Bolesti matične stanice hematopoeze

Petar Gaćina
Klinika za unutarnje bolesti
KBC Sestre milosrdnice

Hematopoetski sustav

Organi i tkiva

- Koštana srž
- Timus
- Slezena
- Limfni čvorovi
- Mukozno limfocitno tkivo

Stanice

- MIJELOPOETSKE STANICE
  Eritrociti
  Granulociti (neutrofilni, eozinofilni,bazofilni)
  Monociti/makrofagi
  Trombociti
  LIMFOPOETSKE STANICE
  (limfociti B i T)

Mjesto stvaranja krvotvornog tkiva

- Fetus 0-2 mjeseca žumančana vreća
  2-7 mjeseci jetra, slezena
  5-9 mjeseci koštana srž
- Djeca koštana srž (sve kosti)
- Odrasli kralješci,sternum,kosti lubanje, sakrum, zdjelica, proksimalni dio femura

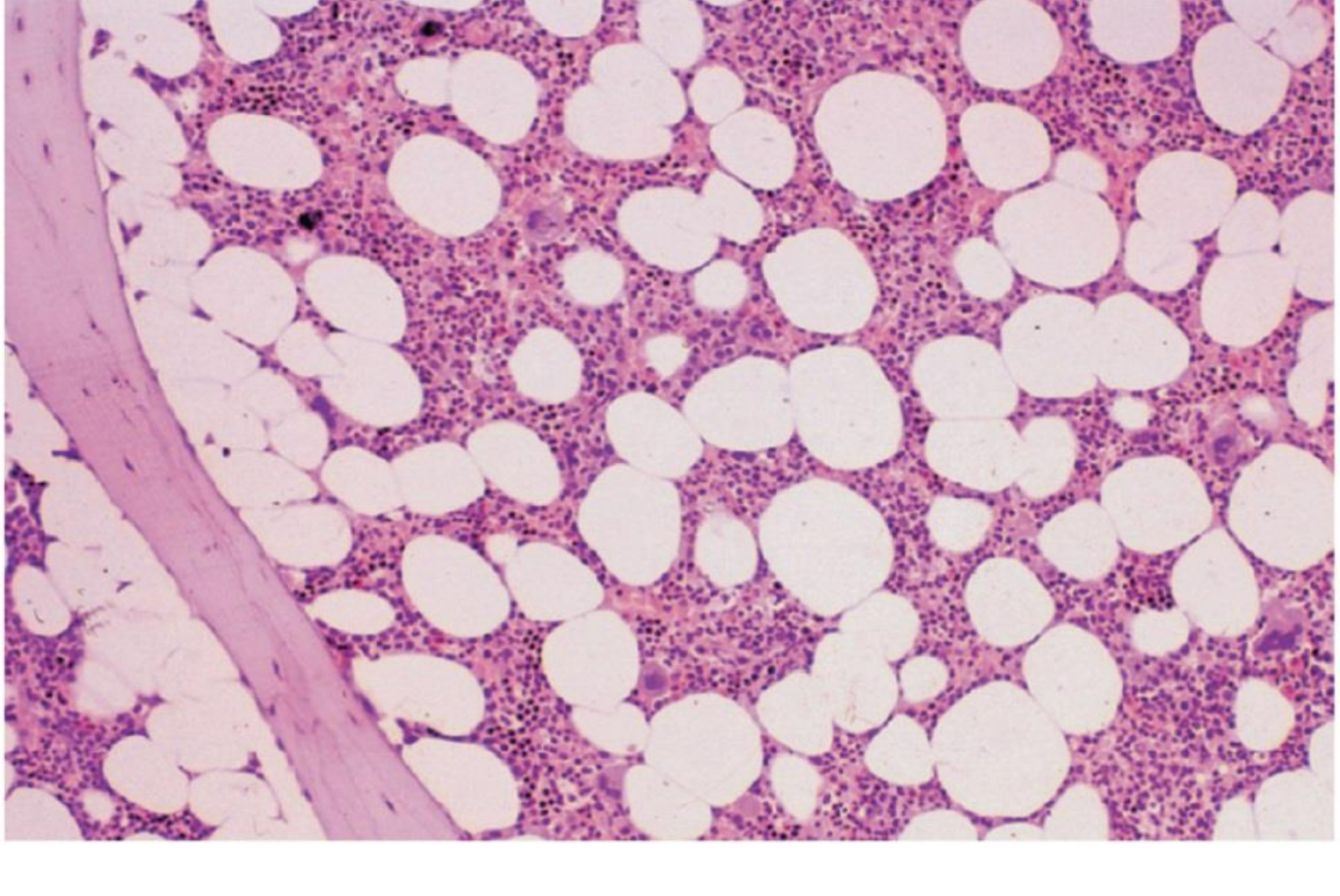

Figure 1.1 Normal bone marrow trephine biopsy (posterior iliac crest). Haematoxylin and eosin stain; approximately 50% of the intertrabecular tissue is haemopoietic tissue and 50% is fat.

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MATIČNA STANICA

- Sposobnost samoobnavljanja
- Diferencijacija i proliferacija ovisno o potrebi
- Izrazita proliferacijska sposobnost
- Važnost mikrookoliša
- Koštana srž 1-4%, a u perifernoj krvi 0,03-0,08% matičnih stanica u odraslih
- Stanice posjeduje biljeg CD34

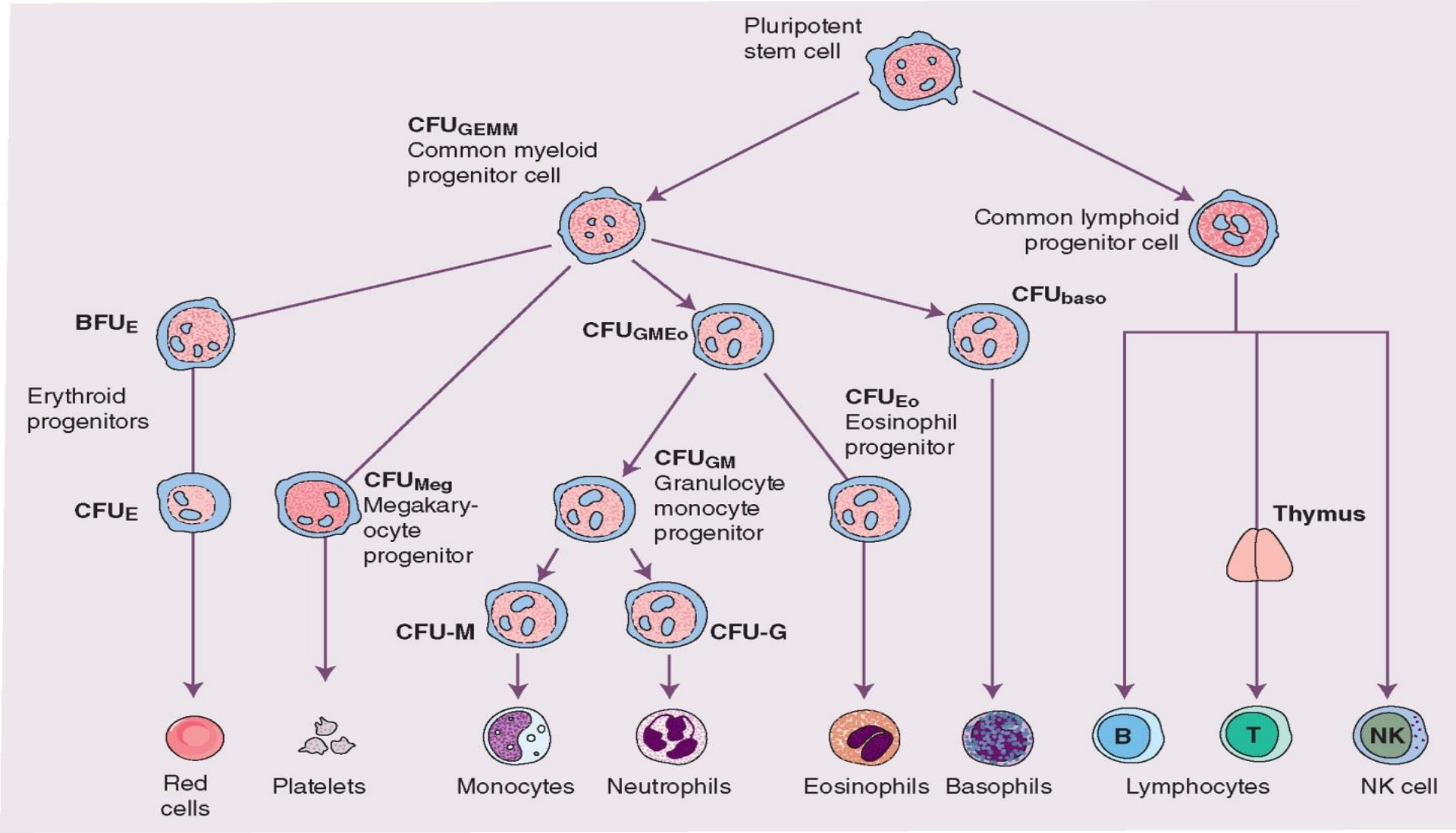

Figure 1.2 Diagrammatic representation of the bone marrow pluripotent stem cell and the cell lines that arise from it. Various progenito cells can be identified by culture in semi-solid medium by the type of colony they form. It is possible that an erythroid/megakaryocytic progenitor may be formed before the common lymphoid progenitor diverges from the mixed granulocytic/monocyte/eosinophil myeloid progenitor. Baso, basophil; BFU, burst-forming unit; CFU, colony-forming unit; E, erythroid; Eo, eosinophil; GEMM, granulocyte, erythro monocyte and megakaryocyte; GM, granulocyte, monocyte; Meg, megakaryocyte; NK, natural killer.

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HEMATOPOETSKI FAKTORI RASTA

- Djeluju na stanice strome (IL-1, M-CSF, IL-6)
- Djeluju na pluripotentnu matičnu stanicu (SCF-faktor matičnih stanica)
- Djeluju na multipotentne matične stanice (IL-3, IL-6, GM-CSF)
- Djeluju na usmjerene matične stanice (G-CSF, M-CSF, IL-5, eritropoetin i trombopoetin)

