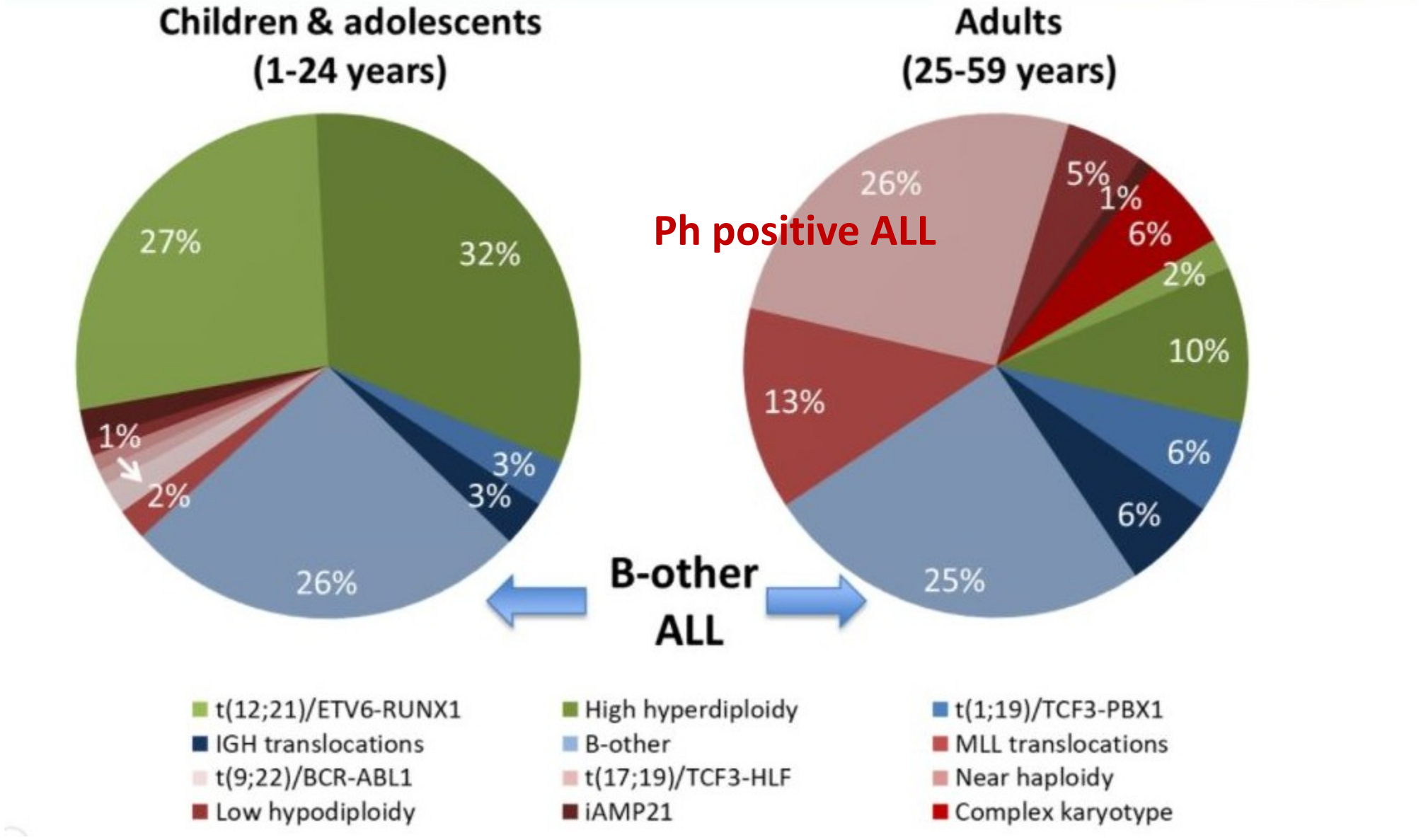
BM - more than 20% of lymphoblasts (usually 80 - 100%).



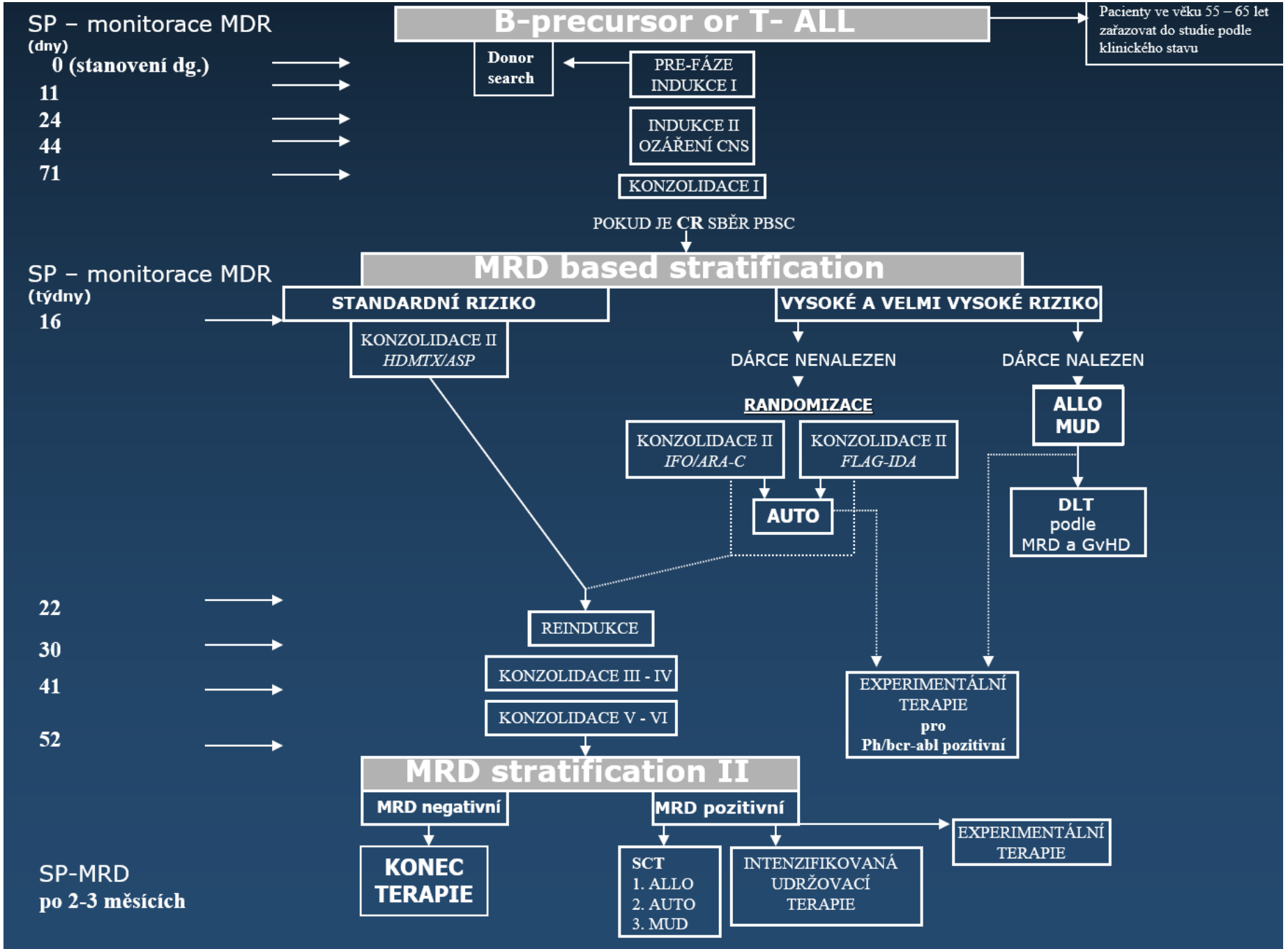
Age specific frequency of genetic subgroups in ALL



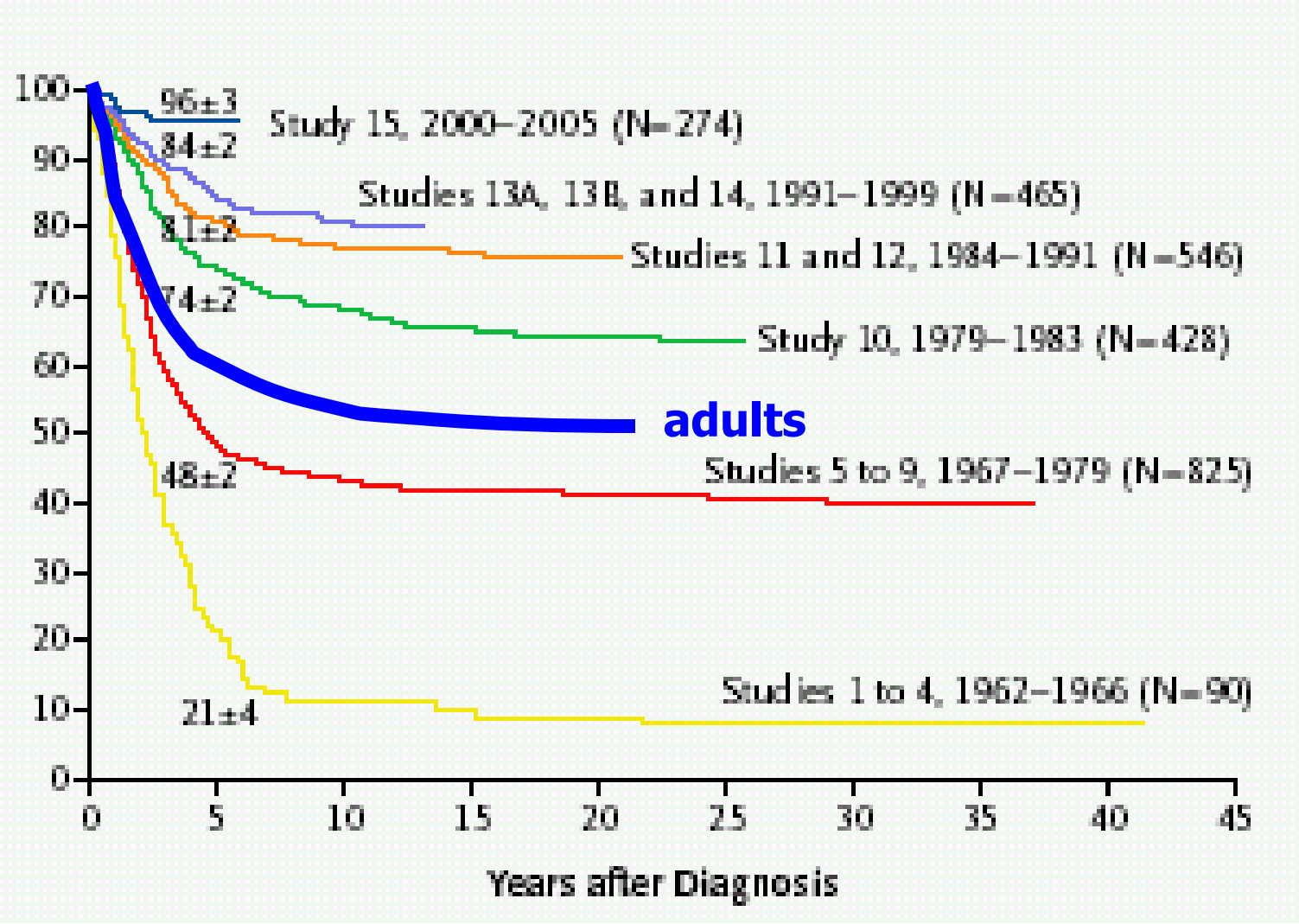
Age specific frequency of genetic subgroups in ALL



ALL – therapy overview



Childhood ALL survival



MYELODYSPLASTIC SYNDROMES

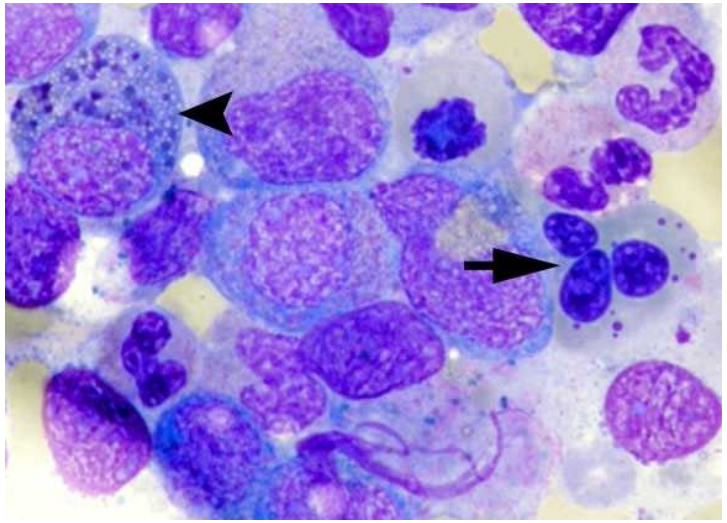
Heterogeneous group of malignant diseases with different prognosis – dysplasia of myeloid lineage.

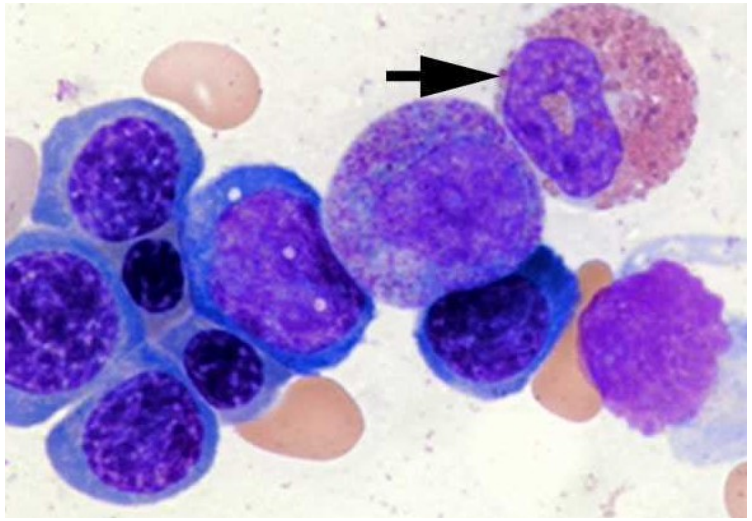
In BM - blasts bellow 20 % and dysplastic features (hypogranular cells, cells with atypical shape of nucleus, hypergranular cells, cells with abnormal plasma)

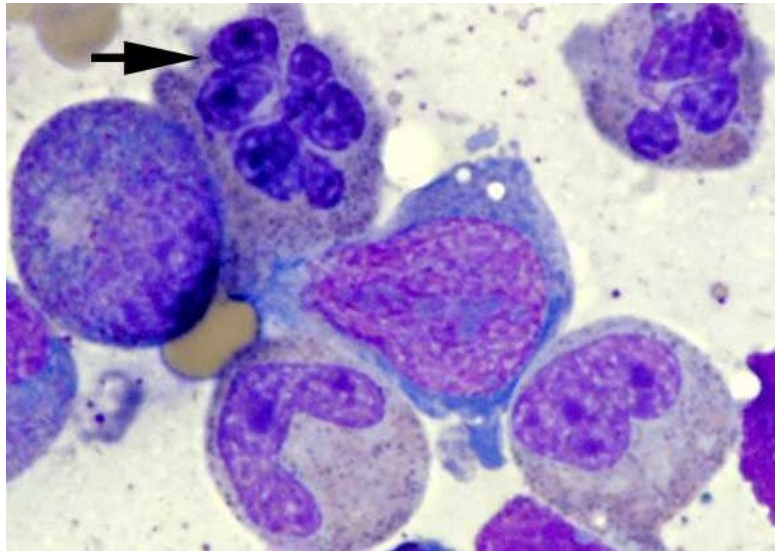
The only curative option is BMT/PBSCT in high risk patients.

Patients asyptomatic or without donor - only symptomatic treatment or watch and wait strategy.

MYELODYSPLASTIC SYNDROMES
- dysplastic features







MDS – classification I

MDS type	Dysplasia	Cytopenia	Ring sideroblasts	Blasts in peripheral blood	Blasts in bone marrow	Cytogenetics
MDS with single lienage dysplasia (MDS-SLD)	1	1 or 2	<15%, < 5%	< 1%, no Auer rods	<5%, no Auer rods	Any except of del(5q)
MDS with mixed lineage dysplasia (MDS-MLD)	2 or 3	1 - 3	<15%, < 5%	< 1%, no Auer rods	<5%, no Auer rods	Any except of del(5q)
MDS with ring sideroblasts (MDS-RS)						
MDS-SLD-RS	1	1 or 2	≥ 15%, ≥ 5%*	< 1%, no Auer rods	<5%, no Auer rods	Any except of del(5q)
MDS-MLD-RS	2 or 3	1 - 3	≥ 15%, ≥ 5%*	< 1%, no Auer rods	<5%, no Auer rods	Any except of del(5q)
MDS with isolated del(5q)	1-3	1-2	No or few	< 1%, no Auer rods	<5%, no Auer rods	del(5q) or 1 more except of -7 or del(7q)

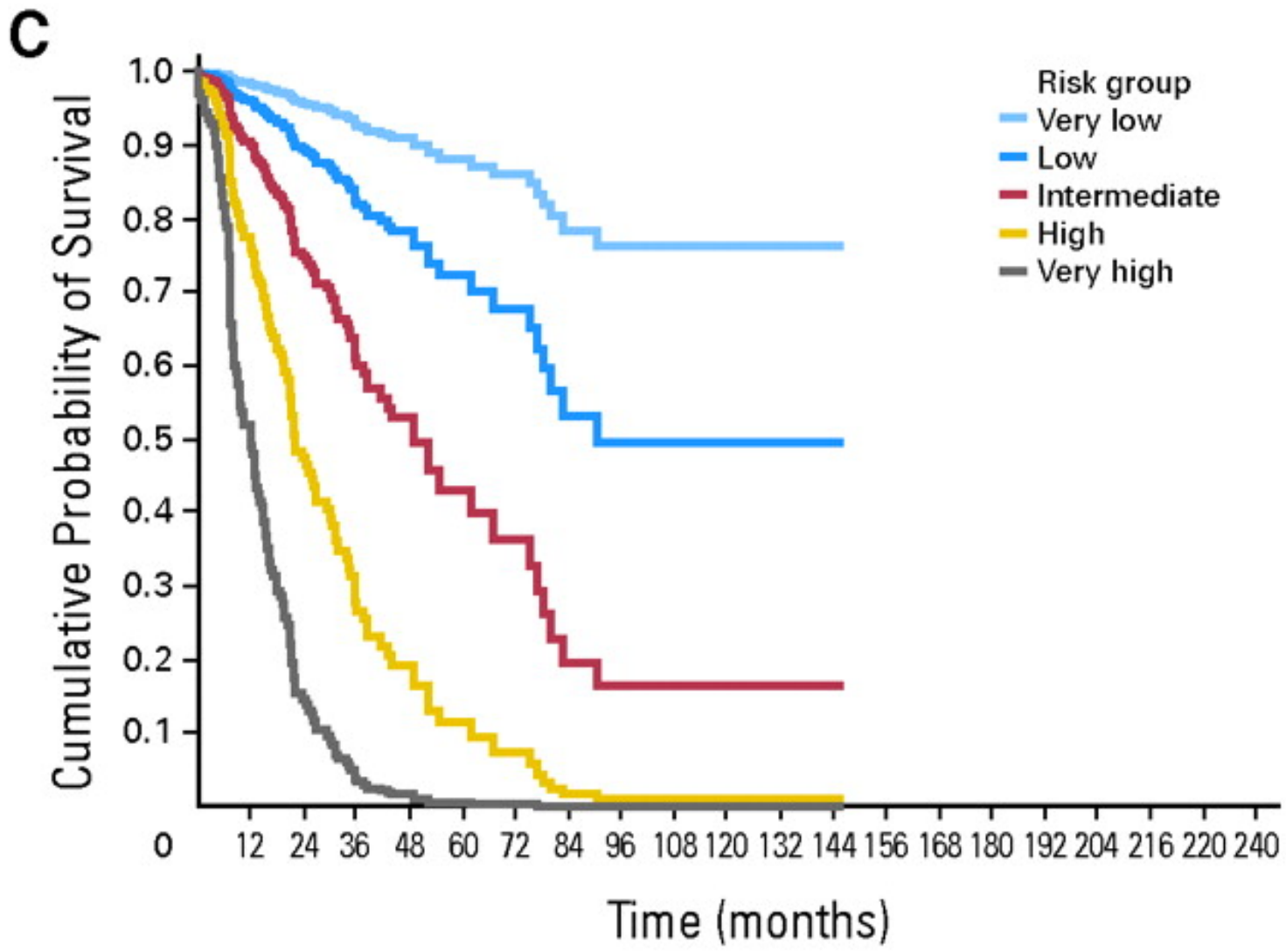
MDS – classification II

MDS type	Dysplasia	Cytopenia	Ring sideroblasts	Blasts in peripheral blood	Blasts in bone marrow	Cytogenetics
MDS with exces of blasts (MDS-EB)						
MDS-EB-1	0-3	1-3	No or few	2-4%, no Auer rods	5-9%, no Auer rods	Any
MDS-EB-2	0-3	1-3	No or few	5-19%, or Auer rods	10-19%, or Auer rods	Any
MDS unclassificable (MDS-U)						
With 1% of blasts in PB	1-3	1-3	No or few	1%, no Auer rods	< 5%, no Auer rods	Any
With 1 lineage dysplasia and pancytopenia	1	3	No or few	< 1%, no Auer rods	< 5%, no Auer rods	Any
With cytogenetic abnormality	0	1-3	< 15%	< 1%, no Auer rods	< 5%, no Auer rods	MDS typical feature
Refractory cytopenia in childhood	1-3	1-3	No	< 2%	< 5%	Any

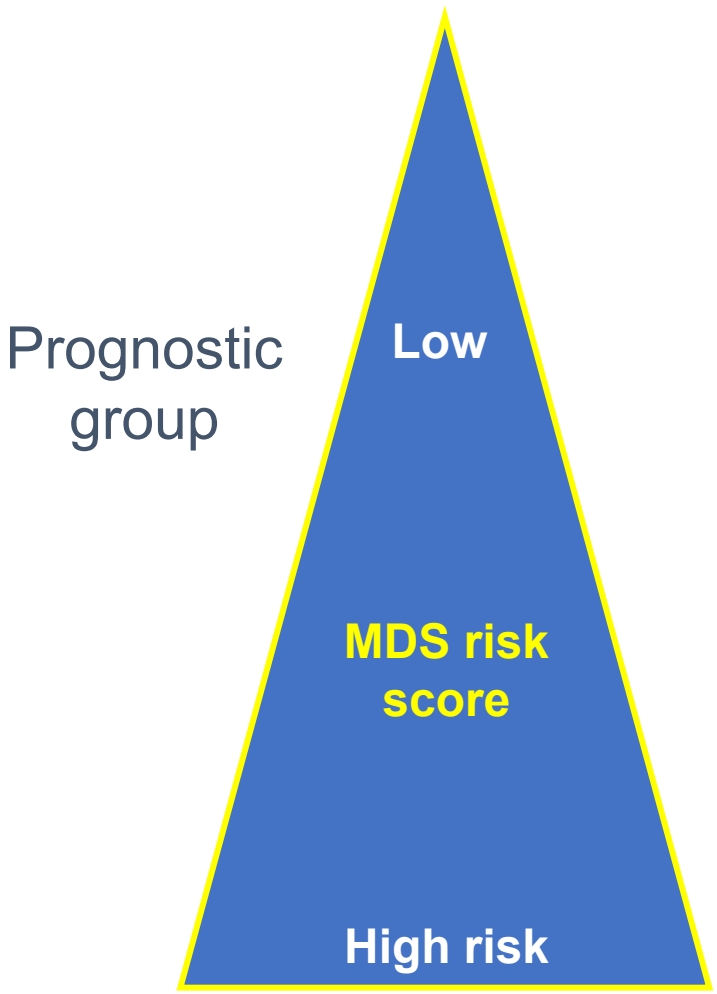
MDS - prognosis

	Score				
Prognostic marker	**0**	**0.5**	**1.0**	**1.5**	**2.0**
Bone marrow blasts (%)	<5	5–10		11–20	21–30
Karyotype	Good	Intermediate	Poor		
Cytopenia	0/1	2/3			

Score	IPSS subgroup	Median survival (years)
0	Low	5.7
0.5–1.0	Int-1	3.5
1.5–2.0	Int-2	1.2
> 2.5	High	0.4

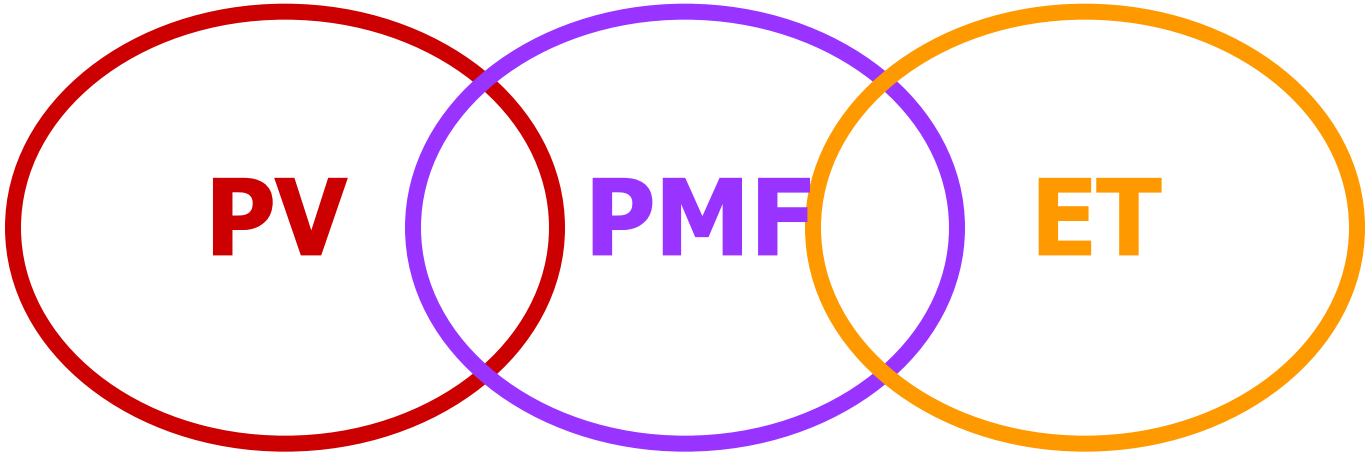

C
Cumulative Probability of Survival
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
12 24 36 48 60 72 84 96 108 120 132 144 156 168 180 192 204 216 220 240
Time (months)
Risk group
Very low
Low
Intermediate
High
Very high

MDS THERAPY



- Supportive care, transfusions, prophylaxis of iron overload
- Erythropoietin
- Immnosupressive therapy
- Low-dose chemotherapy
- Epigenetic therapy (5-azacytidine)
- Allogeneic SCT, clinical trial

MYELOPROLIFERATIVE NEOPLASMS



MYELOPROLIFERATIVE NEOPLASMS

Proliferation of myeloid lineage

(granulocytic, erythroid, megakaryocytic)

MYELOPROLIFERATIVE NEOPLASMS

Myeloproliferative neoplasms (MPN)
Chronic myeloid leukemia (CML), *BCR-ABL1*$^{+}$
Chronic neutrophilic leukemia (CNL)
Polycythemia vera (PV)
Primary myelofibrosis (PMF)
PMF, prefibrotic/early stage
PMF, overt fibrotic stage
Essential thrombocythemia (ET)
Chronic eosinophilic leukemia, not otherwise specified (NOS)
MPN, unclassifiable
Mastocytosis
Myeloid/lymphoid neoplasms with eosinophilia and rearrangement of *PDGFRA*, *PDGFRB*, or *FGFR1*, or with *PCM1-JAK2*
Myeloid/lymphoid neoplasms with *PDGFRA* rearrangement
Myeloid/lymphoid neoplasms with *PDGFRB* rearrangement
Myeloid/lymphoid neoplasms with *FGFR1* rearrangement
Provisional entity: Myeloid/lymphoid neoplasms with PCM1-JAK2

POLYCYTHEMIA

Polycythemia is characterized by an increase of the total red cell volume.
Primary form (PV, clonal neoplastic disorder)
Secondary forms due to appropriate or inappropriate increases in levels of EPO (hemoglobins with high affinity to oxygen, high altitudes, pulomonary and heart diseases, tumours producing EPO)

PV is characterised by increases not only of the number of red cells but also of the granulocytes and platelets and splenomegaly.

POLYCYTHEMIA VERA

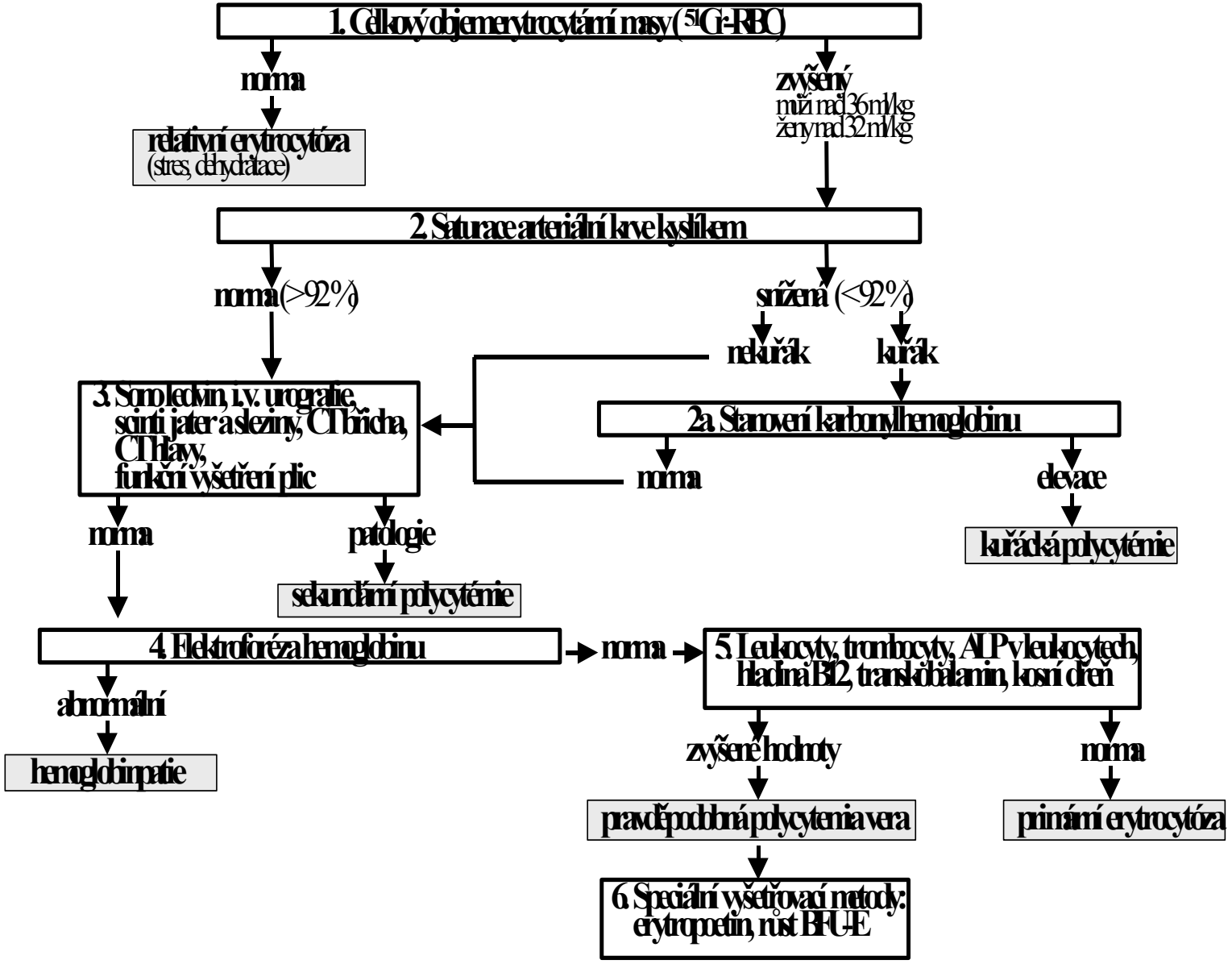
Diagnosis

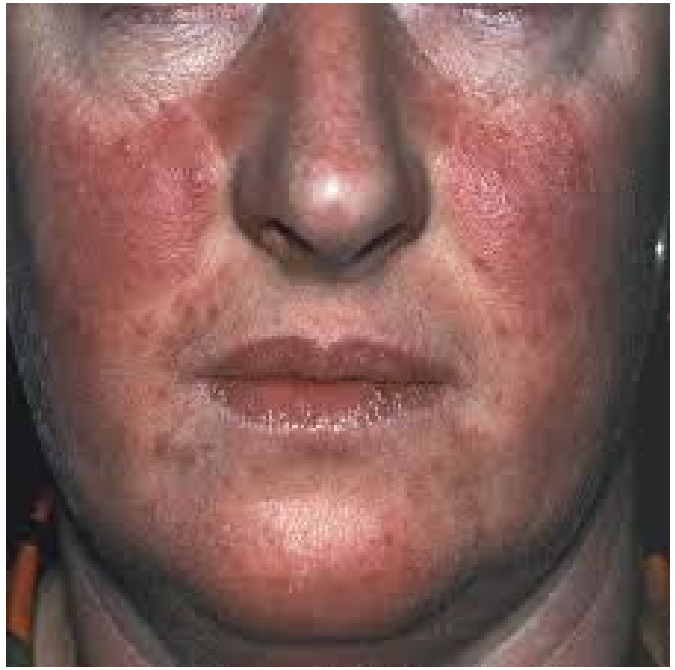
Peripheral blood count
Histology of bone marrow
Total erythrocyte volum
Ertythropoietin level
JAK2 V617F mutation

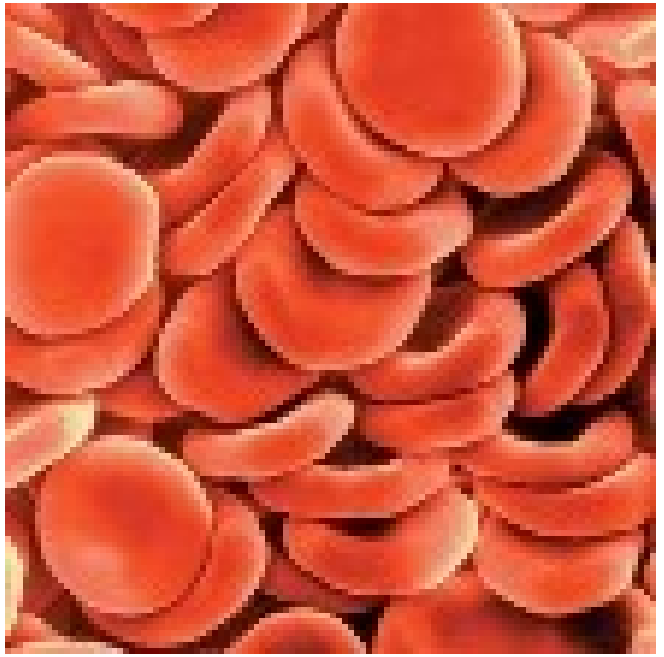
We have to exclude all secondary polycythemias
Secondary polycyhemias are more frequent than PV

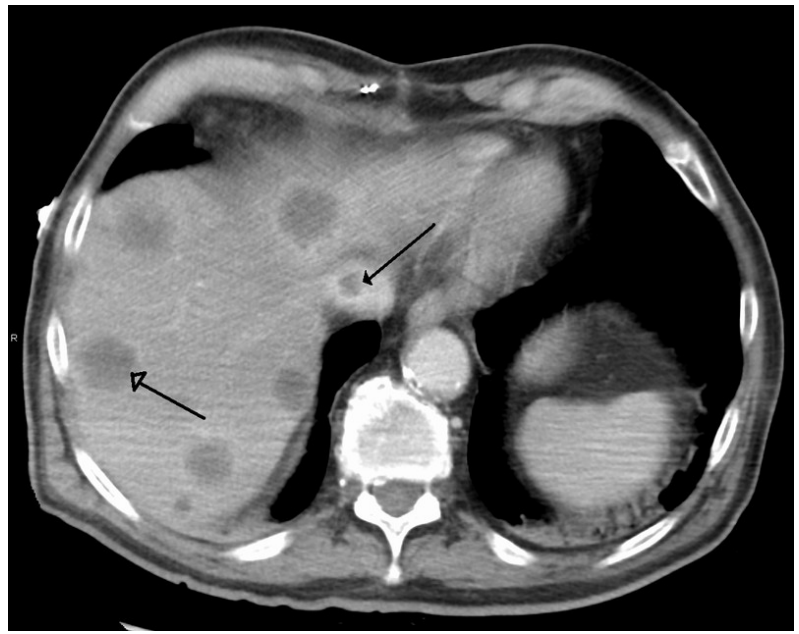
Complications - bleeding, thrombosis, leukemia, bone marrow fibrosis

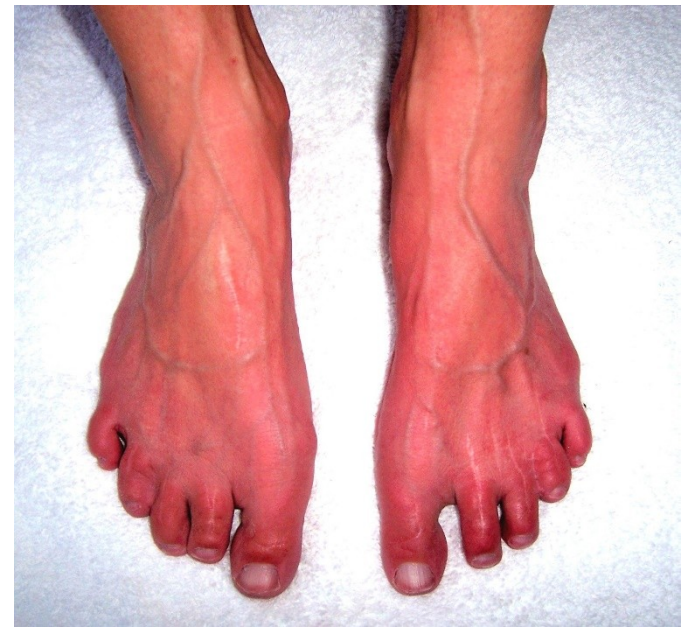
POLYCYTHEMIA VERA – differential diagnosis











POLYCYTHEMIA VERA

Therapy

Phlebotomy
Antiaggregant therapy of anticoagulation therapy

Interferon alpha
Hydroxyurea
Ruxolitinib (JAK2 inhibitor)