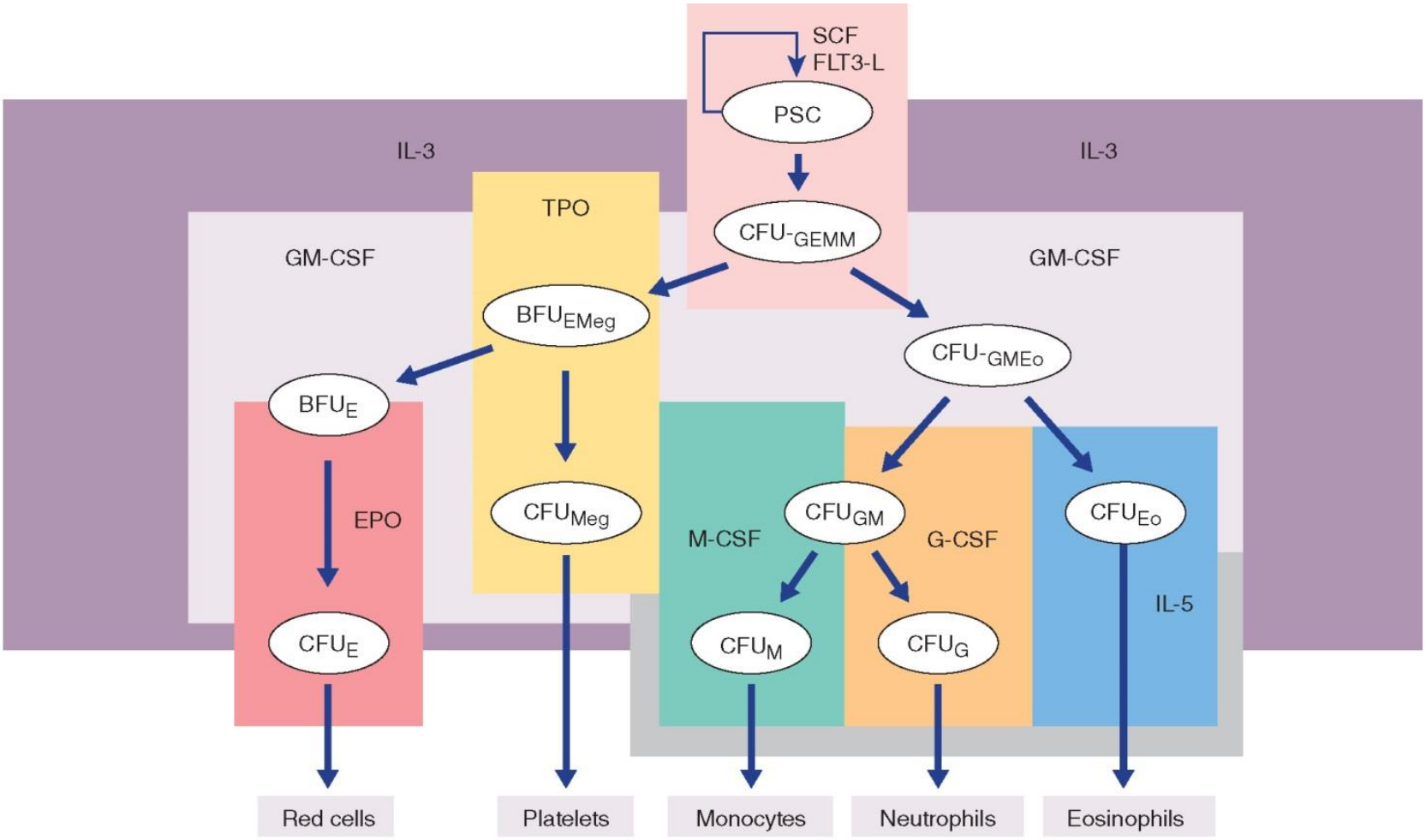

Figure 1.6 A diagram of the role of growth factors in normal haemopoiesis. Multiple growth factors act on the earlier marrow stem and progenitor cells. EPO, erythropoietin; PSC, pluripotential stem cell; SCF, stem cell factor; TPO, thrombopoietin; FLT3-L, FLT3 ligand. For other abbreviations see Fig. 1.2.

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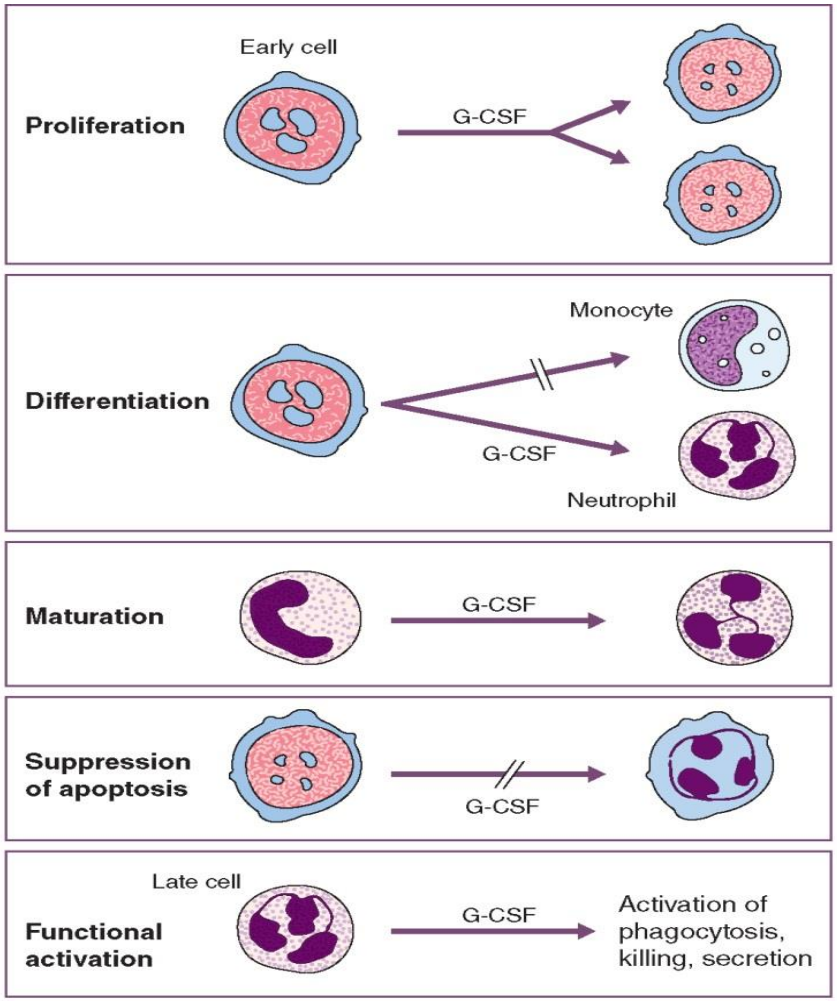

Figure 1.5 Growth factors may stimulate proliferation of early bone marrow cells, direct differentiation to one or other cell type, stimulate cell maturation, suppress apoptosis or affect the function of mature non-dividing cells, as illustrated here for granulocyte colony-stimulating factor (G-CSF) for an early myeloid progenitor and a neutrophil.

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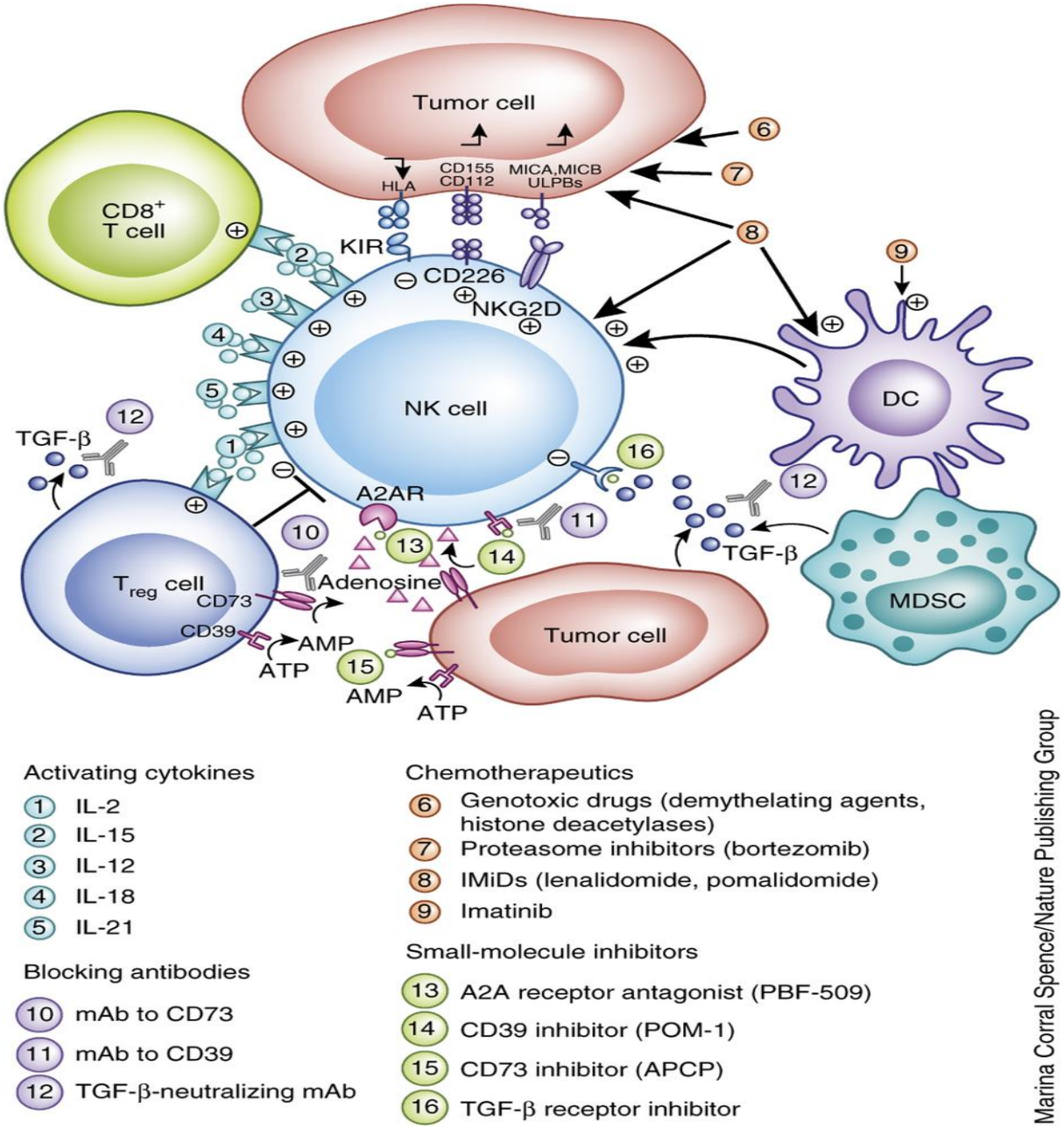
MIKROOKOLIŠ



INHIBITORI DIFERENCIJACIJE I RASTA MATIČNIH STANICA

- TGF-beta
- TNF
- IL-4
- Zračenje
- Lijekovi
- Infiltracija koštane srži stranim stanicama i vezivnim tkivom