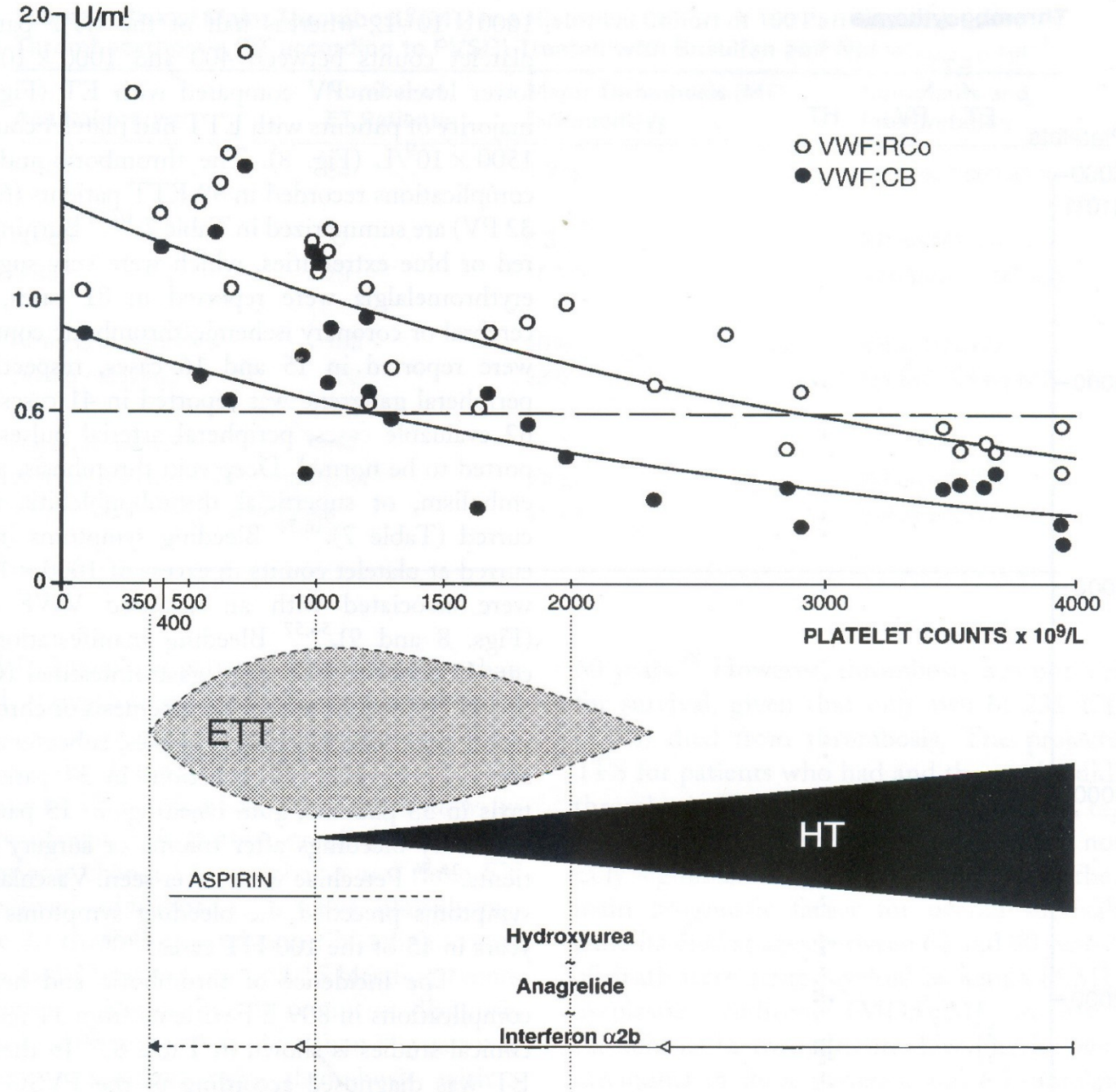
ESSENTIAL THROMBOCYTHEMIA

Clonal proliferation of megakaryocytes in bone marrow and incresed peripheral blood platelet count.
***JAK2 V617F* mutation, calreticulin mutation**

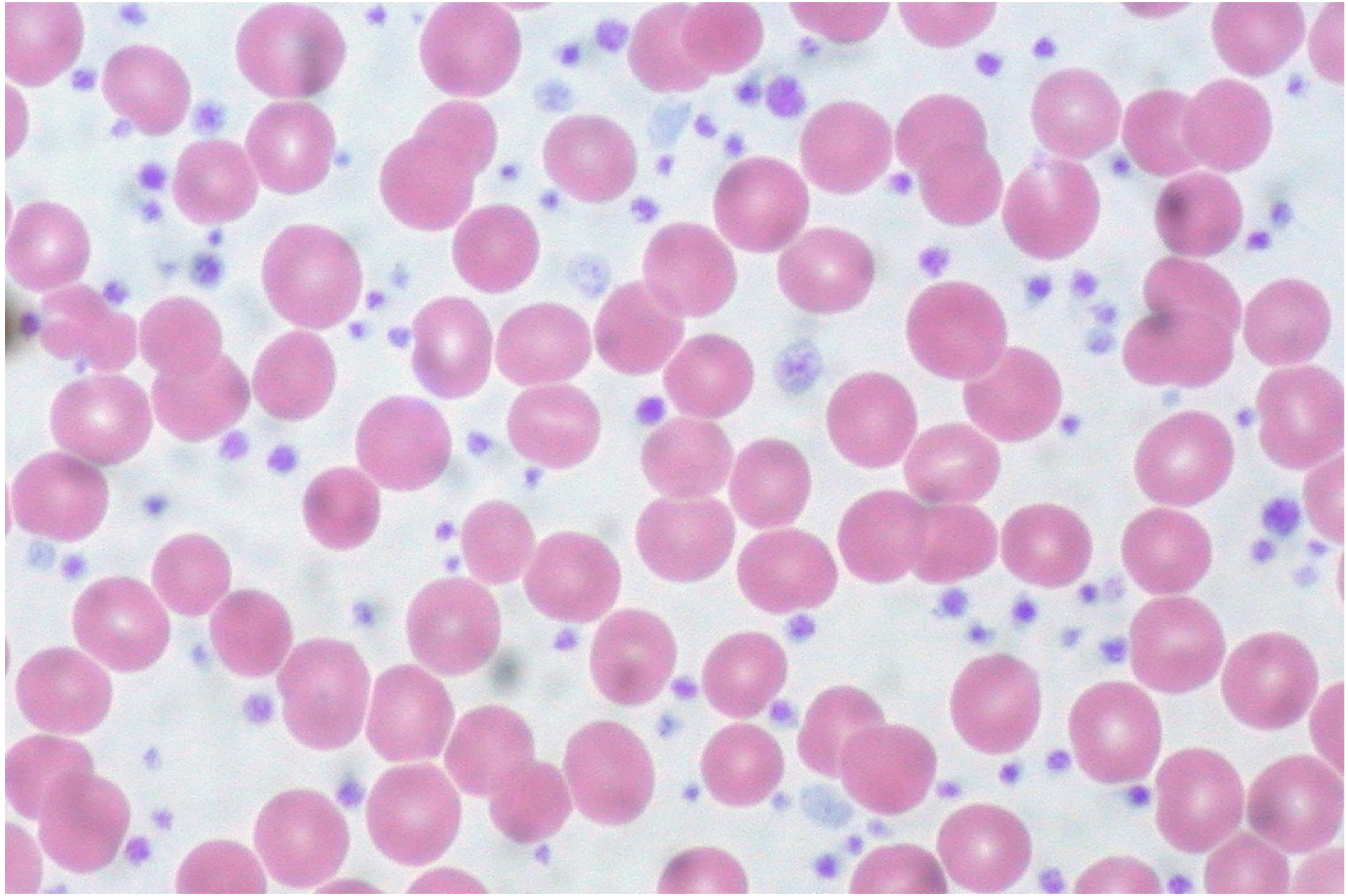
Differential diagnosis:

Secondary thrombocytemias (sideropenia, chronic infection, splenectomy, malignancies, bleeding, hemolysis).
Myeloproliferative disorders, MDS

Complications - bleeding, thrombosis, leukemia, bone marrow fibrosis



ETT: ERYTHROMELALGIC THROMBOTIC THROMBOCYTHEMIA
HT: HEMORRHAGIC THROMBOCYTHEMIA



ESSENTIAL THROMBOCYTHEMIA

Therapy

Antiaggregant therapy of anticoagulation therapy

Interferon alpha
Anagrelide
Hydroxyurea

PRIMARY MYELOFIBROSIS

Clonal disorder chracterized by transformation of normal bone marrow to fibrotic and non-functional bone marrow.
***JAK2 V617F*, *CALR* mutation, *MPL* muation**

Hyperplastic stage - increased precurors of platelets in BM, increased WBC, RBC and PLT.

Late stage – fibrosis (extramedullary hematopoiesis leading to massive splenomegaly).

Prognosis – median shorter than in PV or ET.

PRIMARY MYELOFIBROSIS

Therapy

Interferon alpha
Anagrelide
Hydroxyurea

JAK2 inhibitors (ruxolitinib)

Supportive care

Allogeneic transplantation

Lymphoma

LYMPHOMA

Lymhoid tissue involvement (lymph nodes, other lymphoid tissue)

- Mature B cell neoplasms
- Mature T cell and natural killer (NK) cell neoplasms
- Precursor lymphoid neoplasms
- Hodgkin lymphoma
- Immunodeficiency-associated lymphoproliferative disorders

LYMPHOMA - symptoms

Local expansion symptoms

Systemic symptoms

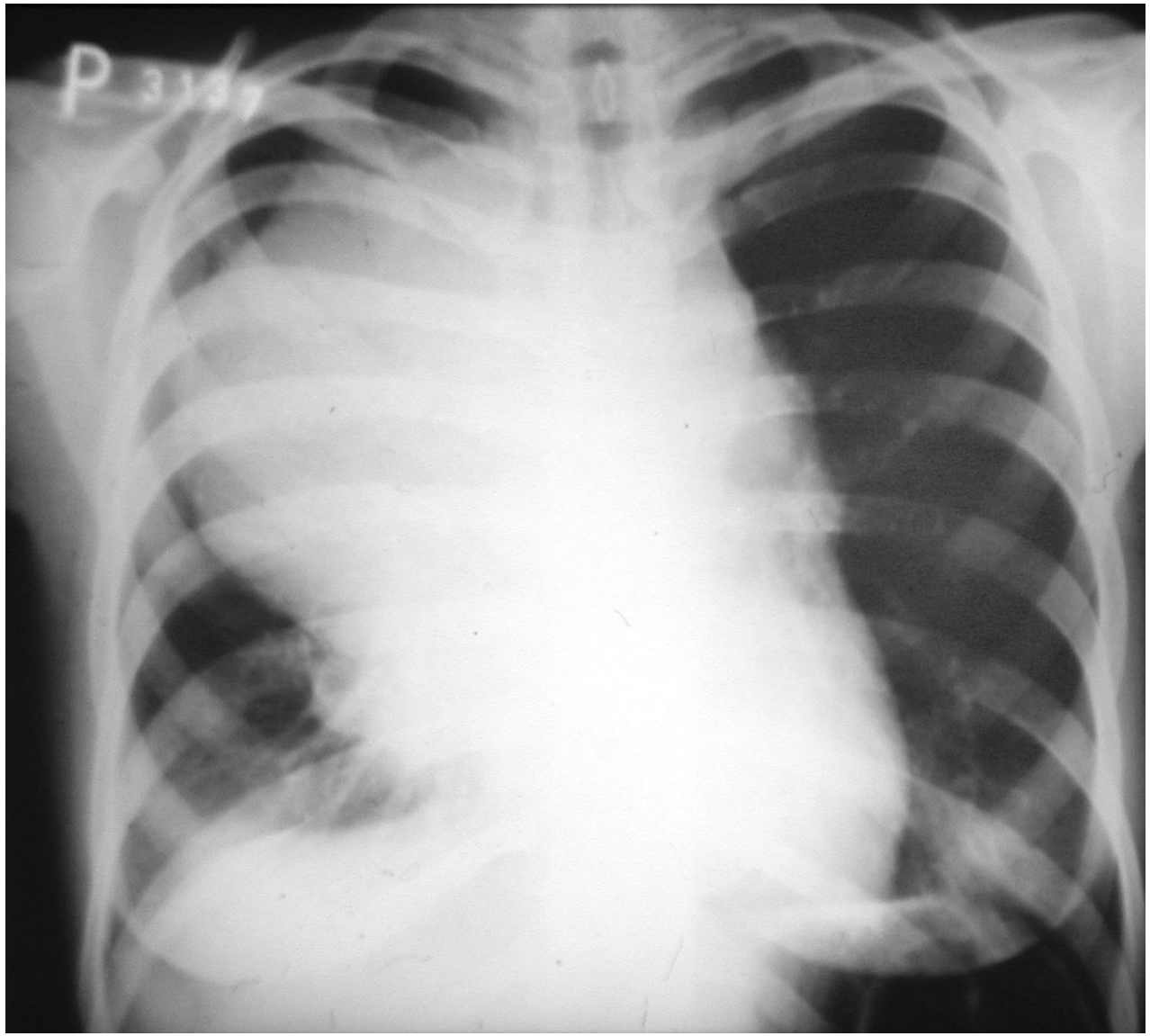
Weight loss
Subfebrilia, fever
(>3 weeks)
Pruritus
Night sweat
Fatigue

LYMPHOMA - symptoms

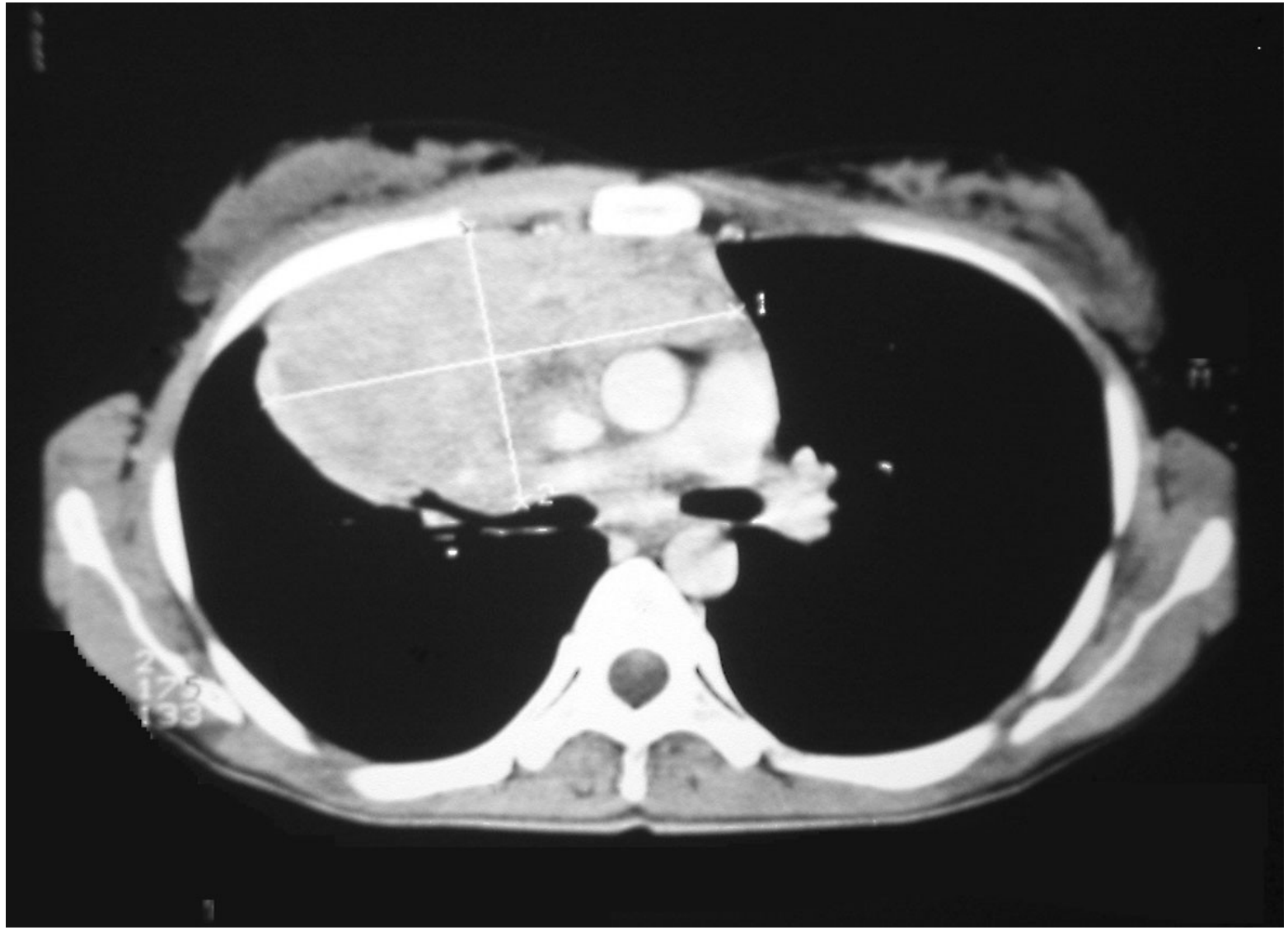
Local expansion symptoms

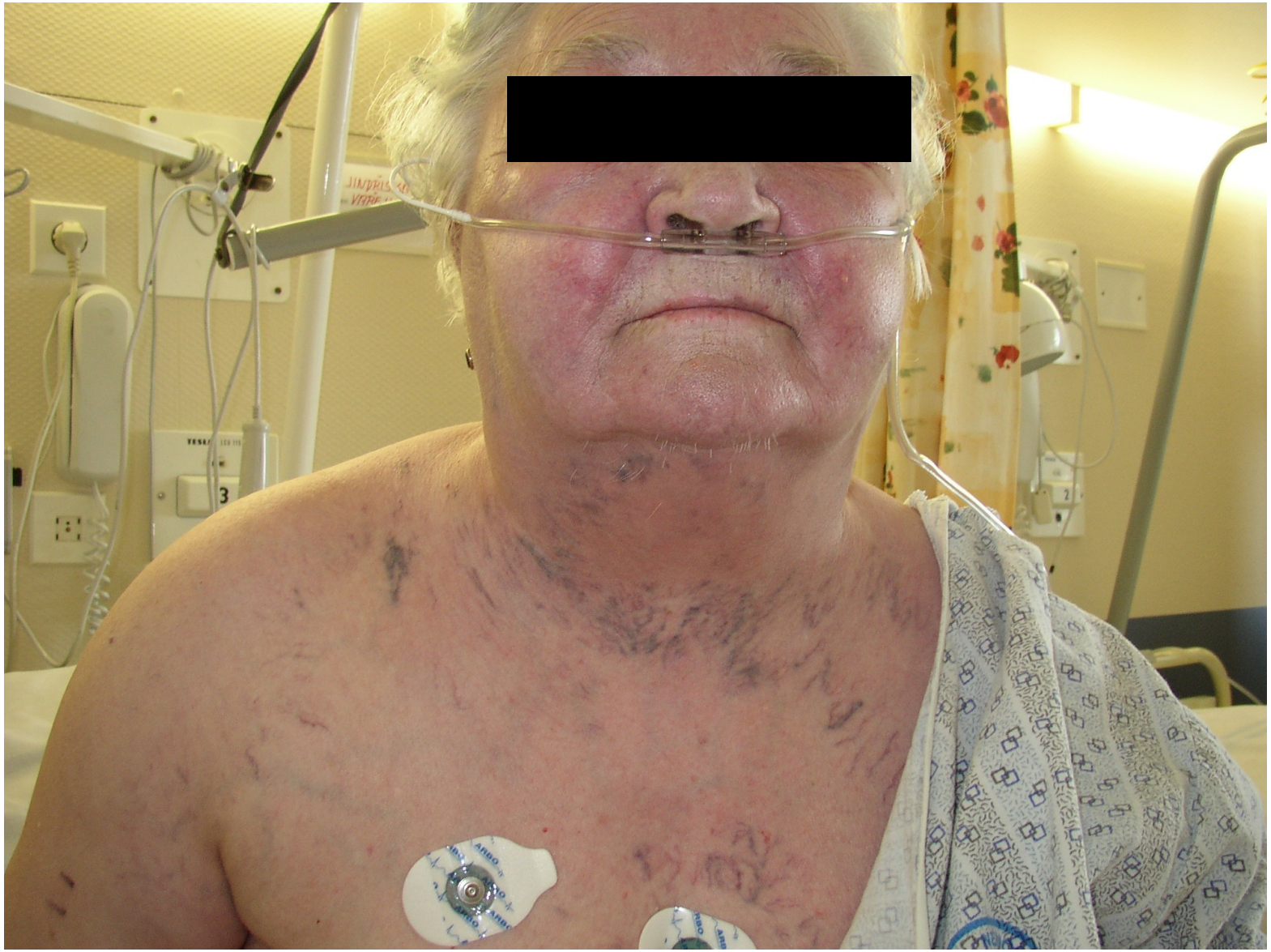
Lymphadenopathy peripheral
Lymphadenopathy mediastinal
(cough, feeling of pressure in the chest, upper vena cava syndrome)
Lymphadenopathy abdominal
(hydronefrosis, abdominal dyscomfort)
Splenomegaly
(abdominal dyscomfort, quick feeling of satiety)
Bone marrow involvment
(cytopenia)
Osteolytic bone leasions