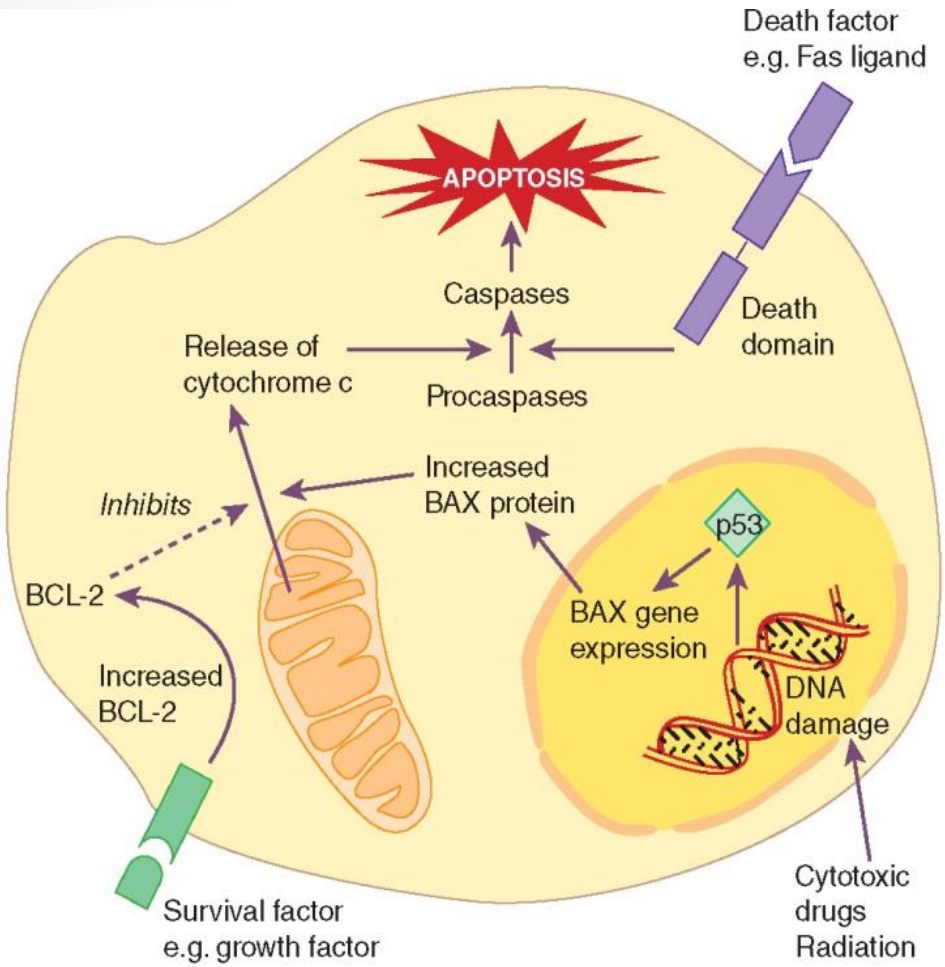


Figure 1.9 Representation of apoptosis. Apoptosis is initiated via two main stimuli: (i) signalling through cell membrane receptors such as FAS or tumour necrosis factor (TNF) receptor; or (ii) release of cytochrome c from mitochondria. Membrane receptors signal apoptosis through an intracellular death domain leading to activation of caspases which digest DNA. Cytochrome c binds to the cytoplasmic protein Apaf-1 leading to activation of caspases. The intracellular ratio of pro-apoptotic (e.g. BAX) or anti-apoptotic (e.g. BCL-2) members of the BCL-2 family may influence mitochondrial cytochrome c release. Growth factors raise the level of BCL-2 inhibiting cytochrome c release, whereas DNA damage, by activating p53, raises the level of BAX which enhances cytochrome c release.

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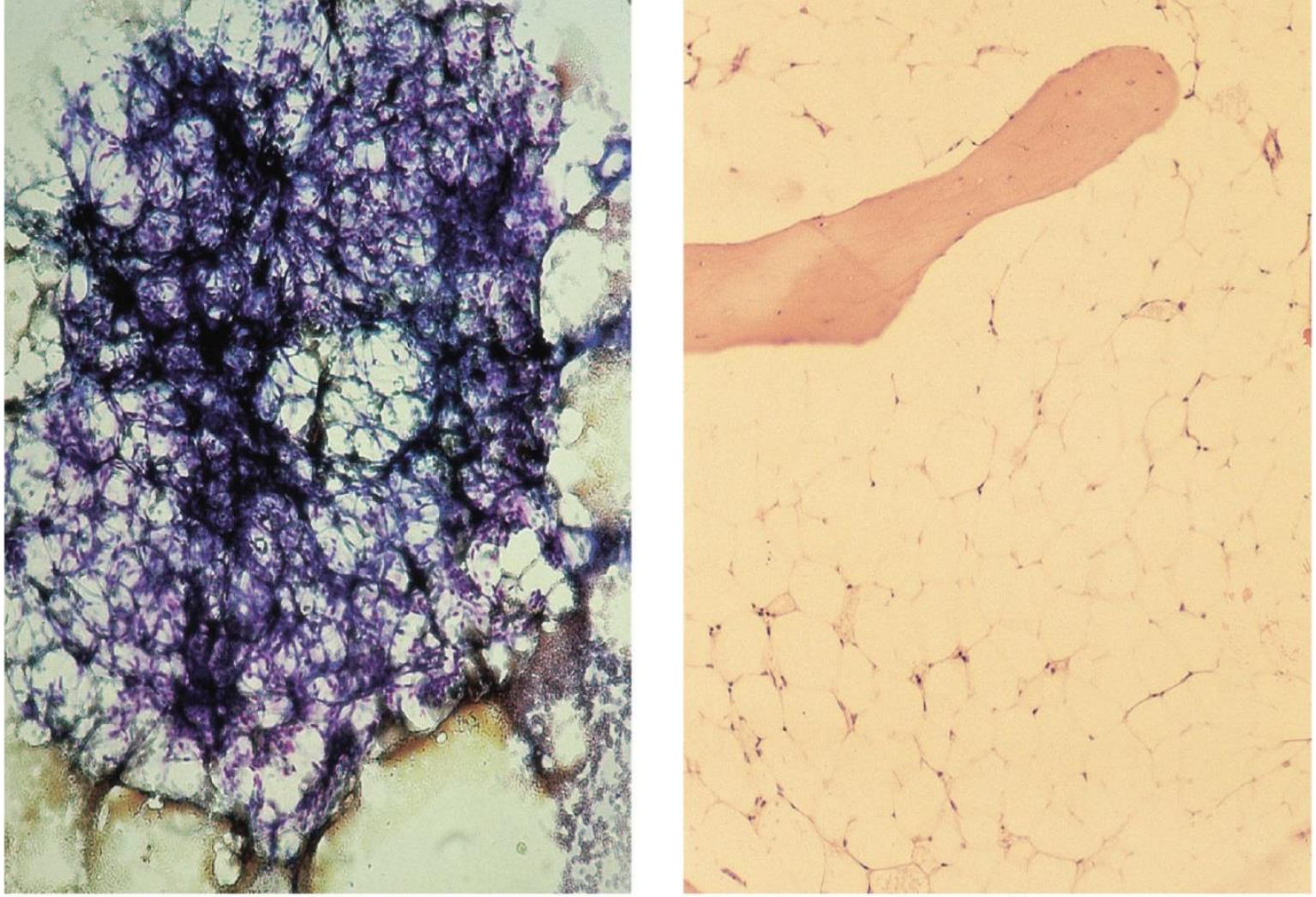
Bolesti matične hematopoetske stanice

- Mijeloaplazije - slabost ili manjak matičnih hematopoetskih stanica
- Mijelodisplazije - klonalne bolesti matičnih hematopoetskih stanica
- Mijelo i limfoproliferacijske bolesti- zloćudna transformacija genoma matičnih hematopoetskih stanica (akutna mijeloična i limfatička leukemija i kronične mijeloproliferativne i limfoproliferativne bolesti)

Etiološka podjela aplastičnih anemija

- PRIMARNE
- kongenitalna (Fanconi anemija)
- idiopatska stečena

- SEKUNDARNE
- ionizantno-zračenje
- benzen i druga organska otapala
- lijekovi
- infekcije
- metabolički
- imunološki
- paroksizmalna noćna hemoglobinurija



(a) (b)

Figure 22.1 Aplastic anaemia: low power views of bone marrow show severe reduction of haemopoietic cells with an increase in fat spaces. **(a)** Aspirated fragment. **(b)** Trephine biopsy.

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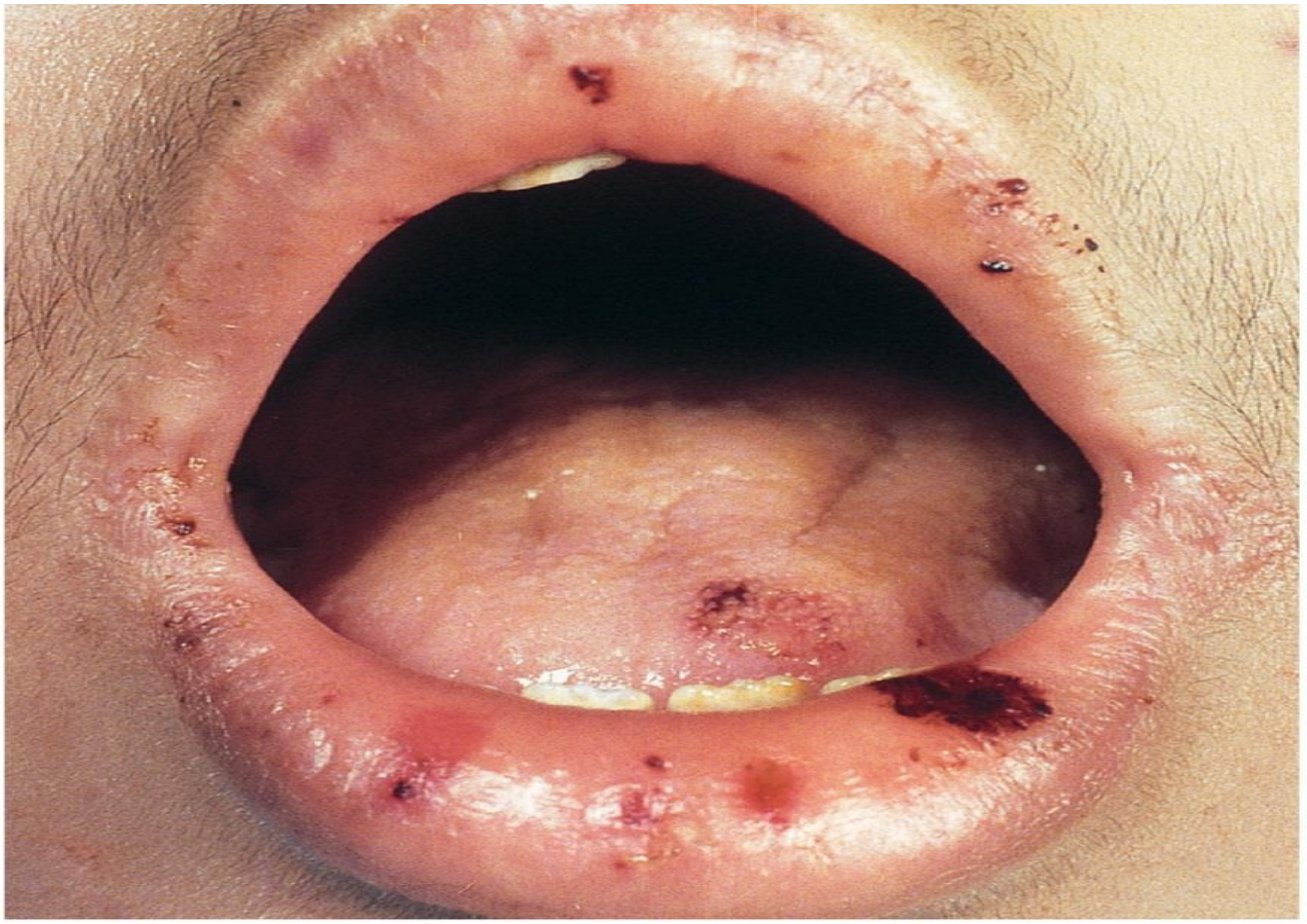

Figure 22.4 Aplastic anaemia: spontaneous mucosal haemorrhages in a 10-year-old boy with severe Fanconi anaemia. Platelet count $<5 \times 10^9$/L. Source: Hoffbrand A.V., Pettit J.E. & Vyas P. (2010) *Color Atlas of Clinical Hematology*, 4th edn. Reproduced with permission of John Wiley & Sons.

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PODJELA MIJELODISPLAZIJA
(WHO klasifikacija)

- MDS s jednolinijskom displazijom (<5%)
- MDS s prstenastim sideroblastima (<5%)
- MDS s multilinijskom displazijom (70%)
- MDS s viškom blasta (MDS EB 1 i 2) (25%)
- MDS s delecijom 5q (5%)
- Neklasificirana MDS (<5%)

Leukemije

- Rak koštane srži
- Očituje se nakupinama određenih krvnih stanica koje čine tumorsku masu
- Nepoznat uzrok (genetski faktor, izloženost kemikalijama, zračenju, bakterijama i virusima)

Tipovi leukemija

- Akutna limfatična leukemija
- Akutna mijeloična leukemija
- Kronična limfocitna leukemija
- Kronična mijeloidna leukemija

Primarne mijeloproliferativne bolesti

- Akutne - akutna mijeloična leukemija

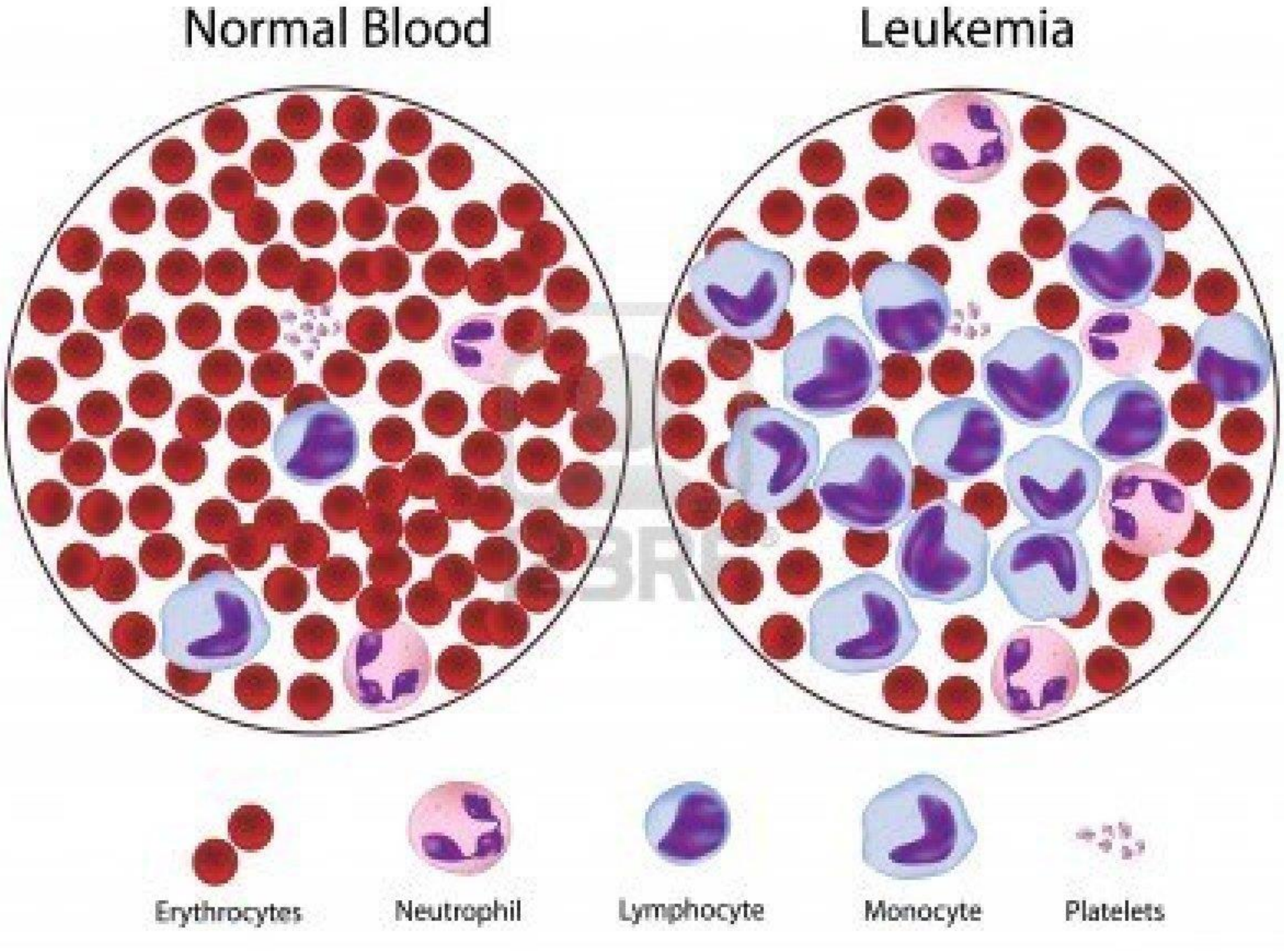
- Kronične - policitemija rubra vera
  esencijalna trombocitopenija
  kronična mijeloična leukemija
  idiopatska mijelofibroza

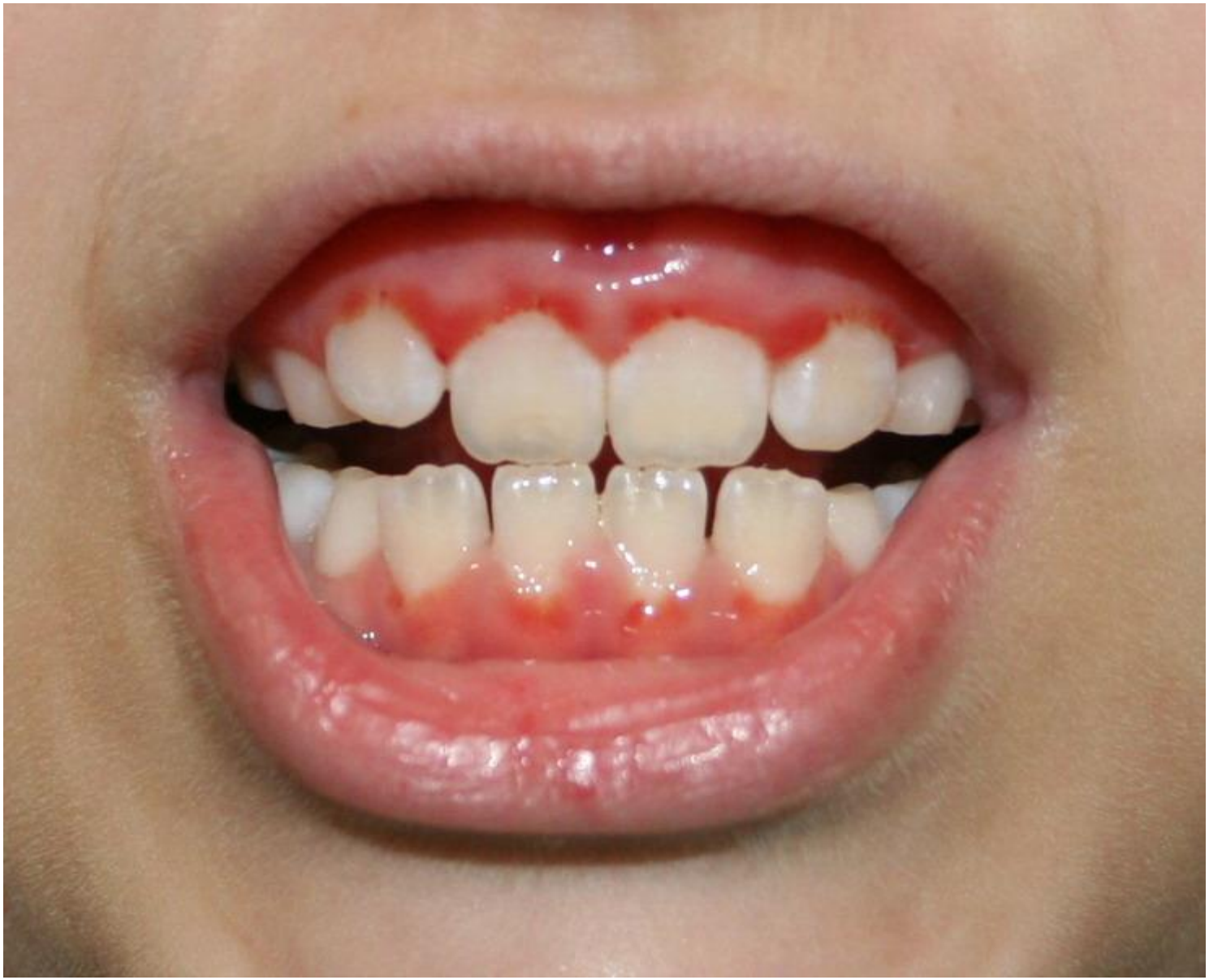
AKUTNE MIJELOIČNE LEUKEMIJE FAB KLASIFIKACIJA

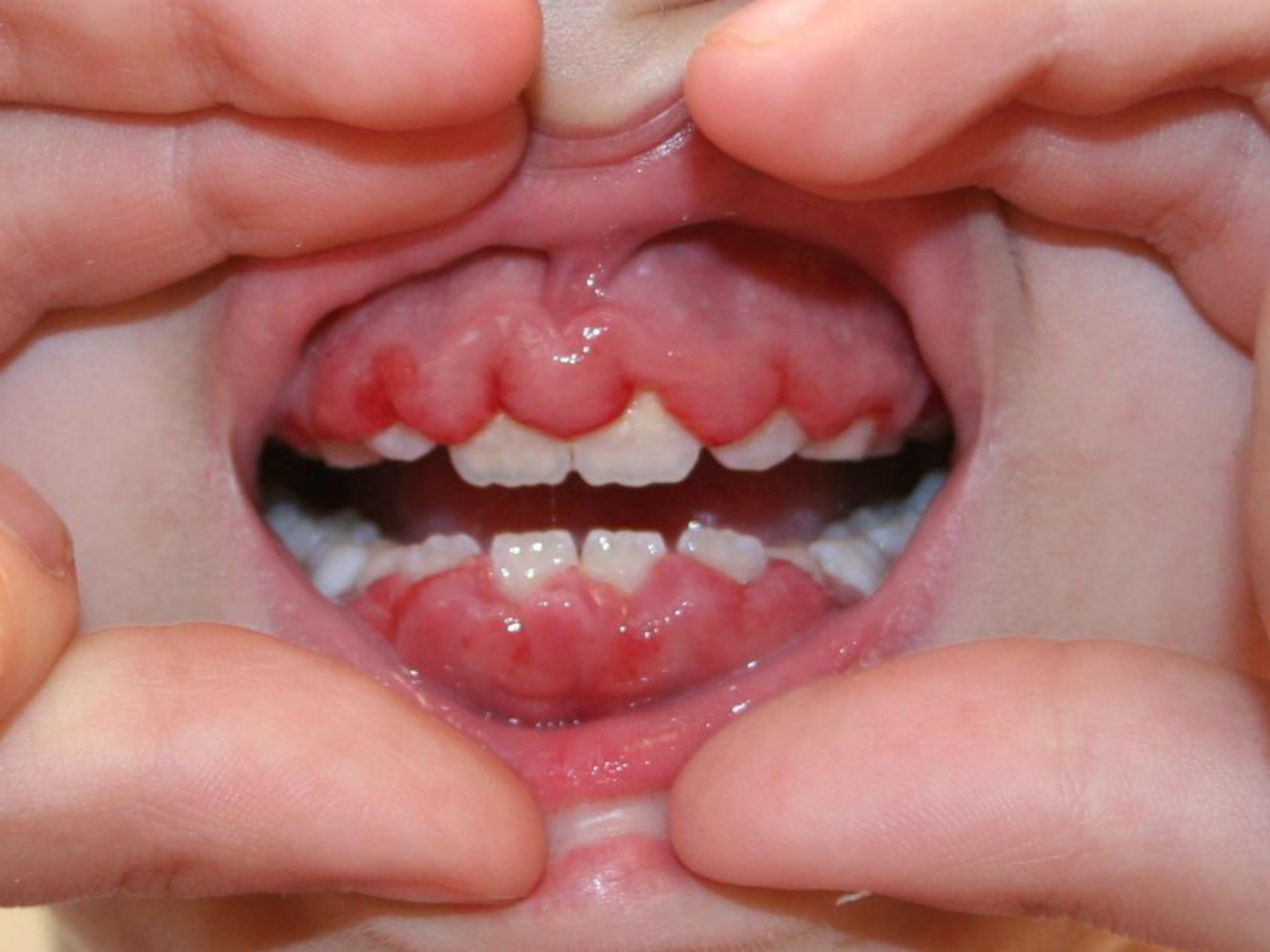
- M_0 – akutna mijeloblastična bez sazrijevanja
- M1 – akutna mijeloblastična
- M2 – akutna mijeloblastična sa sazrijevanjem
- M3 – akutna promijelocitna
- M4 – akutna mijelomonocitna
- M5 – akutna monocitna
- M6 – akutna eritroleukemija
- M7 – akutna megakariocitna