LYMPHOMA - symptoms

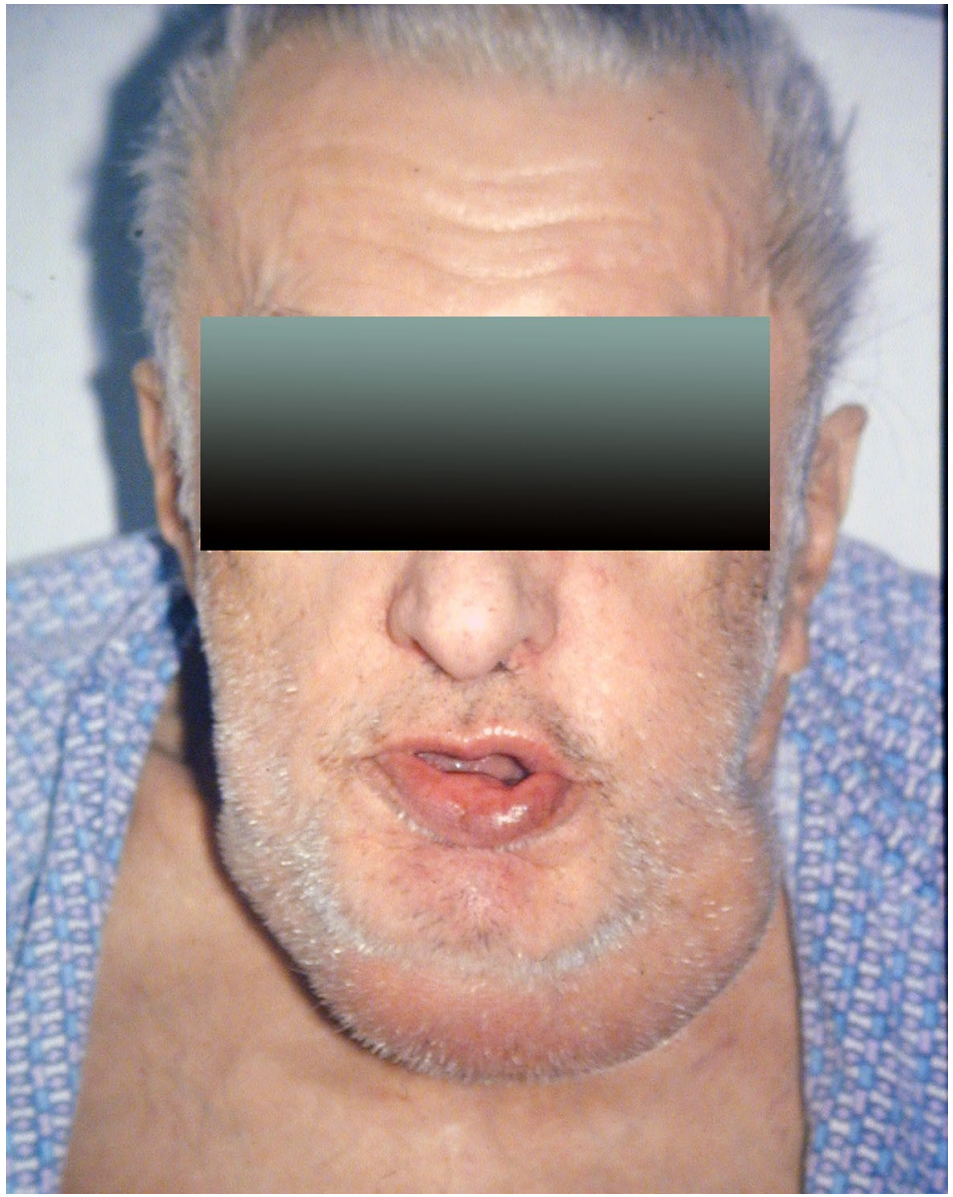


LYMPHOMA - symptoms

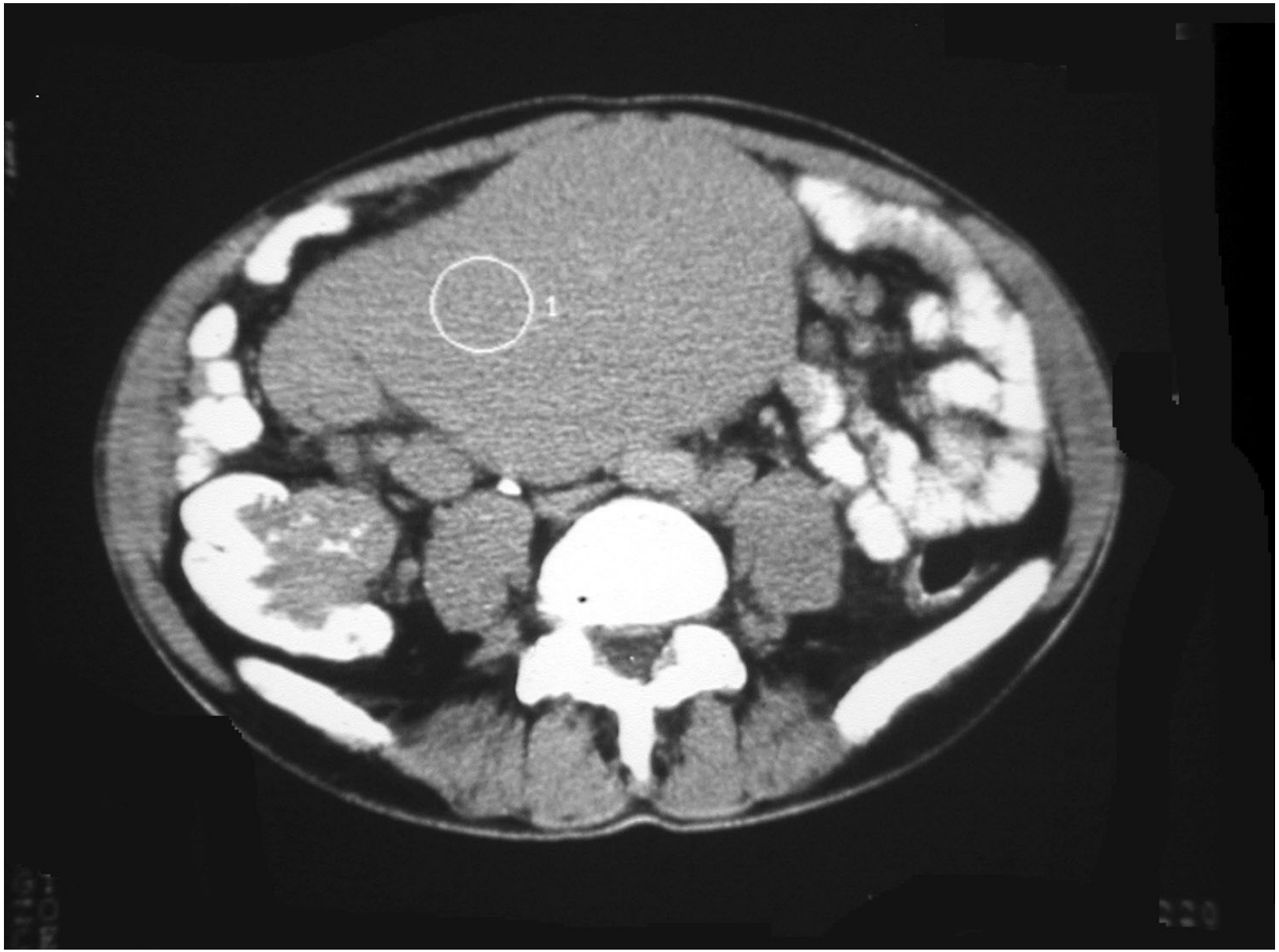




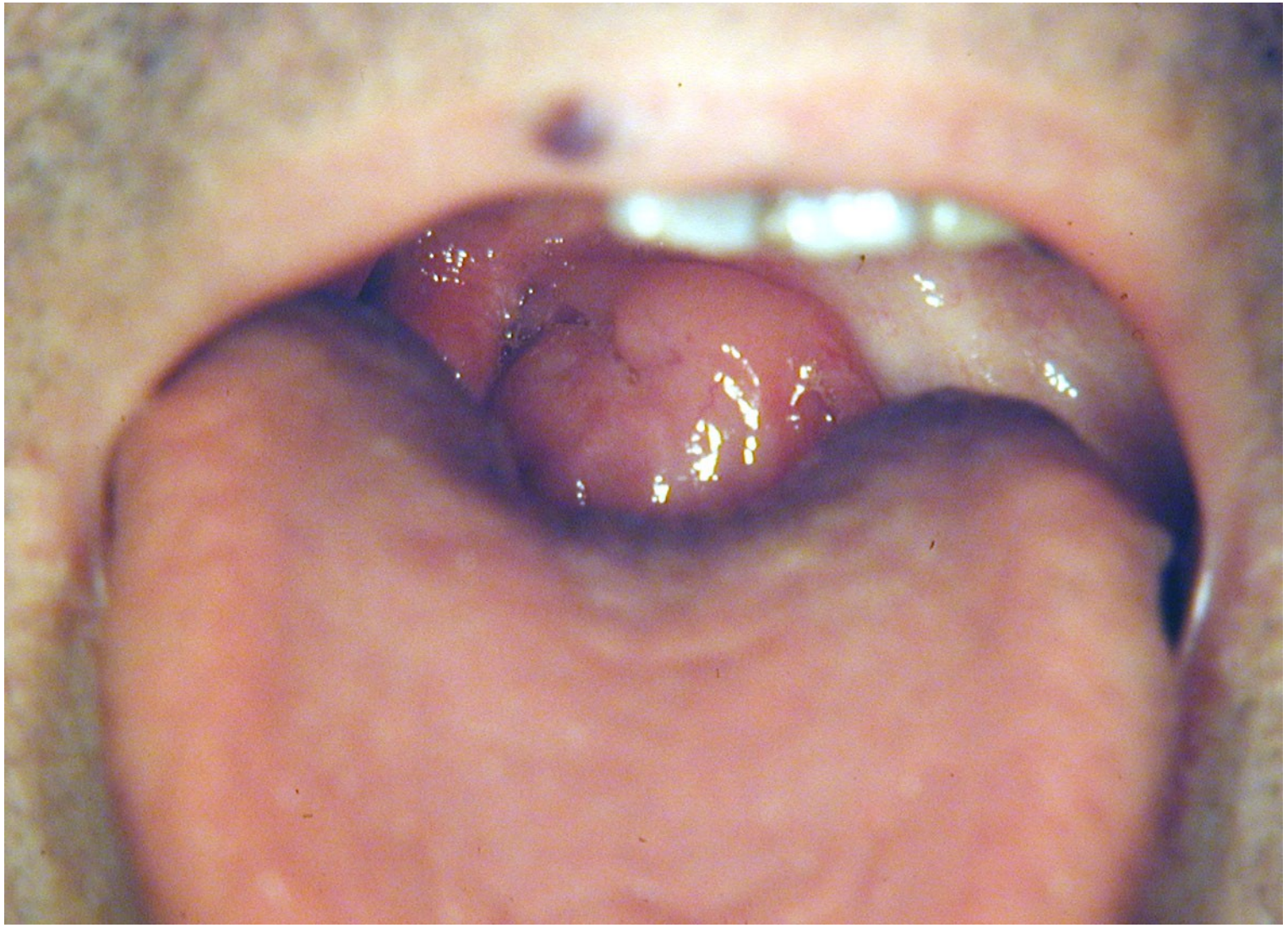
LYMPHOMA - symptoms



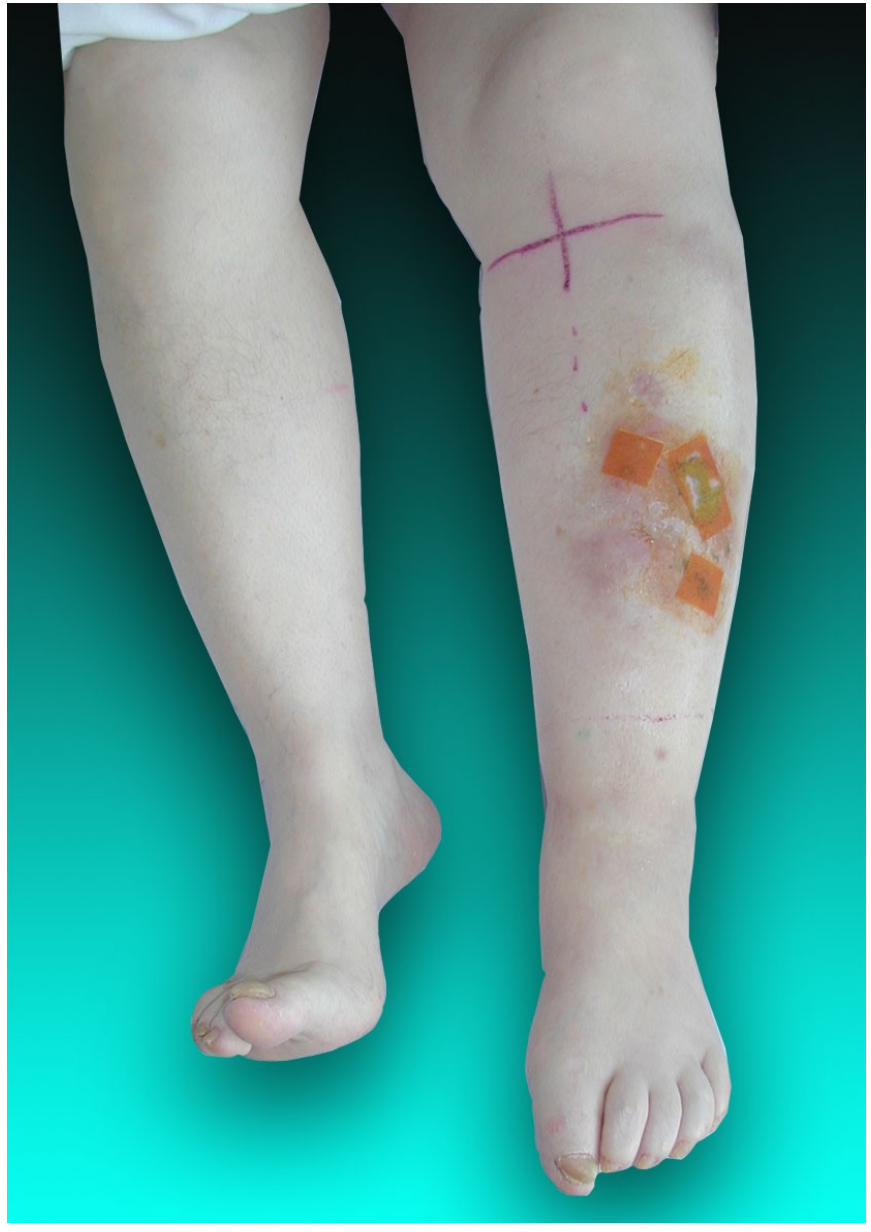
LYMPHOMA - symptoms



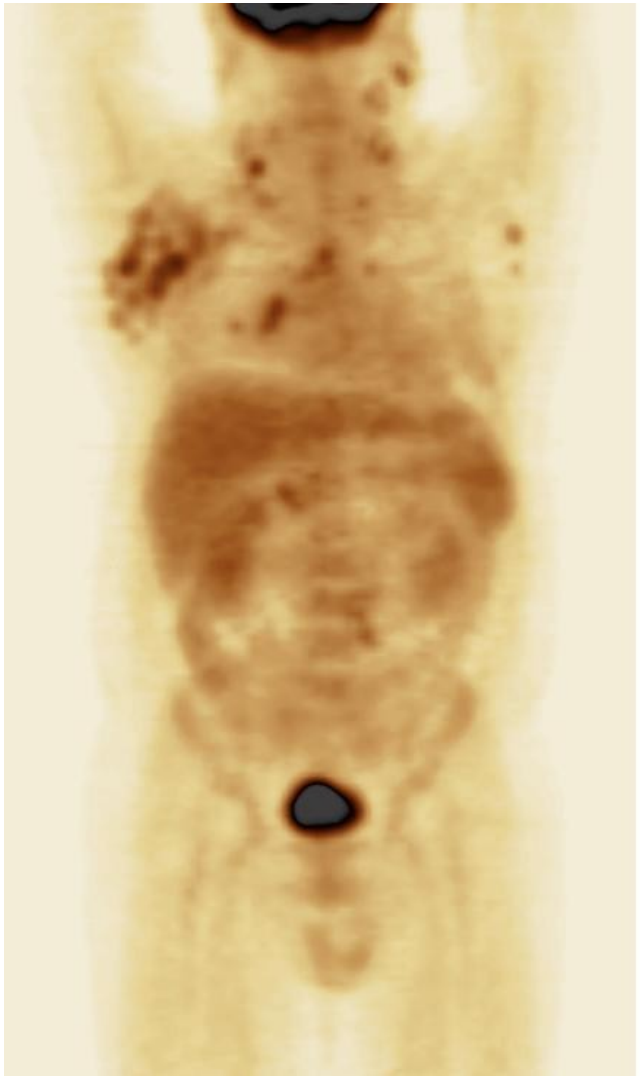
LYMPHOMA - symptoms

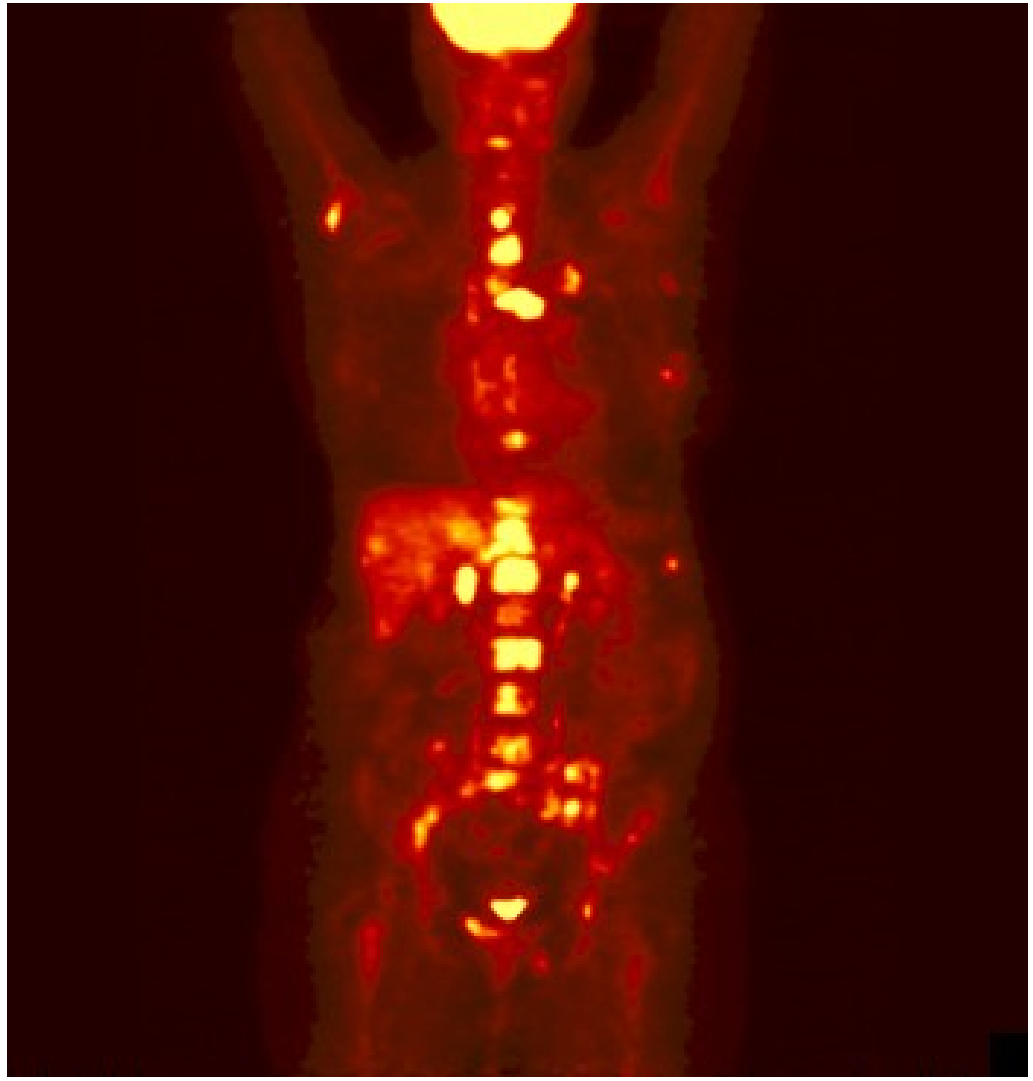


LYMPHOMA - symptoms



LYMPHOMA - symptoms





NON-HODGKIN LYMPHOMA

- **Mature B cell neoplasms**
- **Mature T cell and natural killer (NK) cell neoplasms**