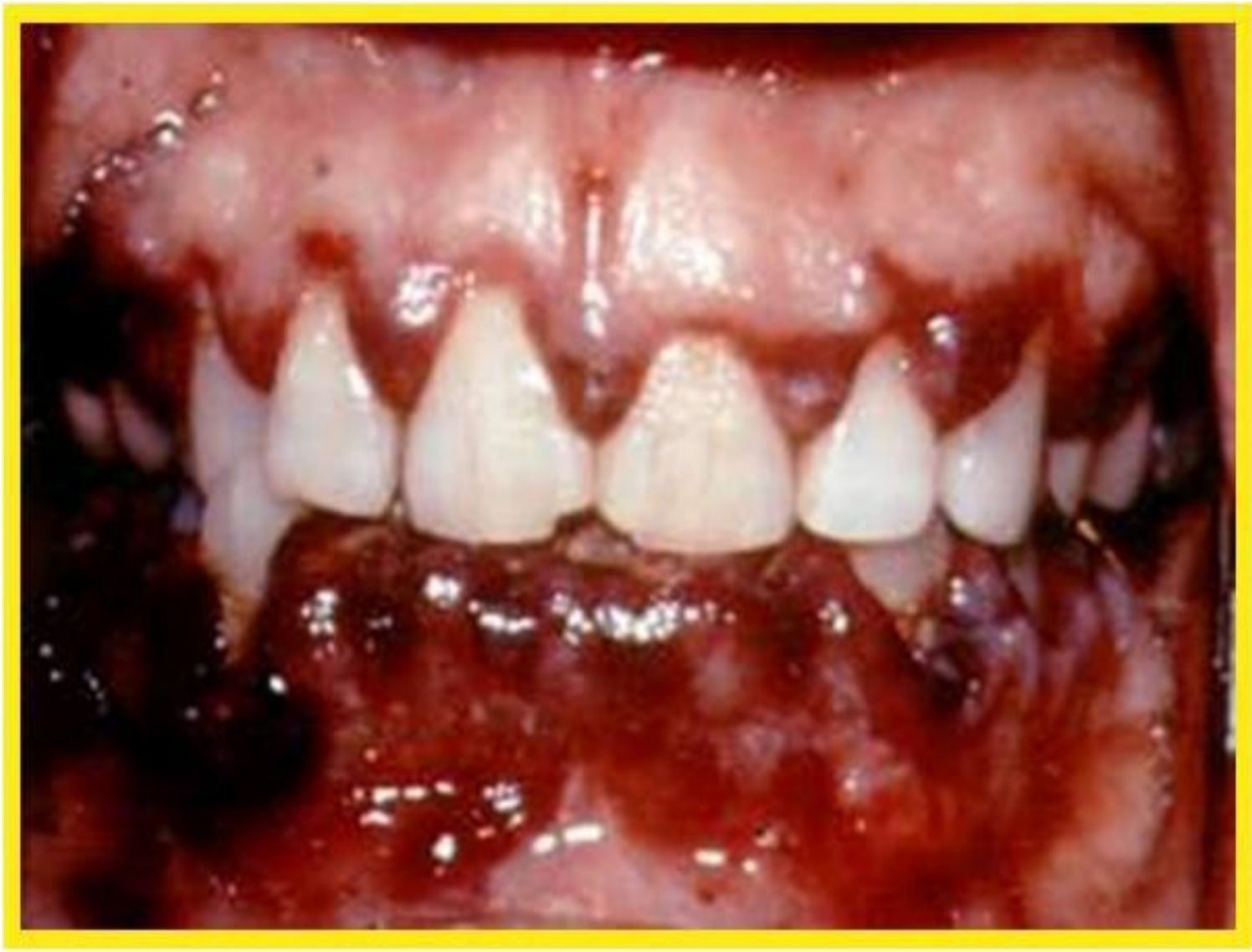
Simptomi leukemije

- Umor
- Gubitak apetita i tjelesne težine
- Povišena tjelesna temperatura
- Učestala znojenja
- Sklonost krvarenju i pojavljivanju modrica
- Bol u kostima


Normal Blood
Leukemia
Erythrocytes
Neutrophil
Lymphocyte
Monocyte
Platelets







Procjena općeg stanja bolesnika

Table 1. The ECOG scoring system versus the Karnofsky scoring system

ECOG/WHO/Zubrod score		Karnofsky score	
Fully active, no restrictions	0	Normal, no evidence of disease Able to perform normal activity with only minor symptoms	100 90
Restricted in strenuous activity Ambulatory, can carry out work	1	Normal activity with effort Able to care for self but unable to do normal activities	80 70
Ambulatory >50% of the time Capable of self-care Unable to work/usual activities	2	Requires occasional assistance, cares for most needs Requires considerable assistance	60 50
Ambulatory ≤50% of the time Capable of limited self-care only	3	Disabled, requires special assistance Severely disabled	40 30
Disabled, no self-care Confined to bed or chair	4	Very sick, requires active support Moribund	20 10

ECOG = Eastern Cooperative Oncology Group; WHO = World Health Organization

Terapija

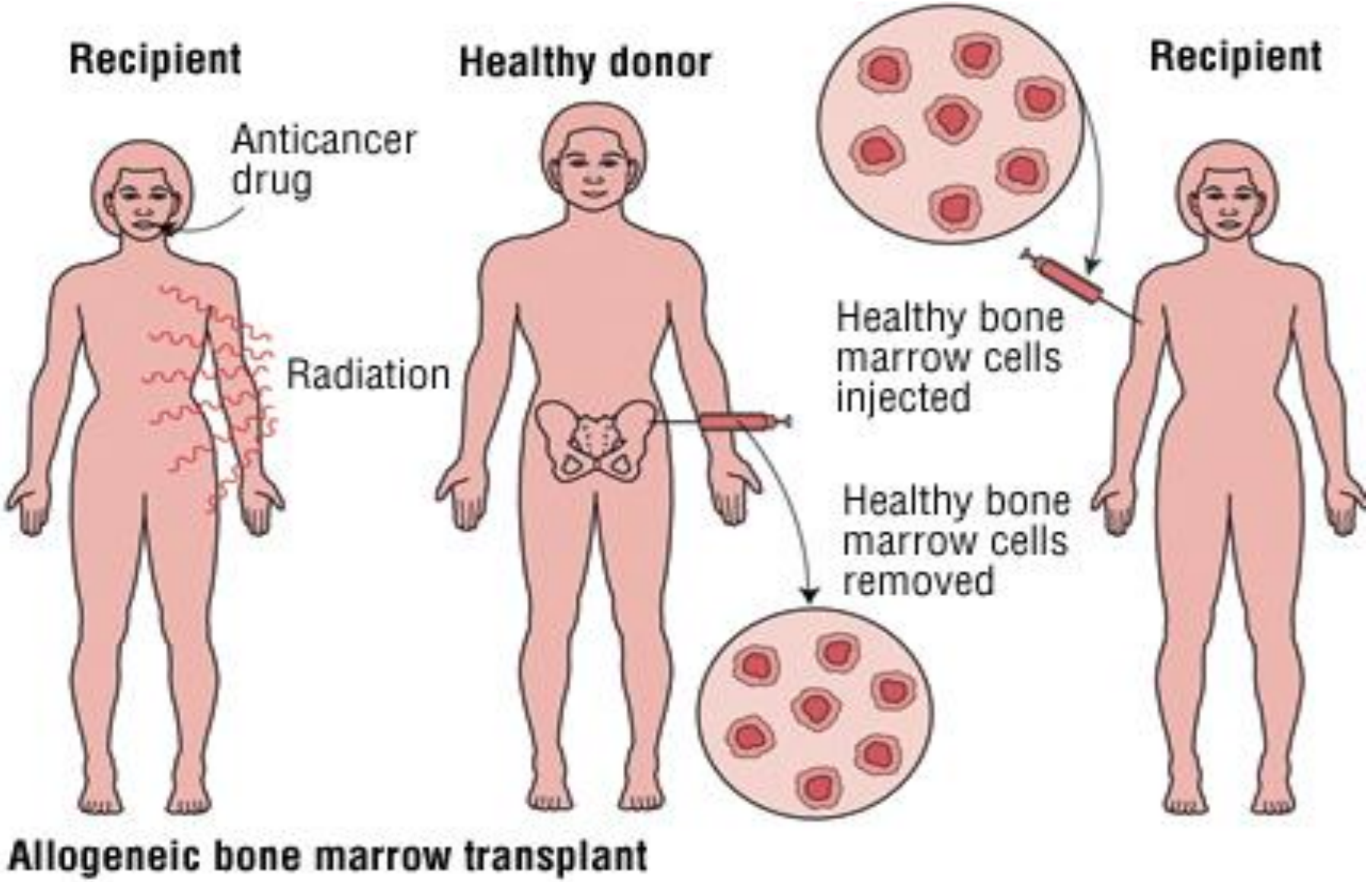
- Ovisi o tipu leukemije, dobi bolesnika, stupnju bolesti, te općem stanju
- Kemoterapija
- Zračenje
- Transplantacija koštane srži
- Potporna terapija

AKUTNA MIJELOIČNA LEUKEMIJA

- TERAPIJA

UVODNA TERAPIJA	II. CIKLUS KEMOTERAPIJE	KOMPLETNA REMISIJA	TRANSPLANTACIJA (alogena; autologna) IMUNOTERAPIJA

- ALL – remisija u 50% slučajeva - recidivi
- AML – remisija u 90% slučajeva- recidivi


Recipient
Anticancer drug
Radiation
Healthy donor
Healthy bone marrow cells removed
Healthy bone marrow cells injected
Recipient
Allogeneic bone marrow transplant

Koje se bolesti mogu liječiti krvotvornim matičnim stanicama?