- **Lymph node involvment**
- **Extranodal lymphoma**

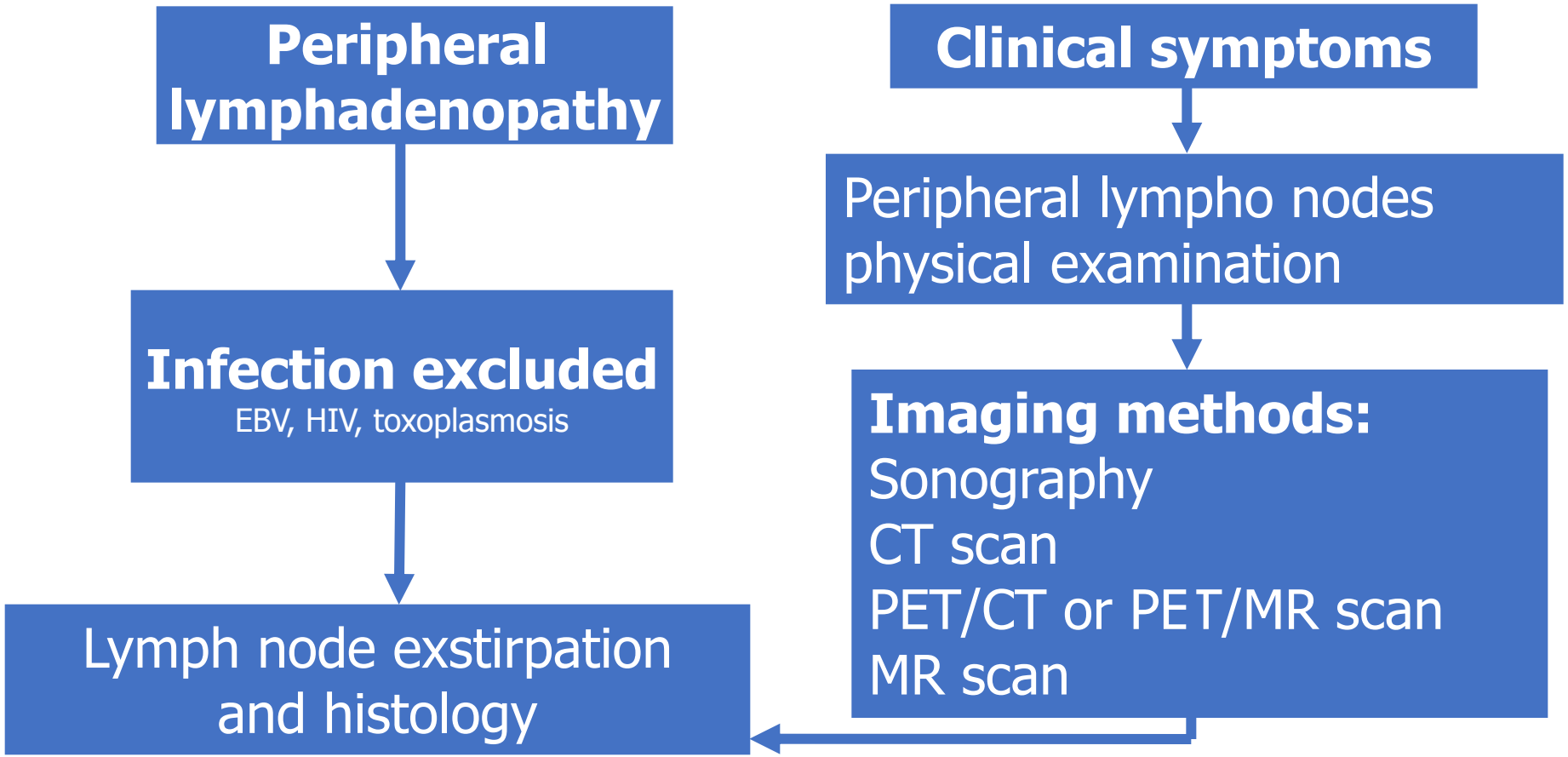
Indolent NHL
slow growth - remission possible, cure unlikely
= start of treatment only with symptoms

Aggressive NHL
potentially curable, treatment start as soon as possible

Very aggressive NHL

NON-HODGKIN LYMPHOMA

Diagnostics



NON-HODGKIN LYMPHOMA

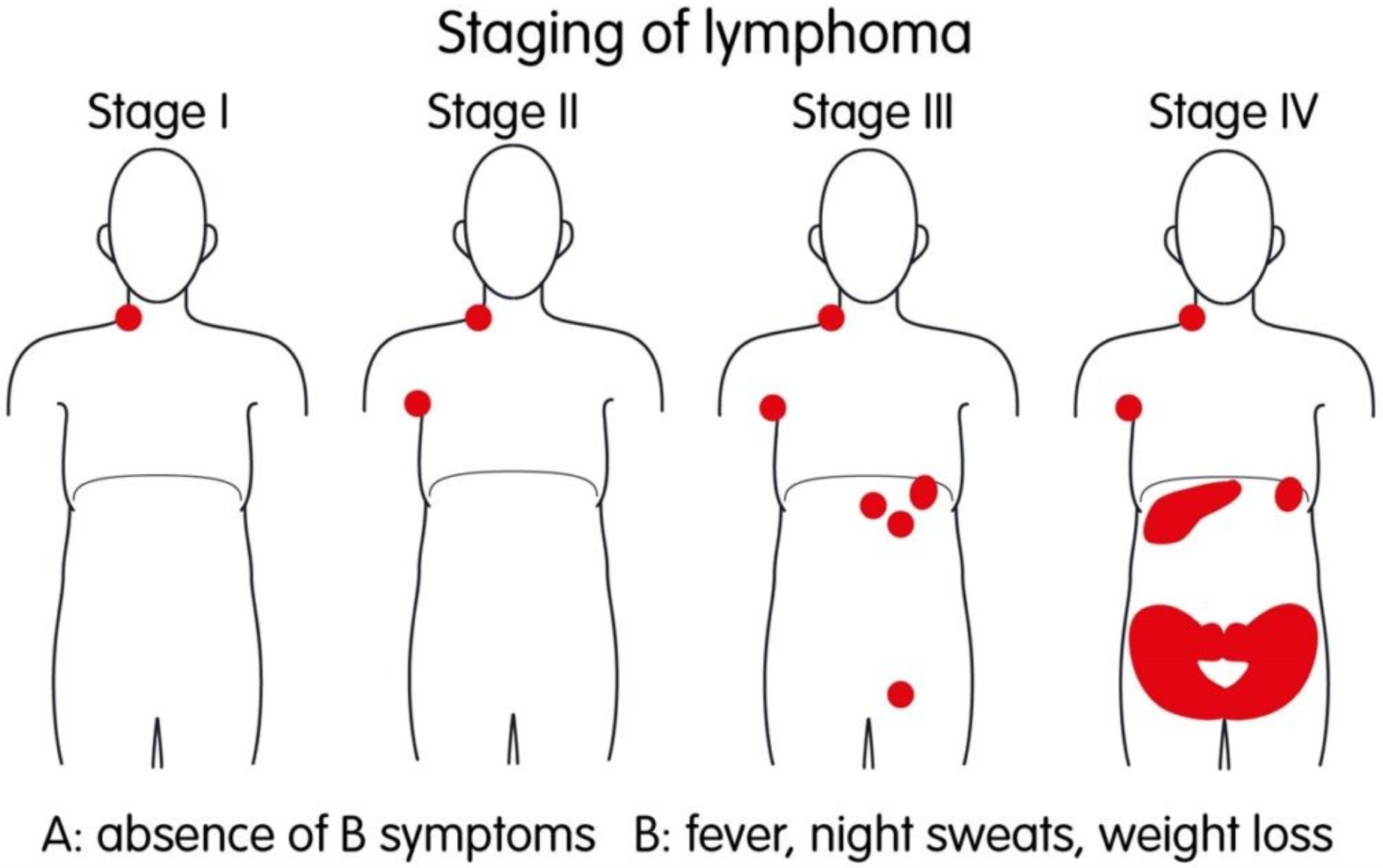
Staging

CT (neck, upper arms, chest, abdomen and pelvis)
- or MRI
- or now PET/CT, alternatively PET/MR

Trephine biopsy and bone marrow histology

Where appropriate, a specialized examination (gastroscopy, colonoscopy, lumbal puncture...)

NON-HODGKIN LYMPHOMA



NON-HODGKIN LYMPHOMA

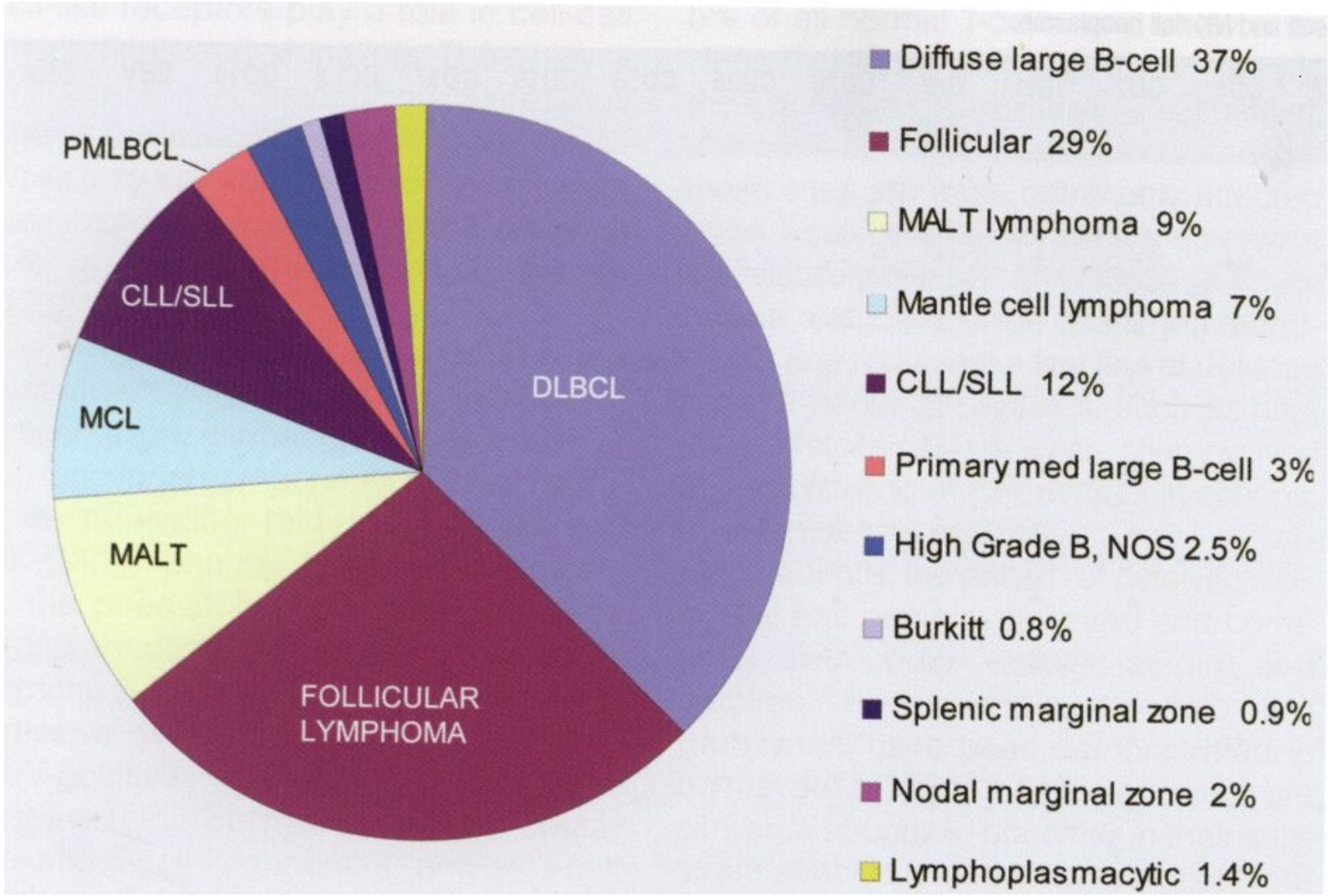
Prognostication

Stage I and II = **limited stage**

Stage III and IV = **advanced stage**
(several prognostic indexes for adnaced stage – IPI, FLIPI…)

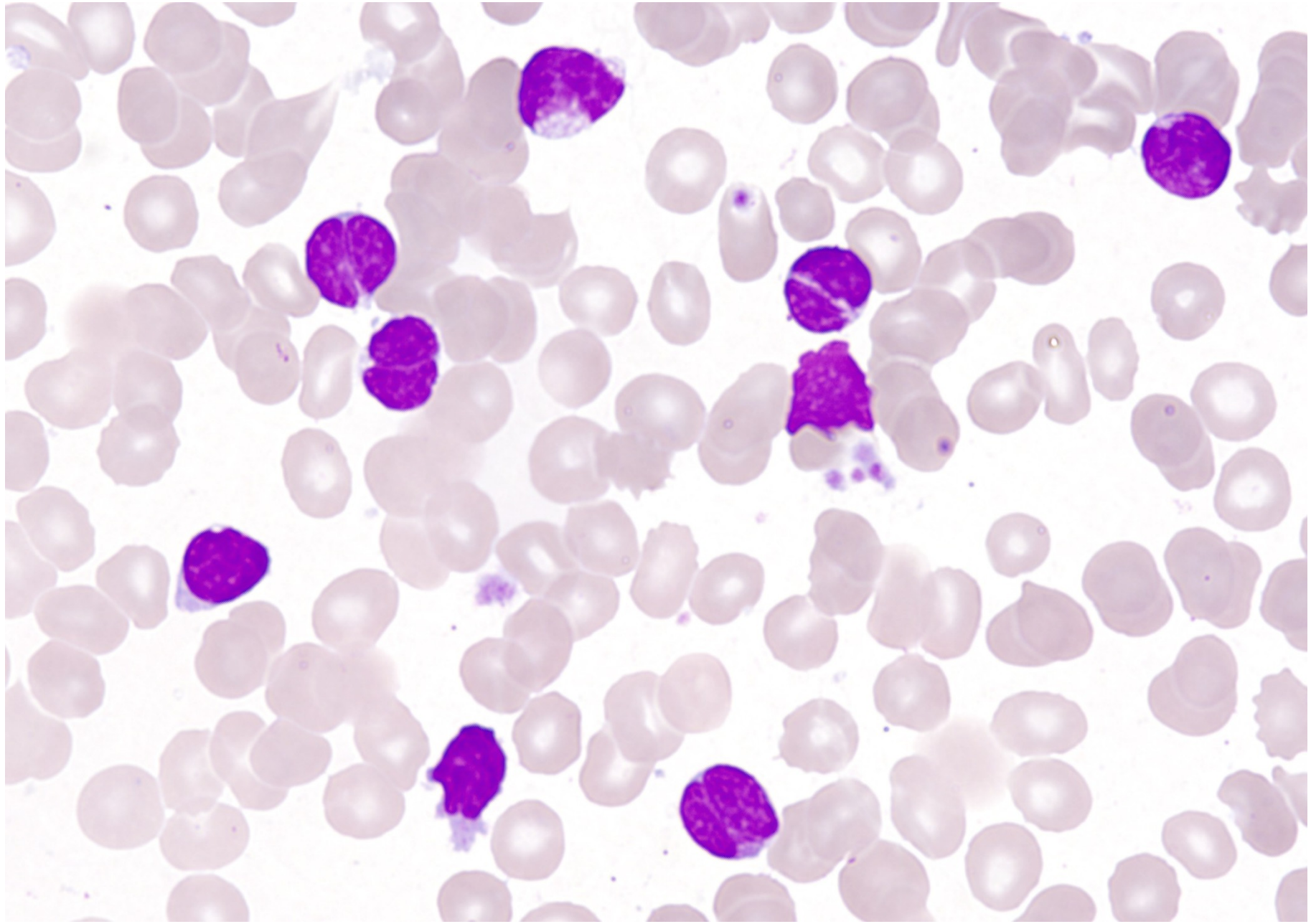
NON-HODGKIN LYMPHOMA

B-cell NHL subtypes

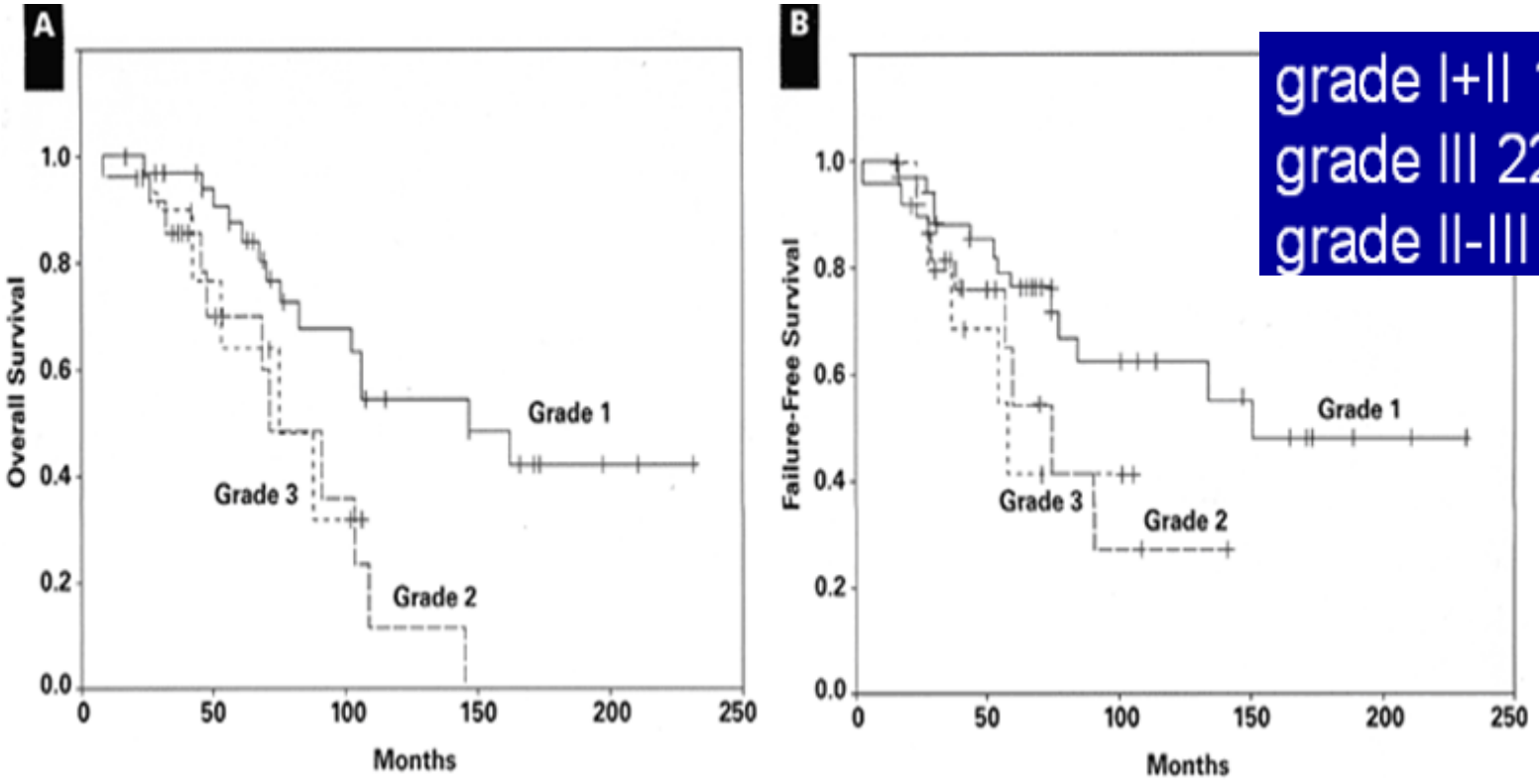


Indolent NHL - folicular lymphoma

- Survival without treatment several years in many patients
- Radiotherapy for limited stage (I.-II. st.) has curative potential
- Systemic treatment leading to remission, but no cure; repeatedly relapsed disease
- Systemic treatment in symptomatic patients only



Folicular lymphoma prognosis according to histology

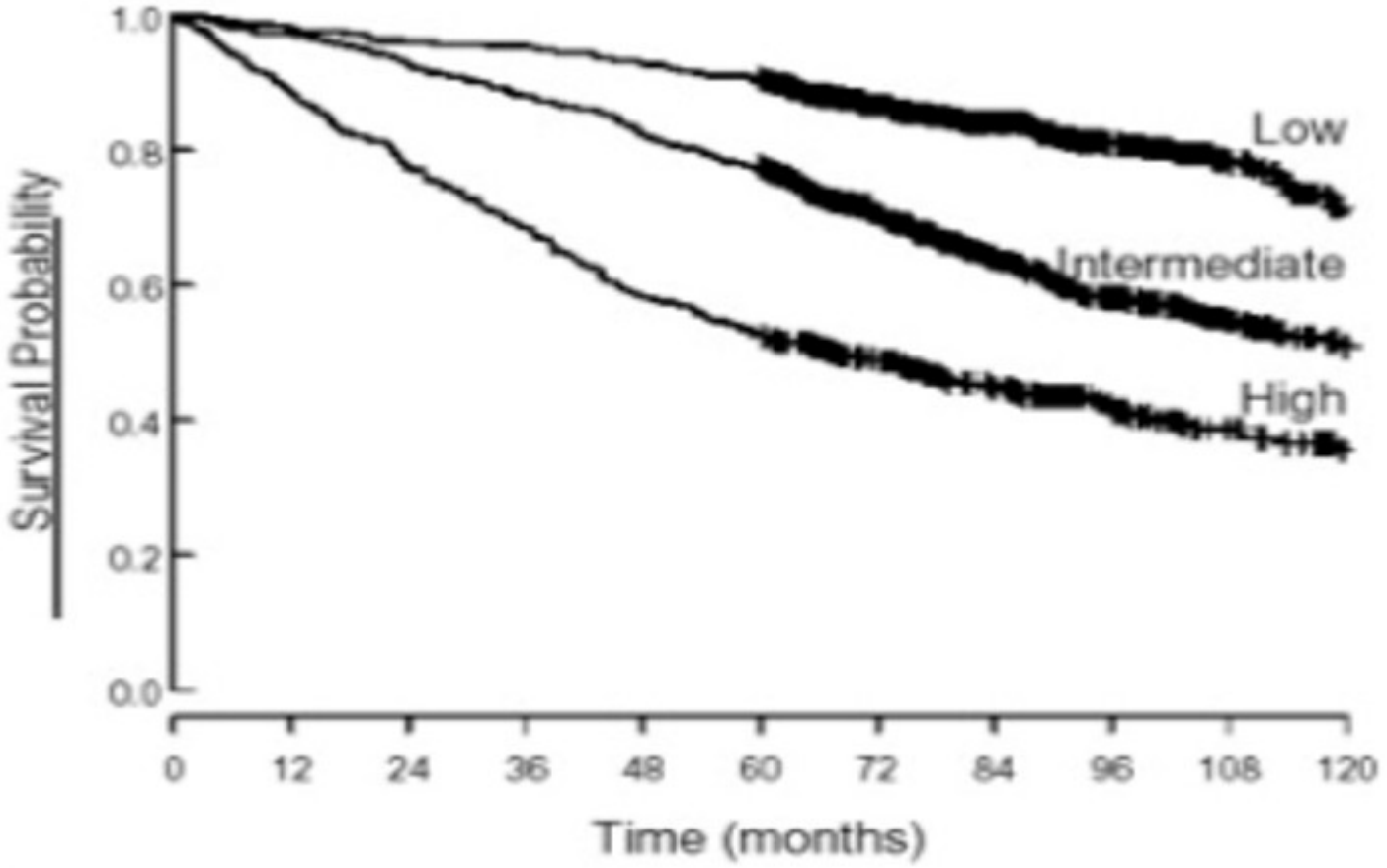


grade I+II 151/178 (85%)
grade III 22/178
grade II-III 5/178

Folicular lymphoma prognostic index (FLIPI)

- Hemoglobin below 120 g/L
- Age over 60 years
- LDH above norm
- Stage II B or higher
- Involved lympho ode areas over 4

Low – FLIPI 0-1
Intermediate – FLIPI 2
High – FLIPI 3 and higher



Folicular lymphoma therapy

First-line therapy

- Limited FL (stadia I+II): IF RT 25-35Gy
- Advanced FL (stadia III+IV): anti-CD20 antibody + chemotherapy (R-CHOP regimen…)

Therapy of relapse

- Chemoimmunotherapy with anti-CD20 antibody +/- maintenance with monoclonal antibody
- High-dose therapy and autologous bone marrow transplant
- Allogeneic bone marrow transplant
- Radioimmunotherapy
- Radiotherapy (limited forms)

Indolent NHL - MALT lymphoma

- MALT – Mucosa Associated Lymphatic Tissuse lymphoma
- Ethiologic role of antigen stimulation, *H. pylori* infection
- Majority: MALT lymphomas of stomach
- Symptoms: non-healing stomach ulcers

MALT lymphoma therapy

Limited clinical stages (I or II)

- Antibiotics, radiotherapy (surgery as alternative)

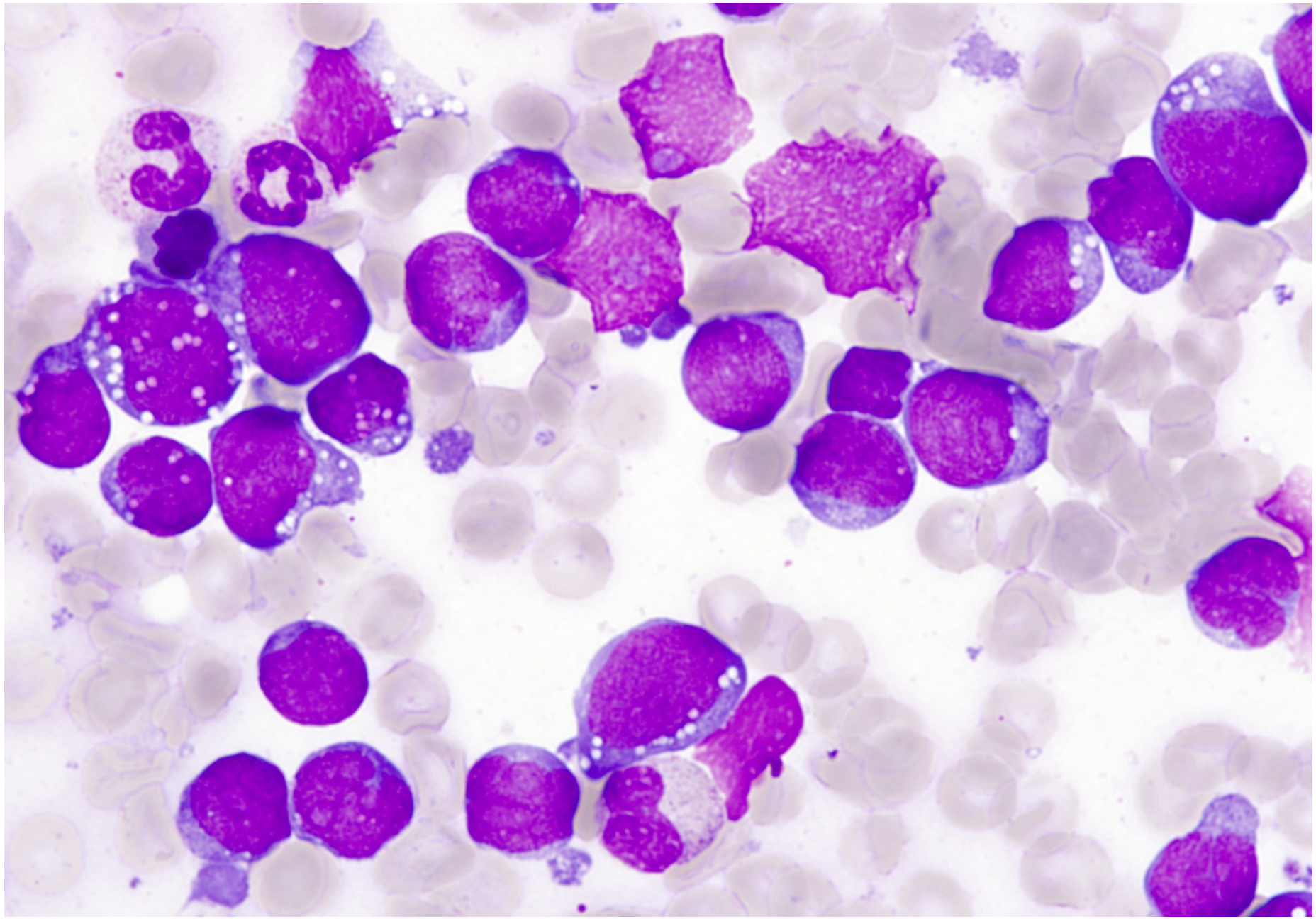
Generalized clinical stages III or IV

- Chemoimmunotherapy (as in folicular lymphoma)

Aggresive NHL – principles of therapy

- **Paliative**
  - Mantle cell lymphoma

- **Curative**
  - DLBCL
  - Burkitt lymphoma



Aggresive NHL - DLBCL

Diffuse large B-cell lymphoma

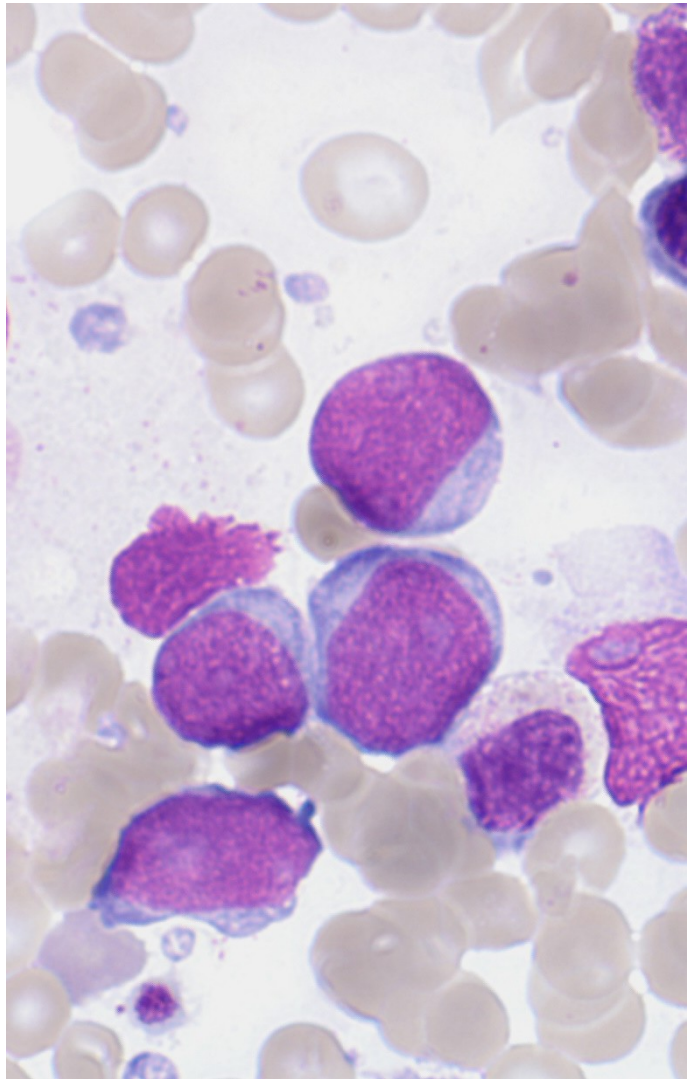
The most common lymphoma

Symptoms

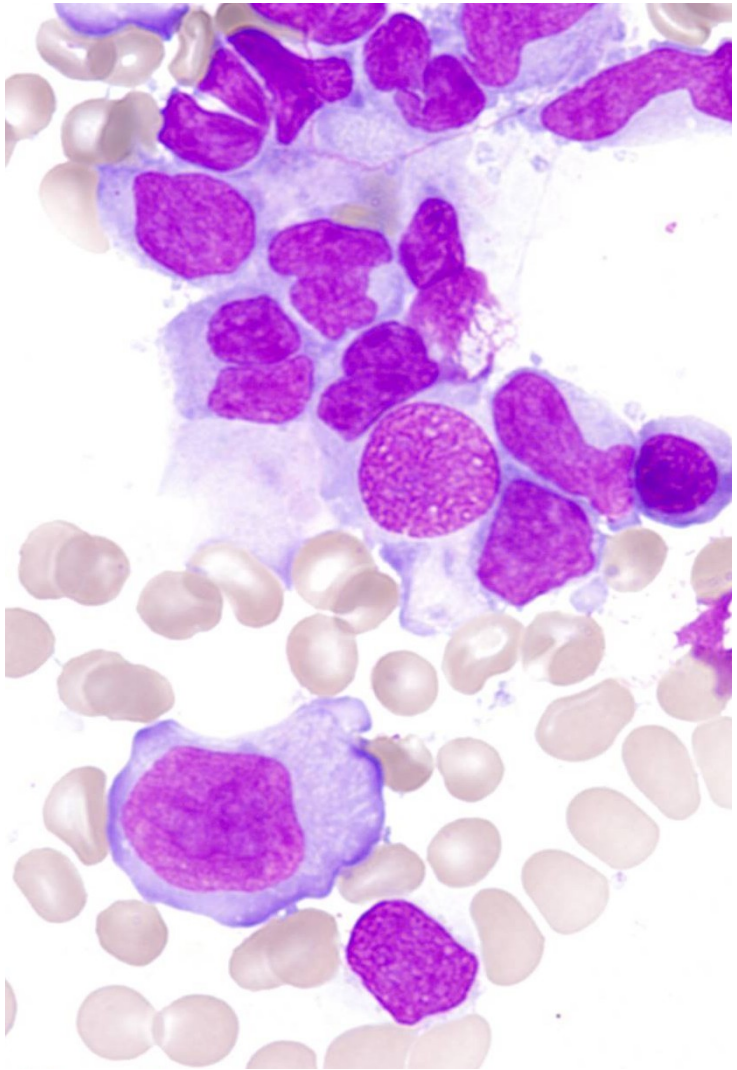
- Rapid local growth
- Large tumor mass
- Continuous generalization
- Frequent involvement of the central nervous system and bones

DLBCL – different morphological forms

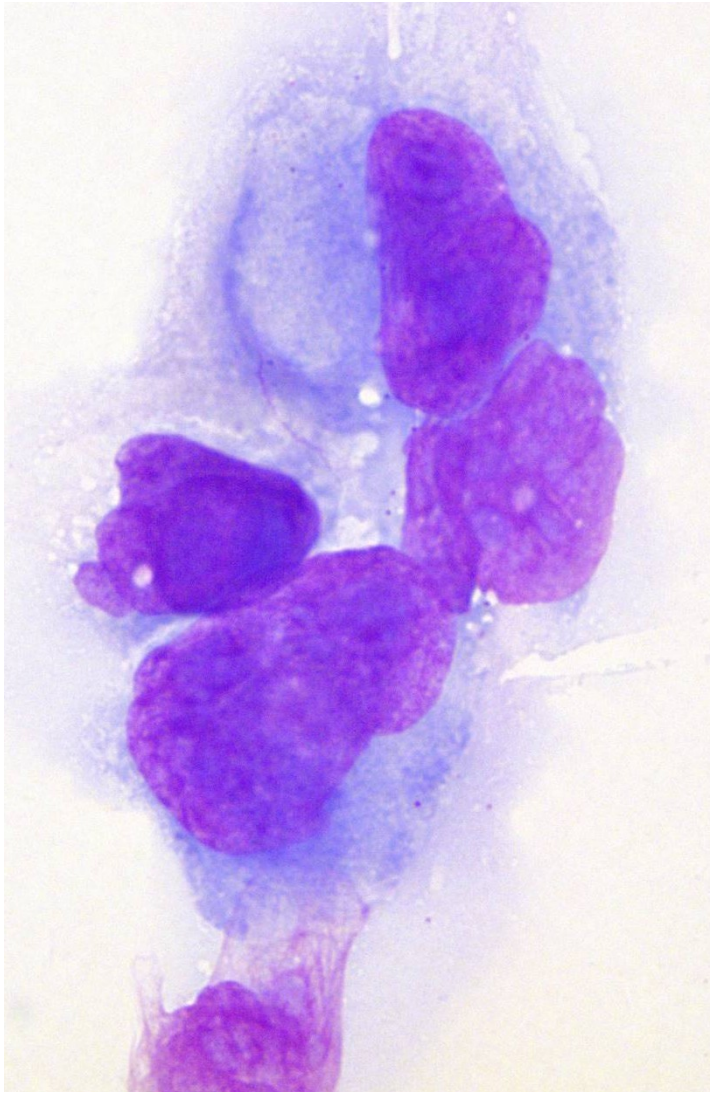
centroblastic



immunoblastic



anaplastic



DLBCL risk factors

- Age over 60 years
- Reduced physical fitness, ECOG higher than 1
- LDH level over upper limit of the norm
- Clinical stage higher than 2
- Extranodal involvement in more than 1 site

DLBCL therapy

First-line therapy

- anti-CD20 antibody + chemotherapy (R-CHOP regimen…)

Therapy of relapse

- Chemoimmunotherapy with anti-CD20 antibody
- High-dose therapy and autologous bone marrow transplant
- Allogeneic bone marrox transplant
- CAR T-cells

Very aggresive NHL

- **Lymphoblastic lymphoma**
  – acute lemphoblastic leukemia based protocols

- **Burkitt lymphoma**
  – agressive therapeutic regimens

HODGKIN DISEASE

- Lymphadenopathy with or without systemic symptoms fever, weigt loss, pruritus
- Pathologic Hodgkin or RS cells
- Two peakc of incidence: young adults and elederly

HODGKIN DISEASE

- Good risk goroup:
  Radiotherapy IF + 2 – 4 cycles of ABVD chemotherapy

- Intermediate risk group:
  BEACOP chemotherapy

- Poor prognosis:
  BEACOP
  Nivolumab
  Brentuximab vedotin (anti CD30)
  Autologous/allogeneic hematopoietic cell transplantation

HODGKIN DISEASE

Prognosis

- CR rate 95 %
- Progression free survival 90 % at 3 years

Multiple myeloma

MM

Proliferation of clonal malignant plasma cells in bone marrow

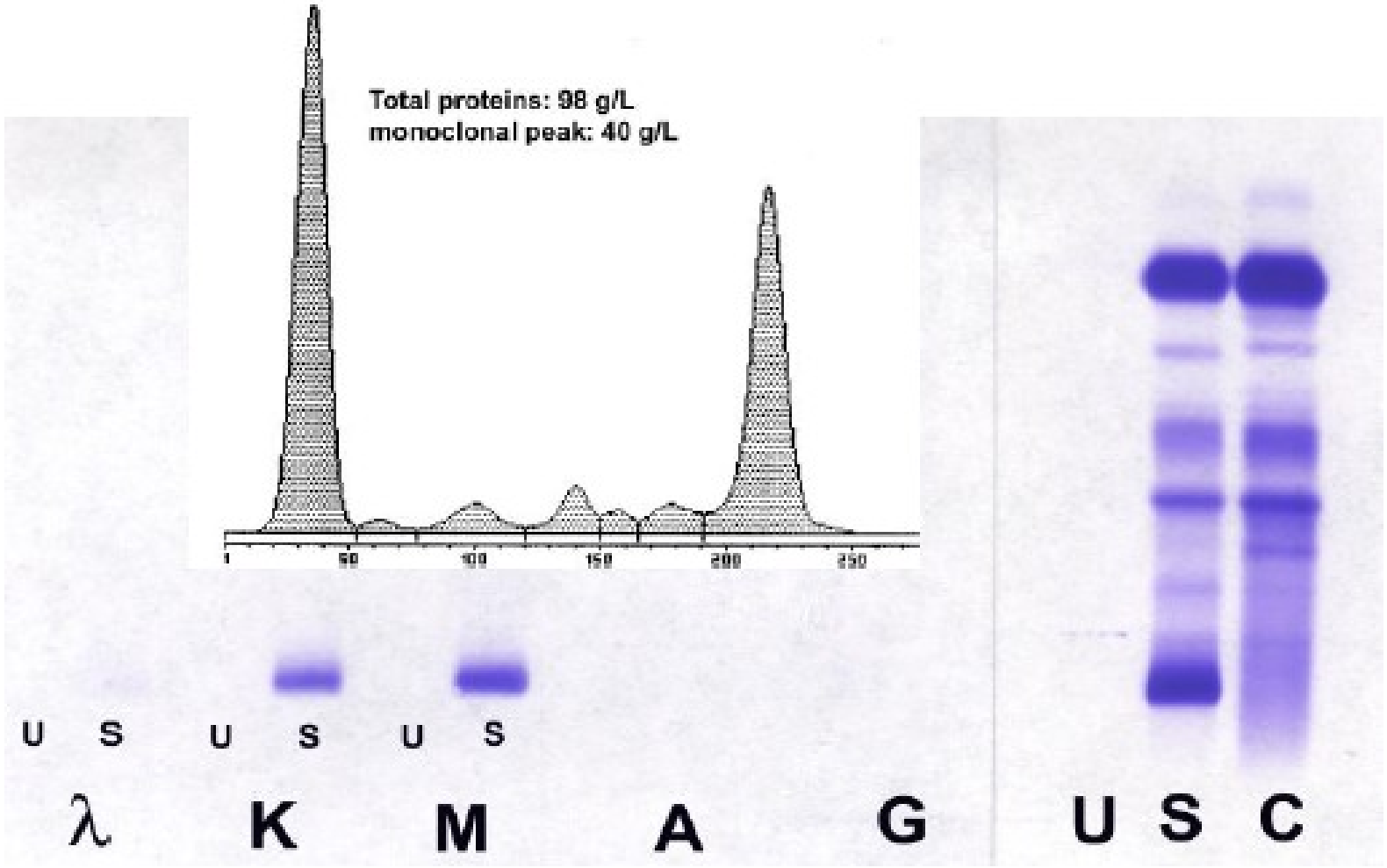
Complete monoclonal immunoglobulin molecule and/or kappa or lambda monoclonal free light chains produced by plasma cells