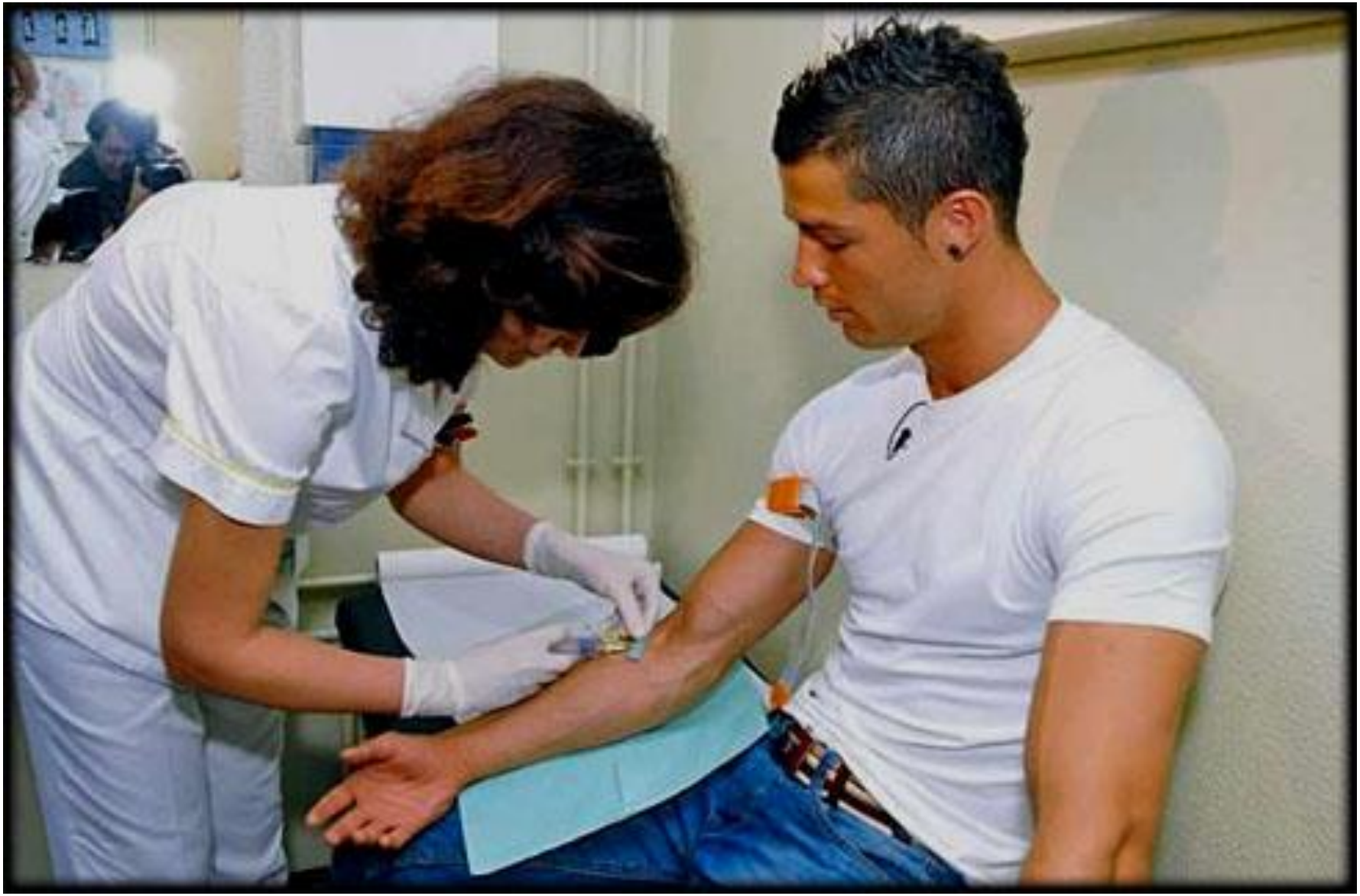
- Akutne i kronične leukemije
- Maligni ne-Hodgkinovi limfomi
- Hodgkinova bolest
- Multipli mijelom
- Aplastična anemija
- Hemoglobinopatije (npr. talasemija)
- Solidni tumori (ca dojke, tumori testisa, neuroblastom)

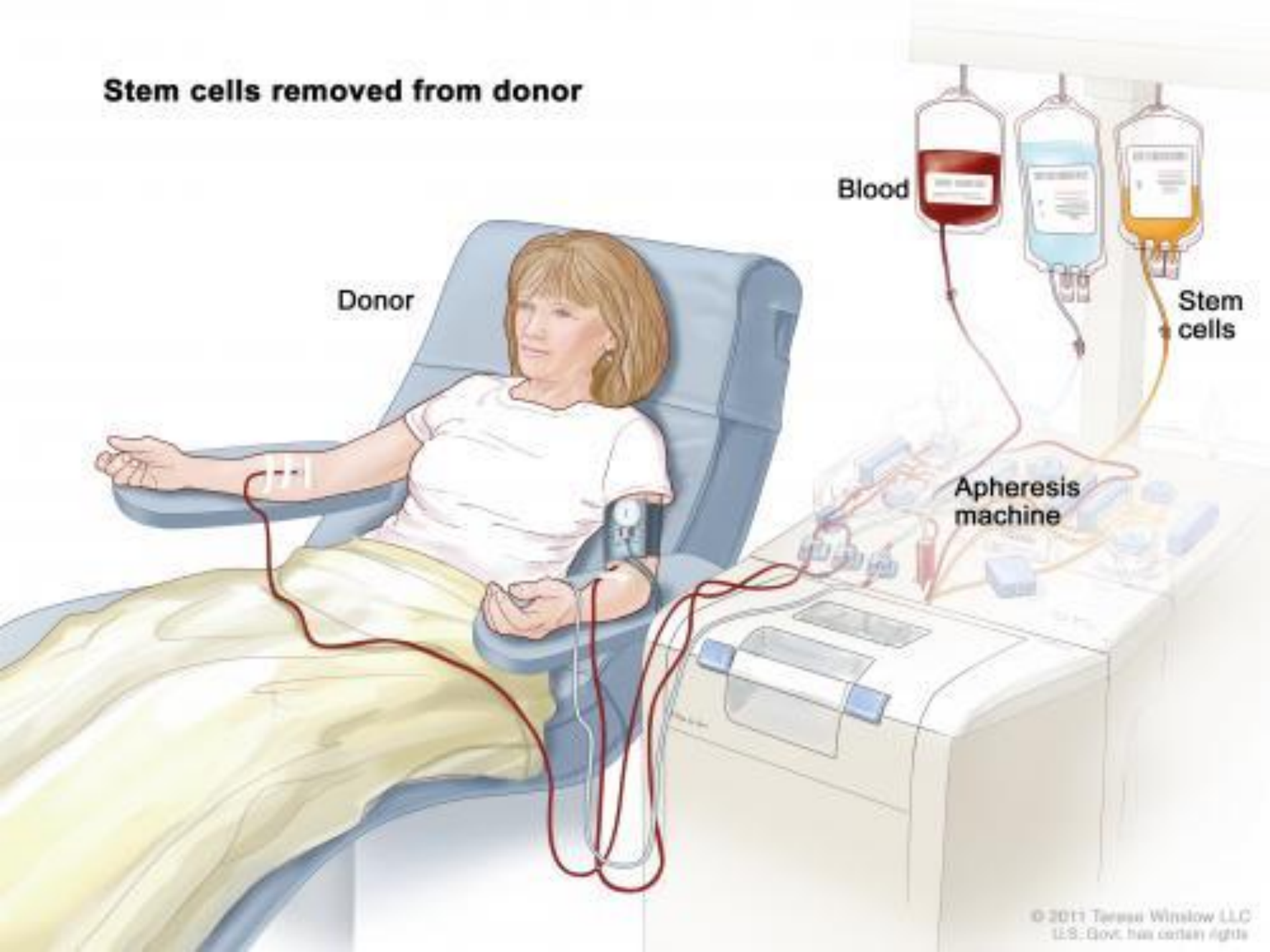
Tko i kako može pomoći?

- Svaka zdrava osoba u dobi od 18-40 godina
- Uzorak krvi (oko 7ml) da bi se odredili antigeni tkivne snošljivosti – registar
- Poziv za doniranje koštane srži ili periferne krvi (matične stanice)

Prikupljanje matičnih stanica iz periferne krvi

- Jednostavan i siguran postupak
- Rijetke i manje značajne neželjene reakcije
- Može se provoditi ambulantno
- Uz matične stanice prisutne i usmjerene prastanice te stanice – preteče što ubrzava oporavak i funkciju koštane srži primatelja




Stem cells removed from donor
Blood
Donor
Stem cells
Apheresis machine

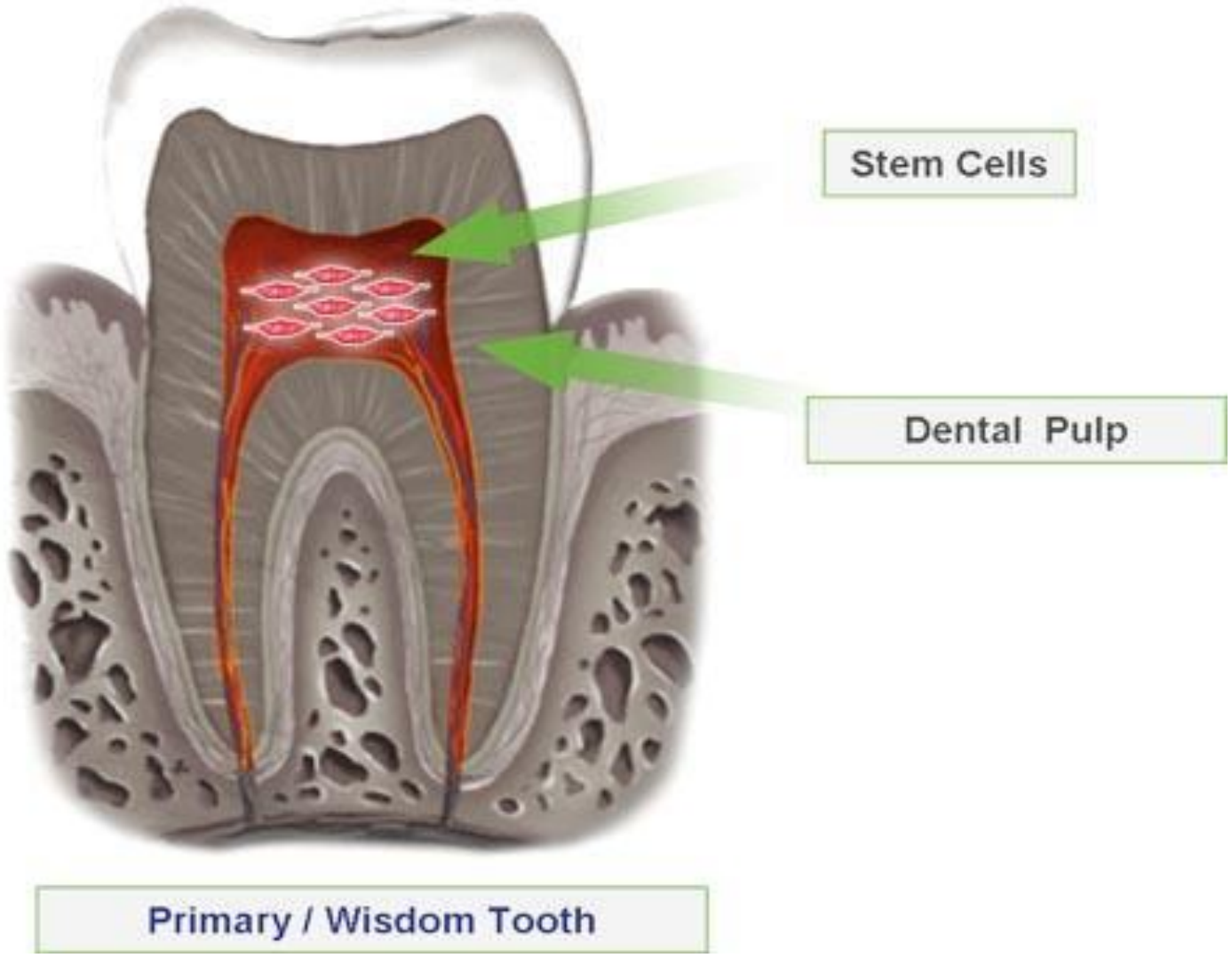

MVE
CONSARCTIC
URO-CY

31. ožujka 2020.