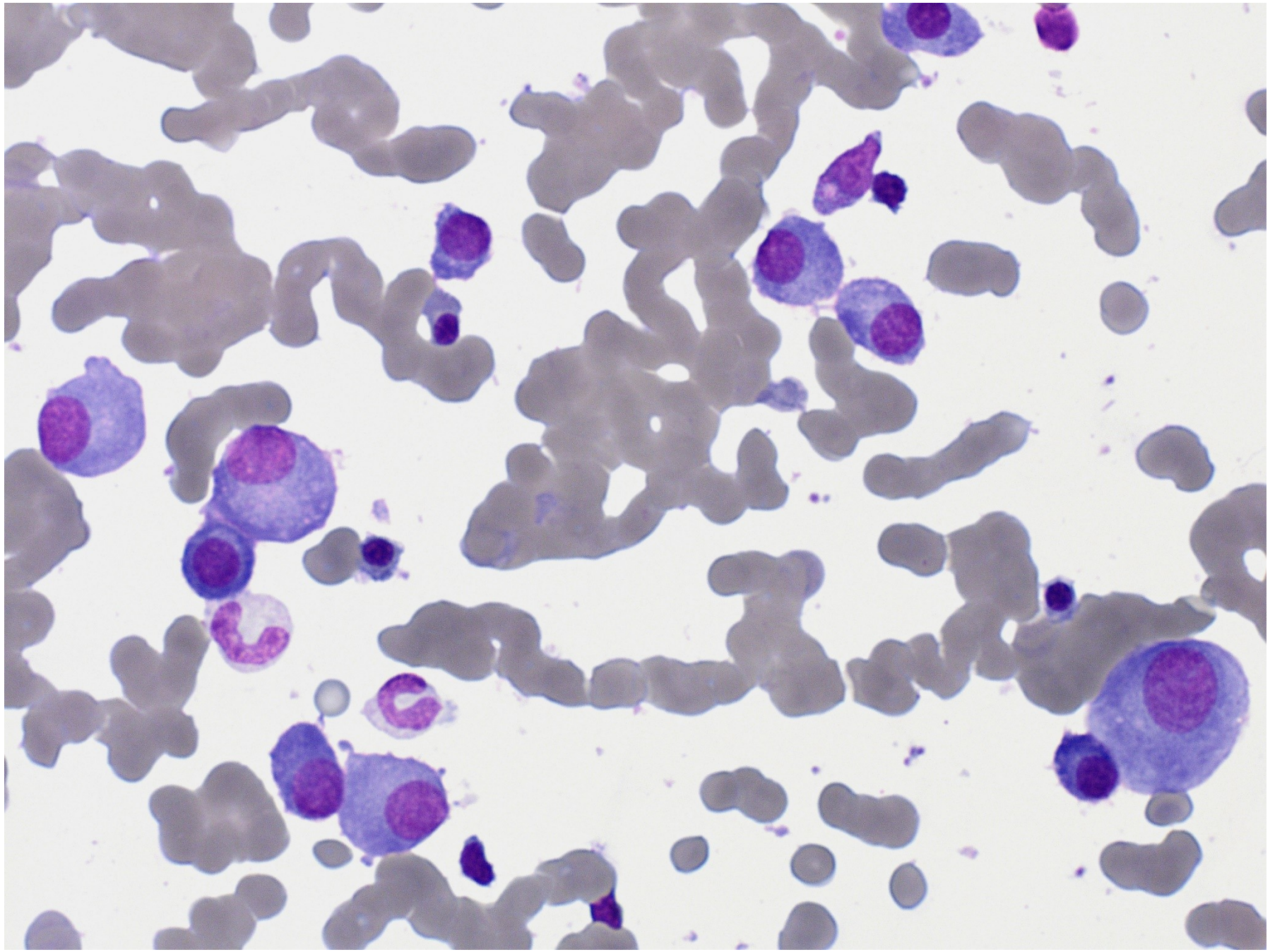
These changes lead to:

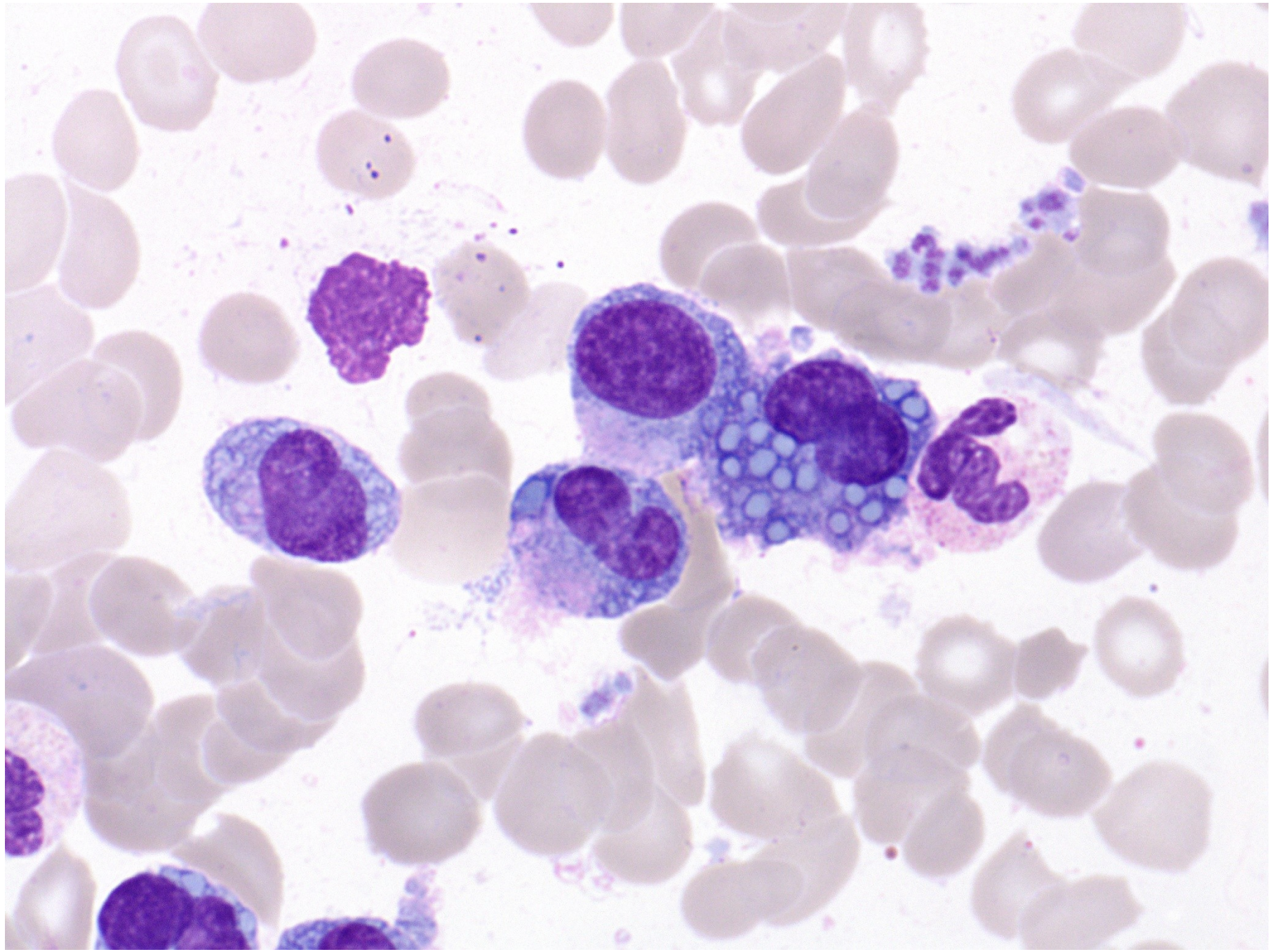
Osteolysis, osteoporosis, bone pain
Hypercalcemia
Hyperproteinemia
Renal failure
Coagulopathy
Neuropathy
Cytopenia

Incidence 4 / 100 000

MM – immunofixation, electrophoresis, densitometry







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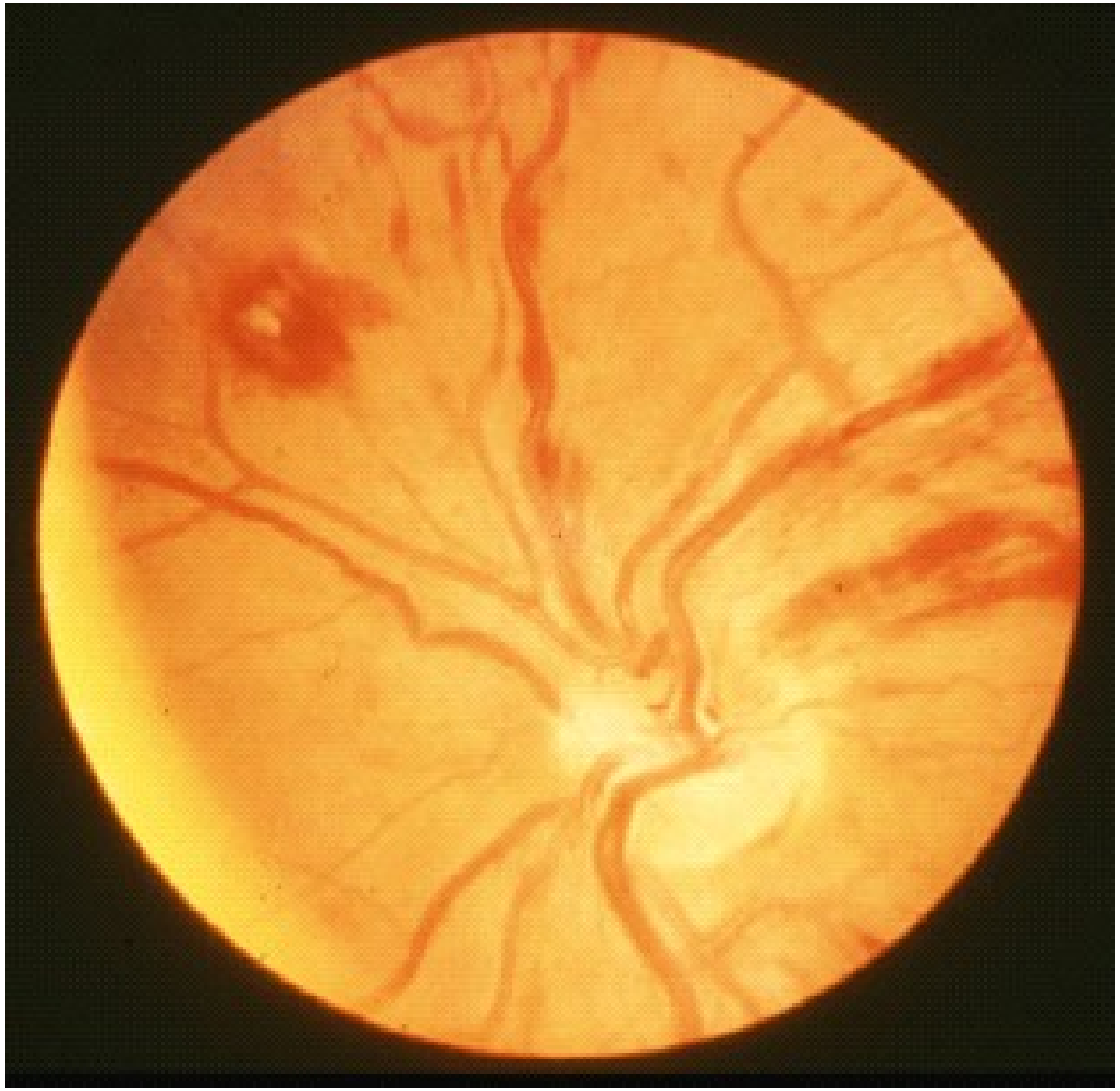