Broj potencijalnih darivatelja u registru
64 249

broj tipiziranih darivatelja

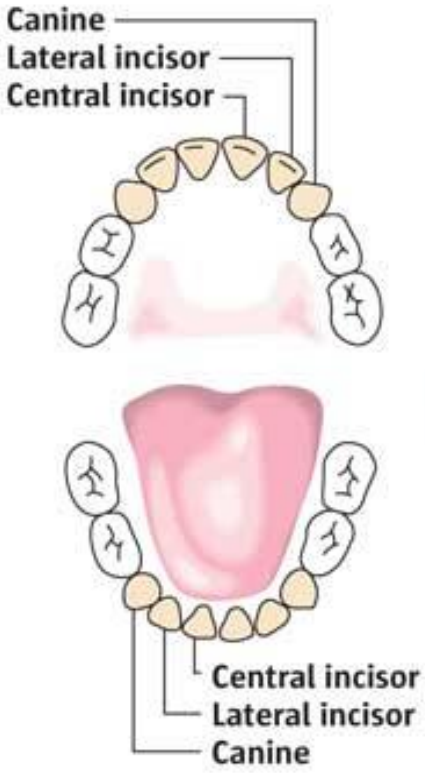
61 524


Stem Cells
Dental Pulp
Primary / Wisdom Tooth

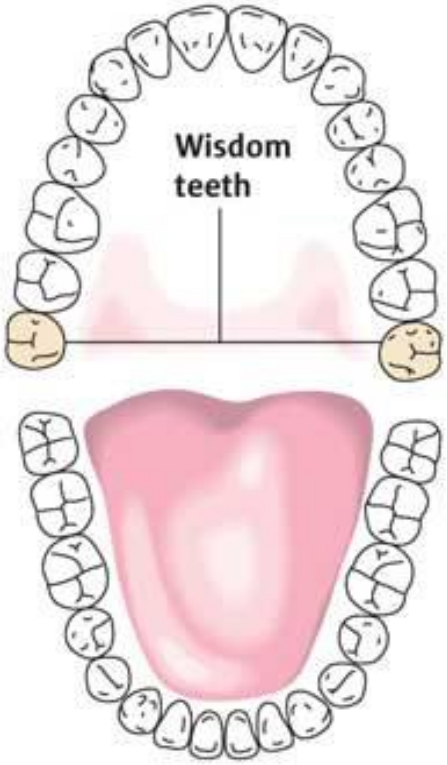
Recovering stem cells

Companies collecting stem cells say it is best to recover them when patients are young, but they can be retrieved at any age if the teeth are healthy.

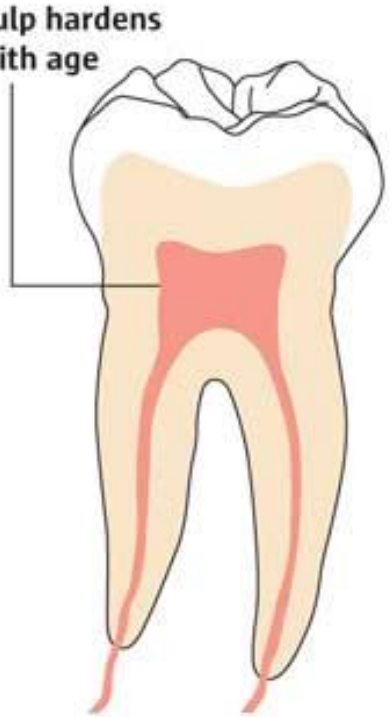
Baby teeth, *age 6-12*
Best sources are from canine to canine before the teeth fall out on their own.



Wisdom teeth, *age 16-20*
Third molars are a good source for stem cells, although it is best when the teeth are still developing.



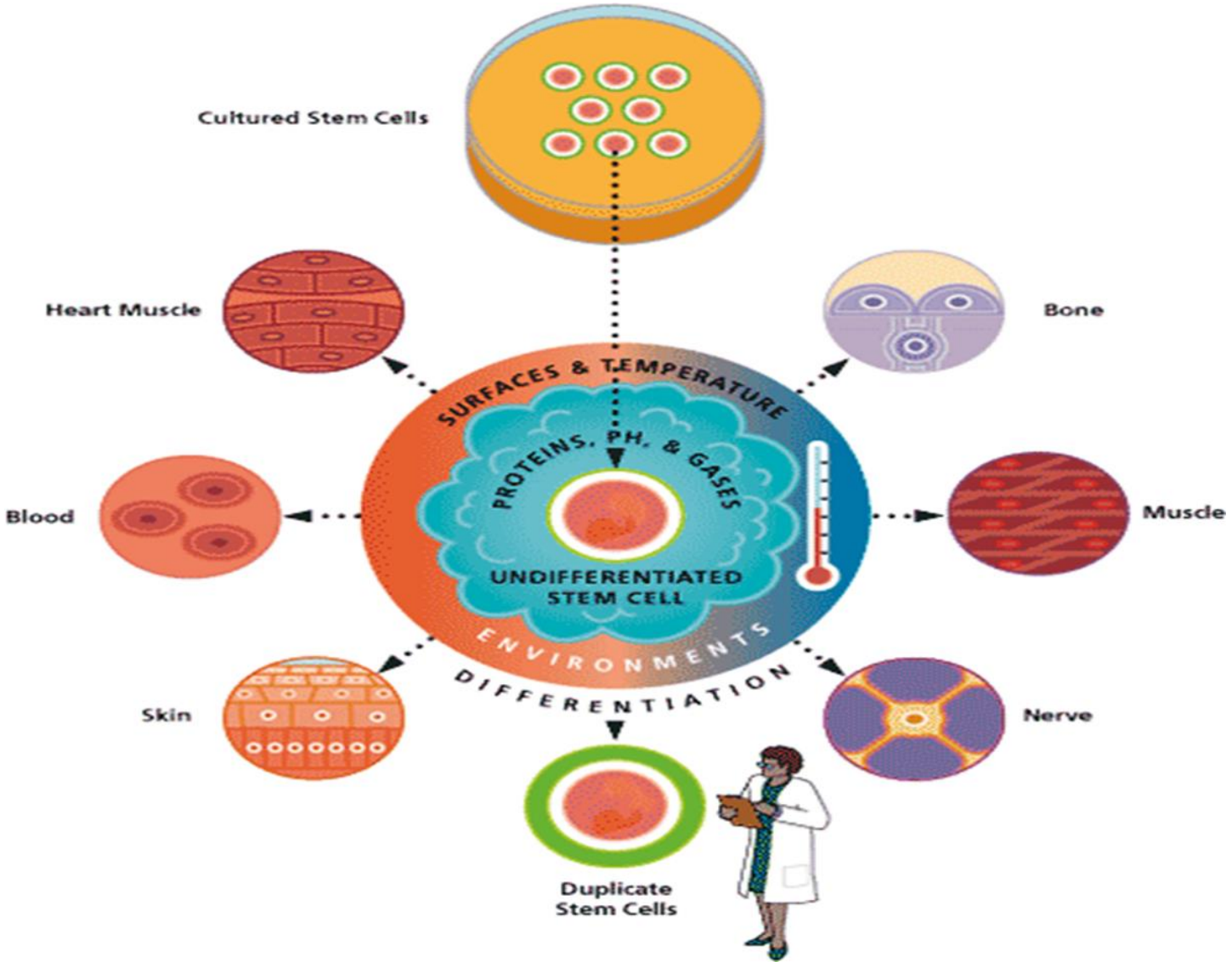
Permanent teeth, *over age 20*
All adult teeth with healthy pulp are potential sources. As people age, dental stem cells become less useful.



Source: StemSave; National Institutes of Health; The Human Body

Thomas McKay, The Denver Post

Matične stanice iz pupkovine

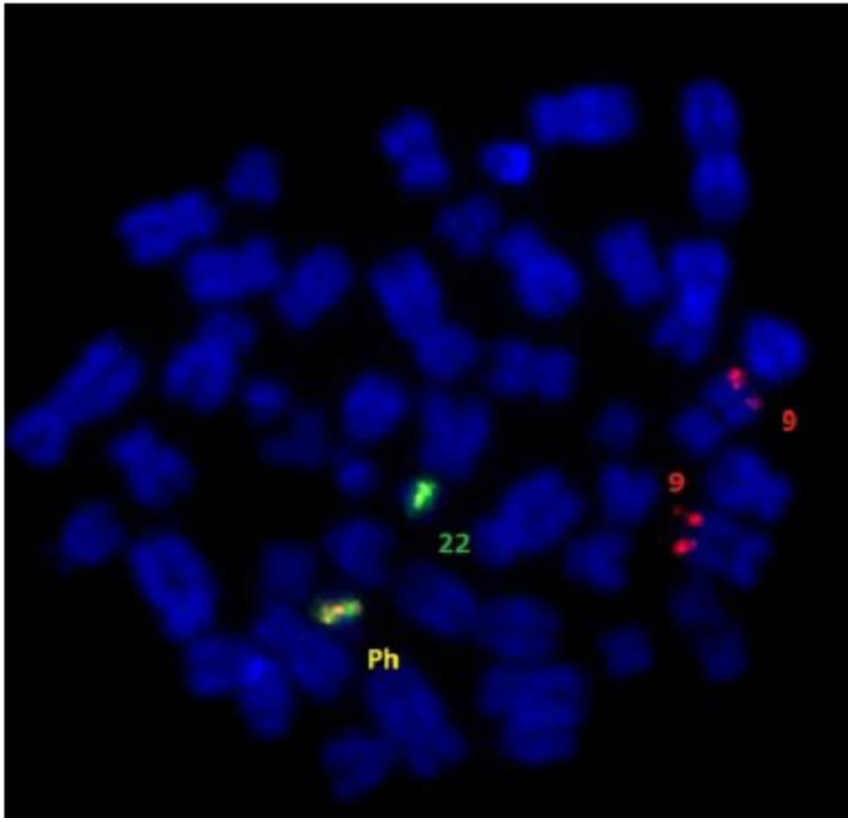


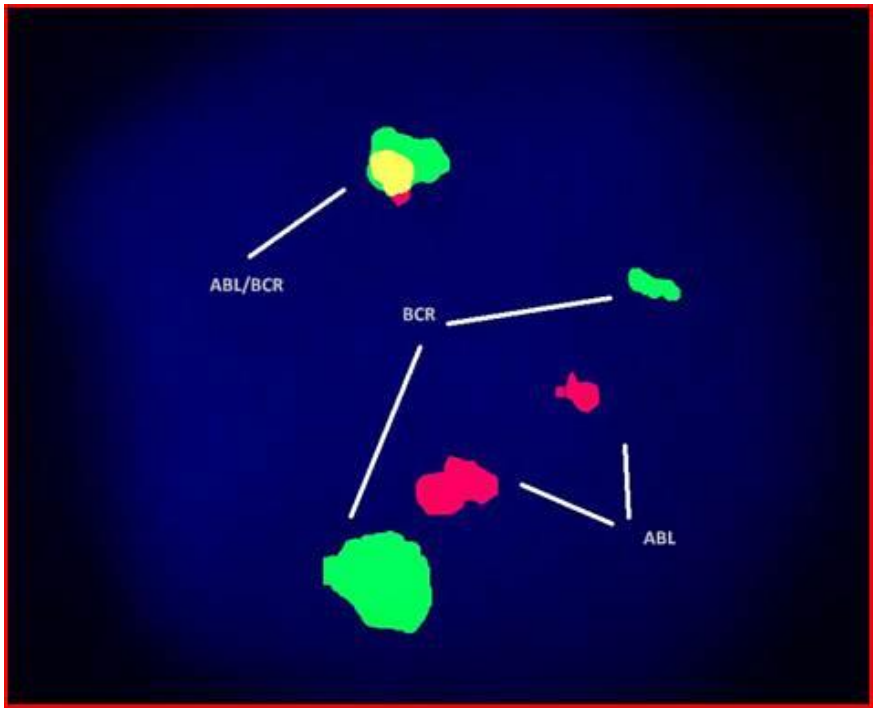
KRONIČNE MIJELOPROLIFERATIVNE BOLESTI

KRONIČNA MIJELOIČNA LEUKEMIJA

- Bolest pluripotentne matične stanice hematopoeze, karakterizirana masivnom ekpanzijom mijeloidne loze
- U više od 90% bolesnika nalazi se kromosomski poremećaj Ph1 kromosom