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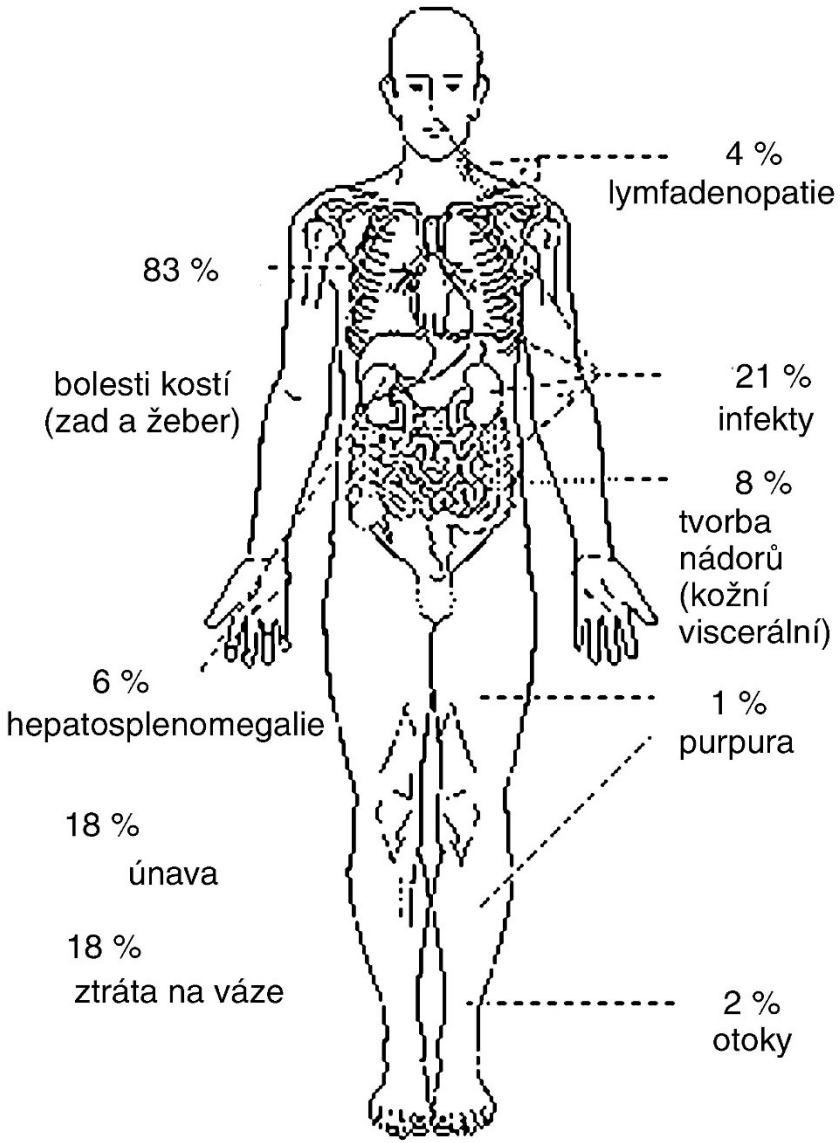
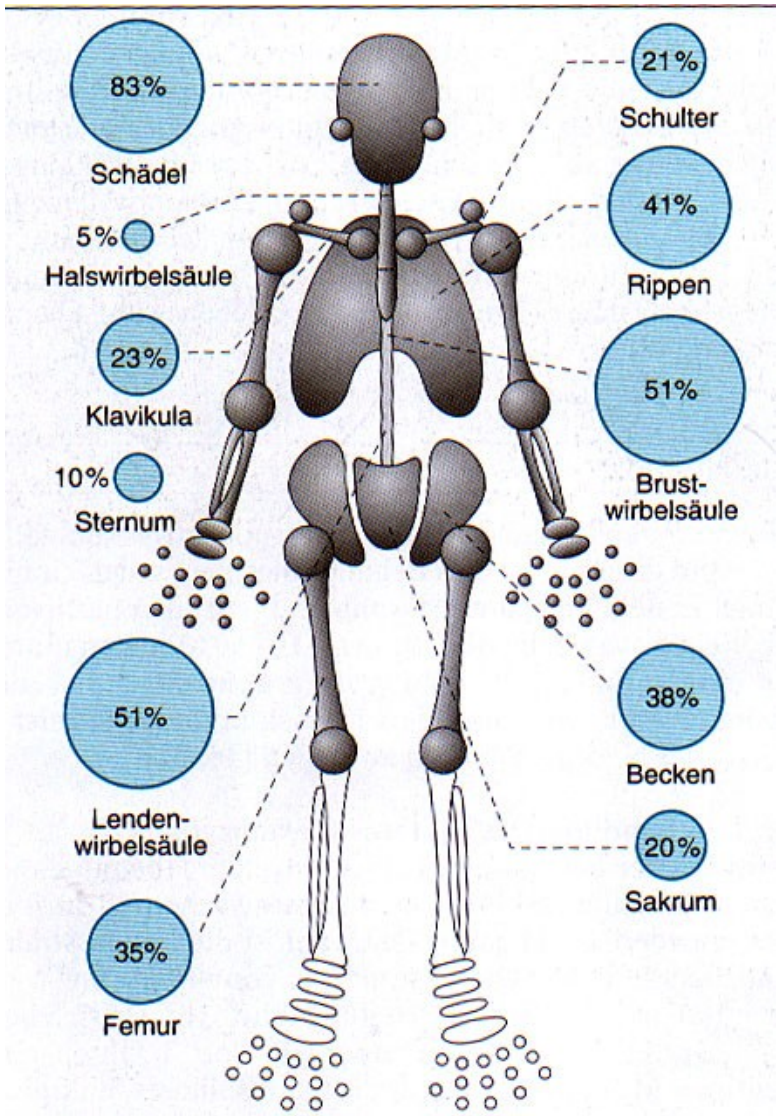
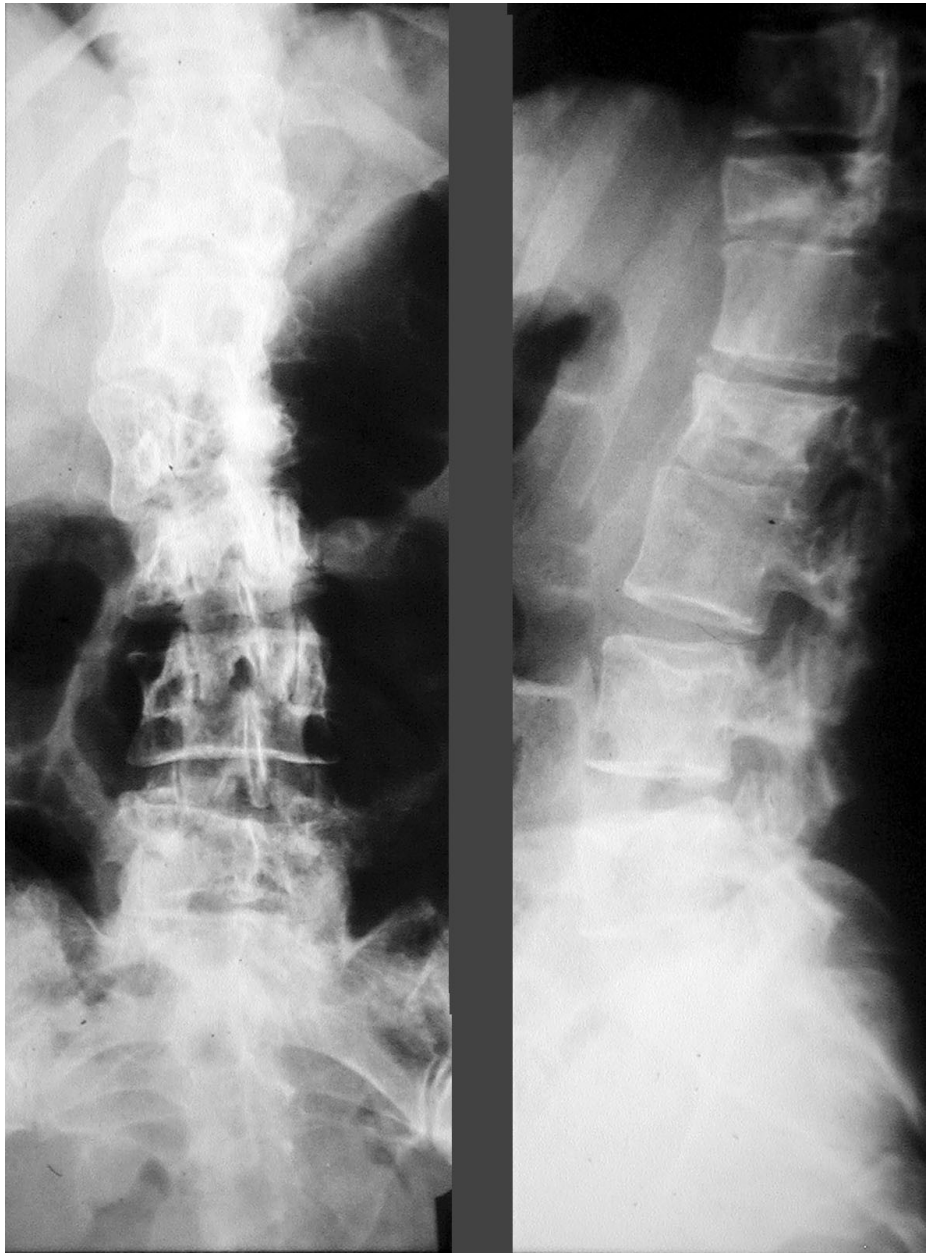
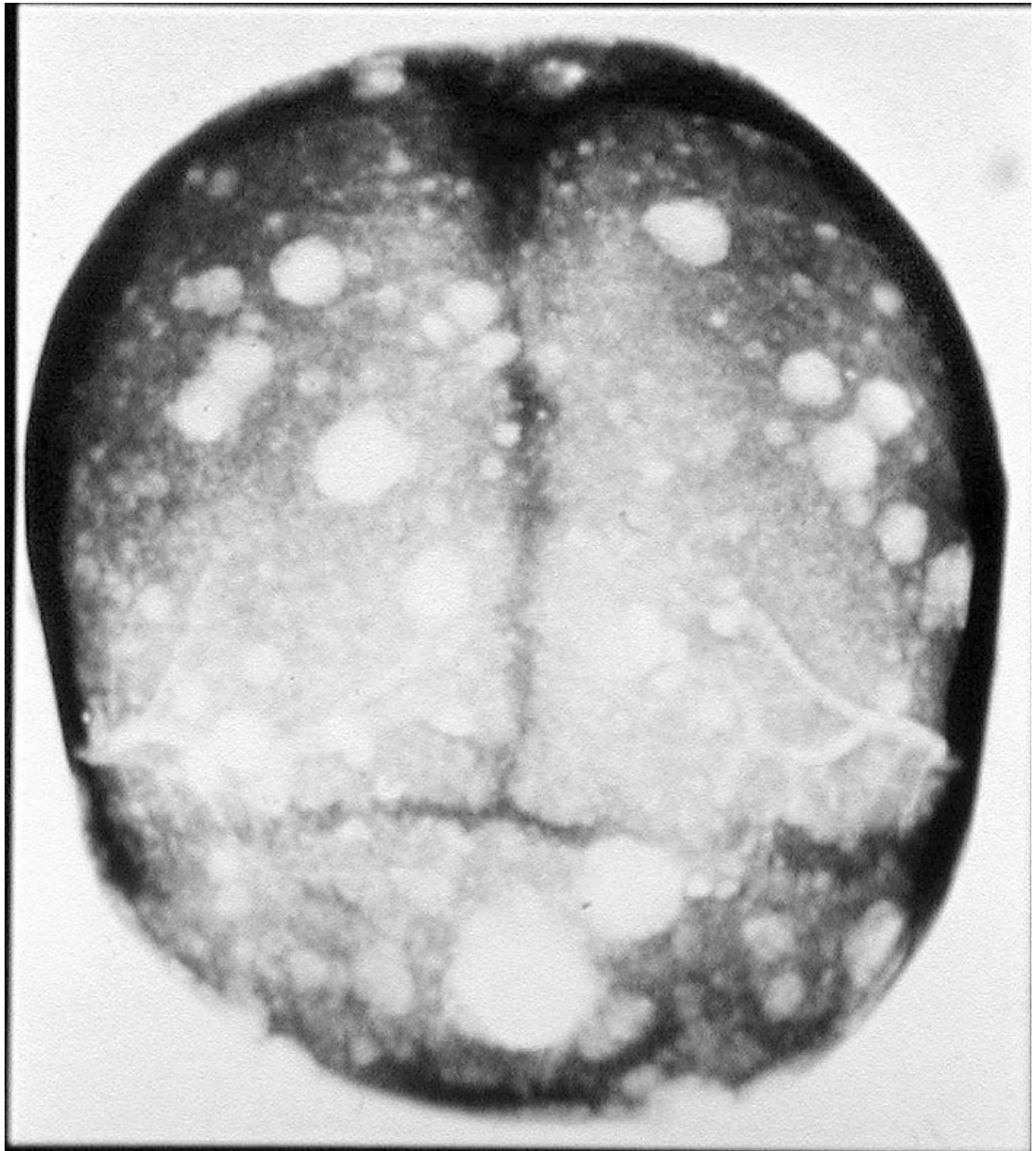

4 %
lymfadenopatie
83 %
bolesti kostí
(zad a žeber)
21 %
infekty
8 %
tvorba
nádorů
(kožní
viscerální)
6 %
hepatosplenomegalie
1 %
purpura
18 %
únava
18 %
ztráta na váze
2 %
otoky


83%
Schädel
5%
Halswirbelsäule
23%
Klavikula
10%
Sternum
51%
Lenden-
wirbelsäule
35%
Femur
21%
Schulter
41%
Rippen
51%
Brust-
wirbelsäule
38%
Becken
20%
Sakrum

MM





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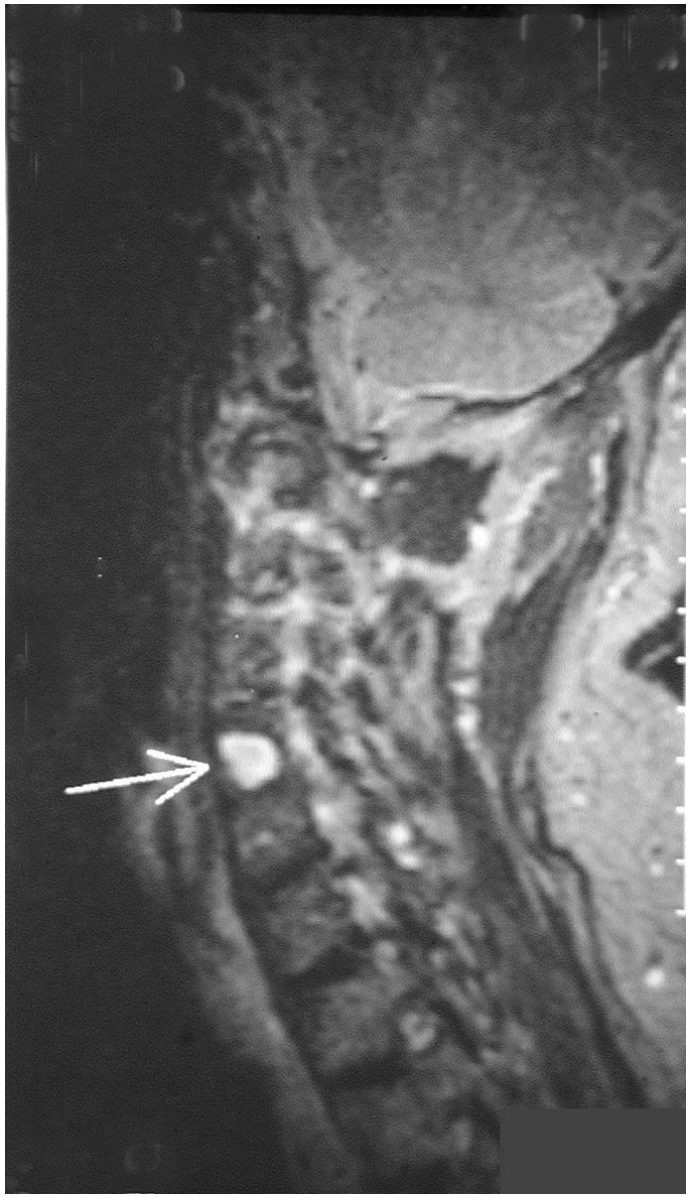

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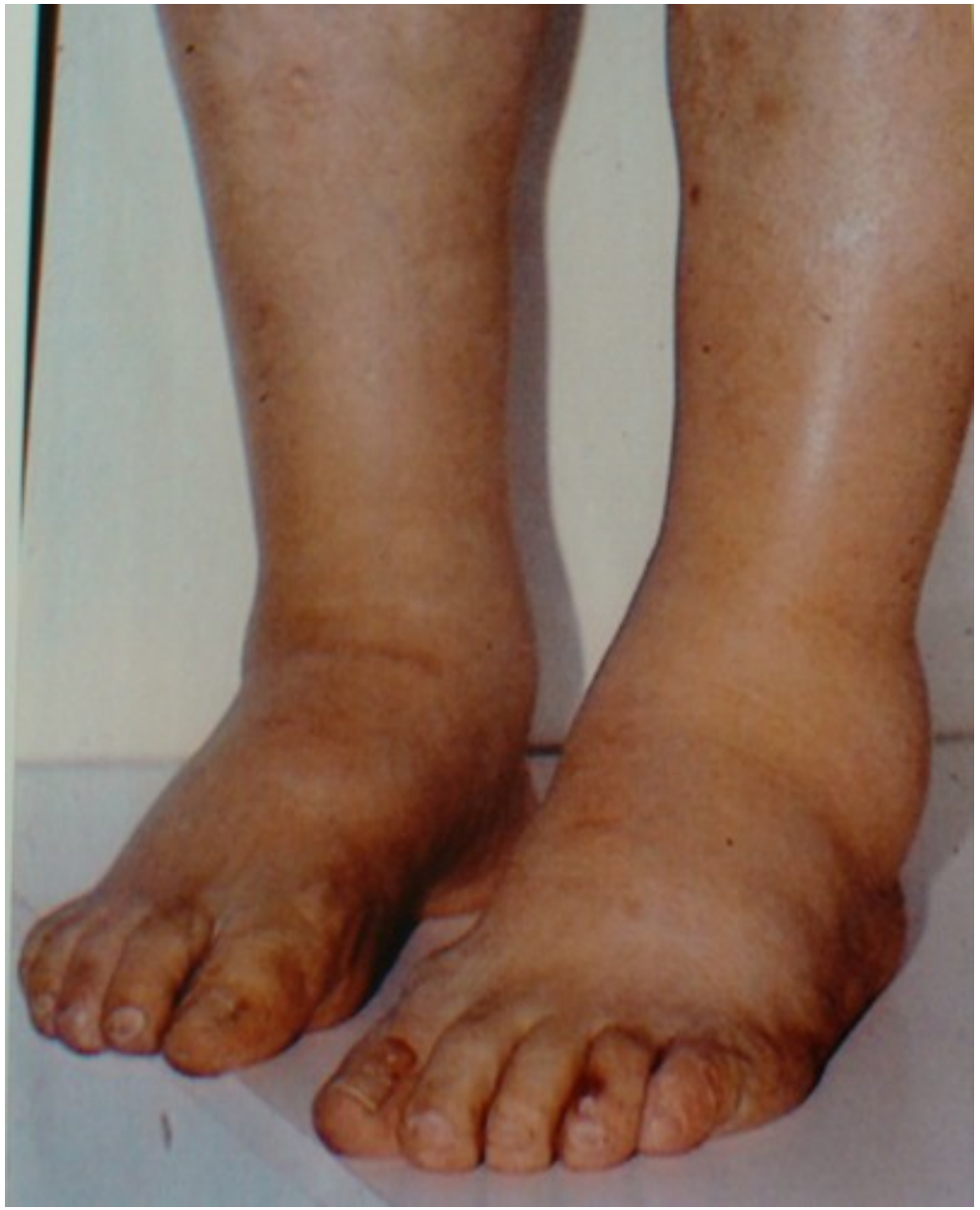


MM

Diagnostics:

Monoclonal immunoglobulin (or light chains) in peripheral blood and urine

Bone marrow histology/cytology

Imaging methods: X-ray, MR, PET/CT

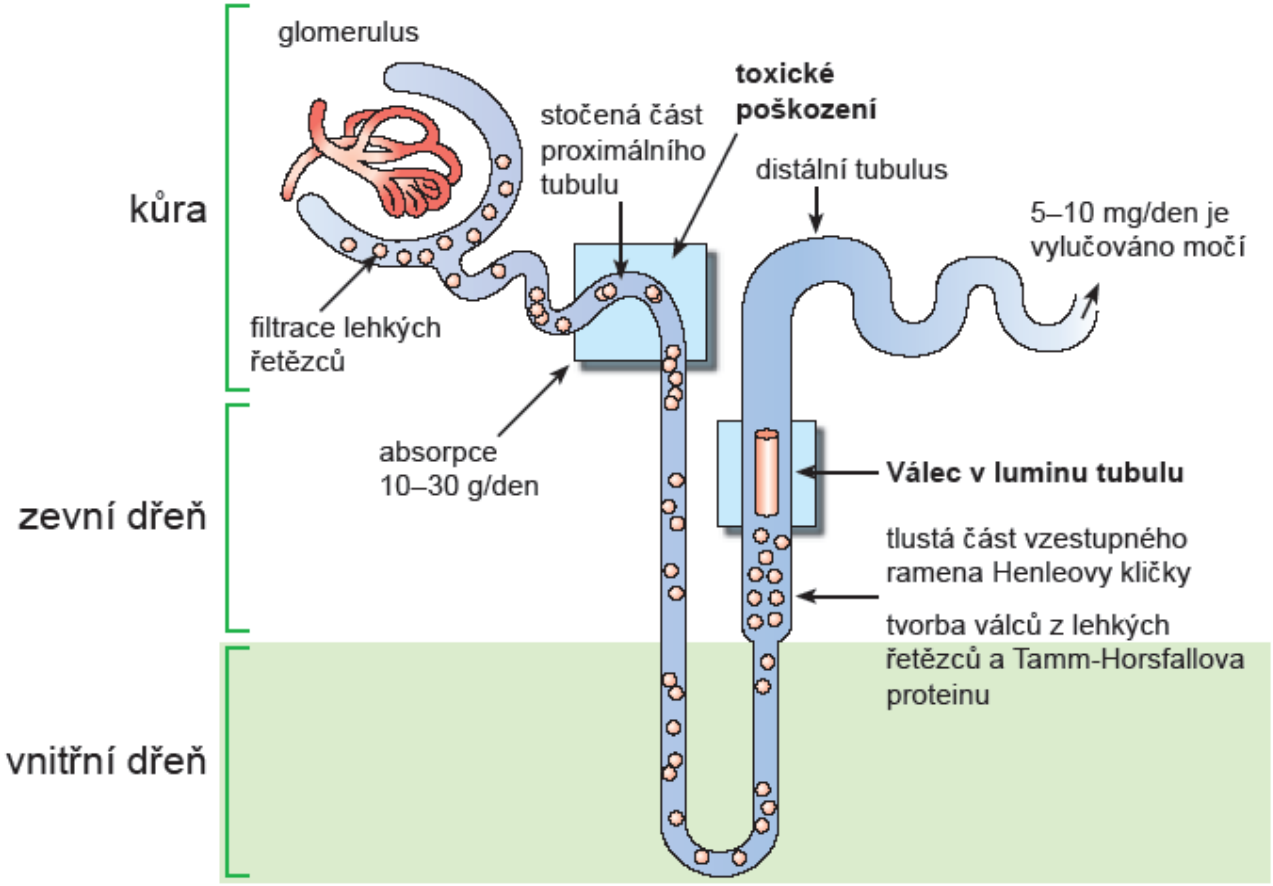
Serum immunoglobulins

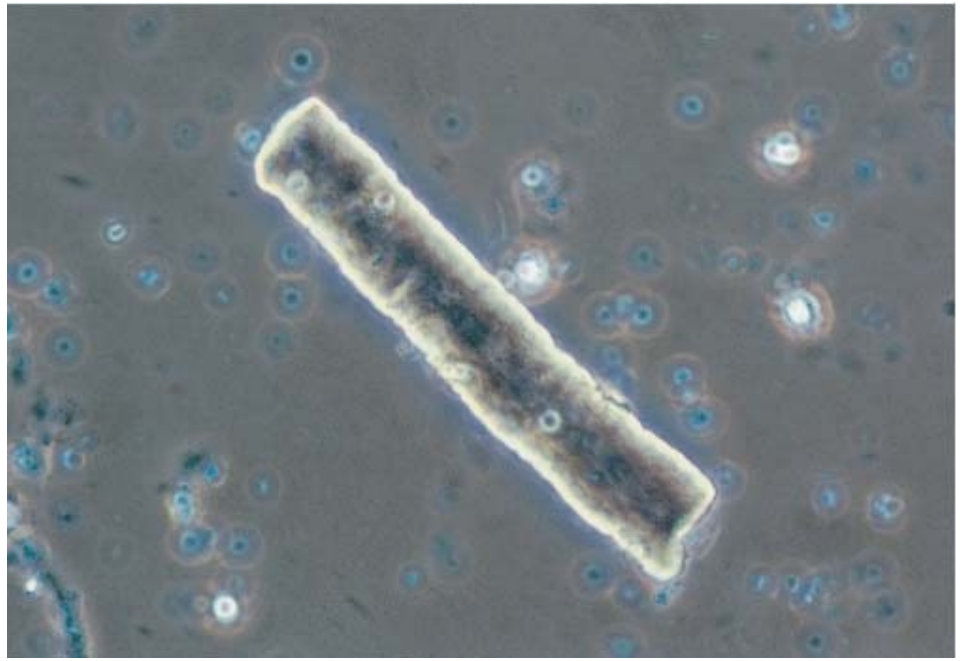
Serum calcium level

Serum protein

Peripheral blood count

MM – kidney failure





MM - therapy

Indication for therapy:
Symptomatic patients: cytopenia, bone leasions, hypercalcemia, kidney failure…

Drugs:
Chemotherapy (vincristine, melfalan)
Corticosteroids (dexamethasone)
Proteasome inhibitors (bortezomib, ixazomib, carfilzomib)
IMIDs (lenalidomide, pomalidomide)
Anti-CD38 (daratumumab)

High-dose therapy + autologous hematopoietic stem cell transplantation

MM – supportive care

- Bisfosfonates
- Dialysis
- Plasmaferesis
- Radiotherapy
- Pain killers
- Prophylaxis of infection
- Tranfusions

positive mutation