FISH na Philadelphia kromosom (t (9,22))

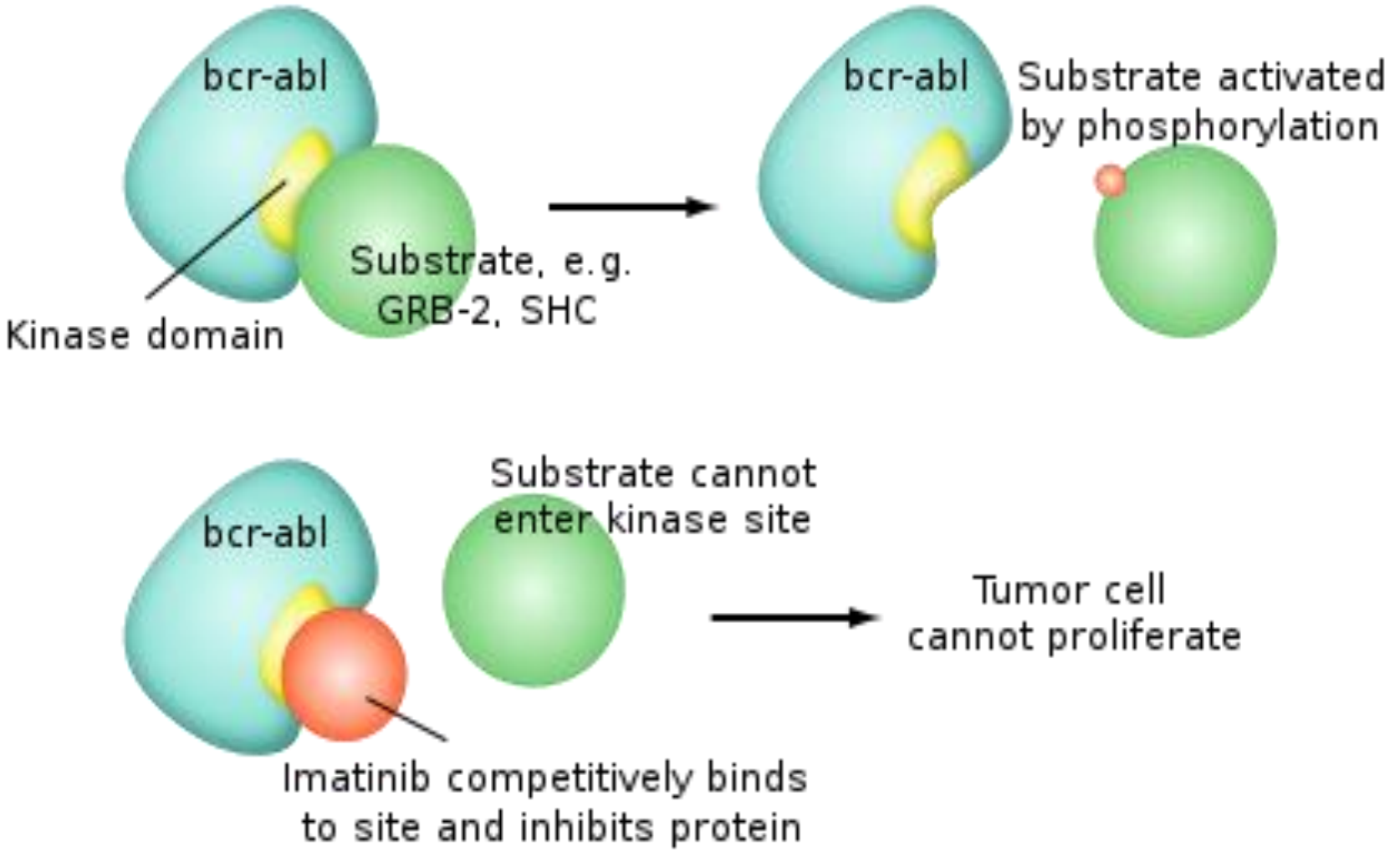




KRONIČNA MIJELOIČNA LEUKEMIJA

- Dijagnoza – periferna krv i koštana srž
  - leukocitoza 50-200x10^9 (zreli neutrofilni granulociti i nezreli oblici sve do mijeloblasta; broj promijelocita i mijeloblasta nije veći od 20% - znak blastne transformacije)
  - Normocitna anemija, obično trombocitoza, bazofilija i eozinofilija
  - alkalna fosfataza u leukocitima je niska ili odsutna


TIME
THERE IS NEW AMMUNITION IN THE WAR AGAINST
CANCER.
THESE ARE THE BULLETS.
Revolutionary new pills like GLEEVEC combat cancer by targeting only the diseased cells. Is this the breakthrough we've been waiting for?


bcr-abl
Substrate, e.g.
GRB-2, SHC
Kinase domain
bcr-abl
Substrate activated
by phosphorylation
bcr-abl
Substrate cannot
enter kinase site
Tumor cell
cannot proliferate
Imatinib competitively binds
to site and inhibits protein

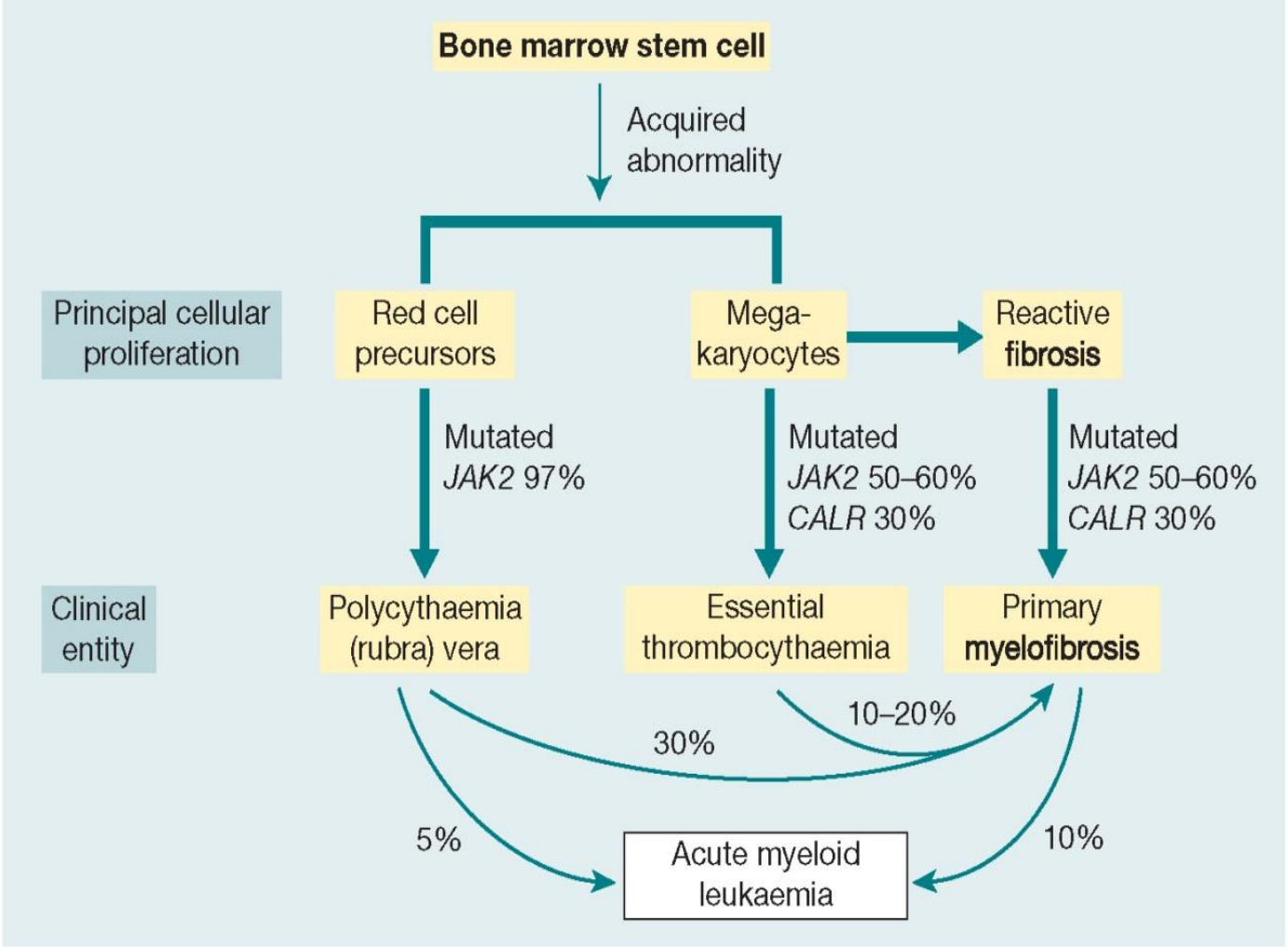


Figure 15.1 Relationship between the three myeloproliferative diseases. They may all arise by somatic mutation in the pluripotential stem and progenitor cells. Many transitional cases occur showing features of two conditions and, in other cases, the disease transforms during its course from one of these diseases to another or to acute myeloid leukaemia. The three diseases, polycythaemia rubra vera, essential thrombocythaemia and primary myelofibrosis, are characterized by *JAK2* or *CALR* mutation in a varying proportion of cases.

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POLICITEMIJA RUBRA VERA

- Neoplastična, klonalna mijeloprolifearativna bolest nastala transformacijom pluripotentne matične stanice i zahvaća sve krvne loze
- Nepoznate etiologije
- Bolest starije životne dobi (50-70g.)
- Incidencija 0,5-2/100 000 stanovnika

POLICITEMIJA RUBRA VERA SIMPTOMI

- Izrazito crvenilo lica
- Slabost
- Glavobolja
- Vrtoglavice
- Zujanje u ušima
- TIA
- Cijanoza distalnih dijelova ekstremiteta
- Klaudikacije

Figure 15.4 Polycythaemia vera: facial plethora and conjunctival suffusion in a 63-year-old woman. Haemoglobin 180 g/L; total red cell volume 45 mL/kg.

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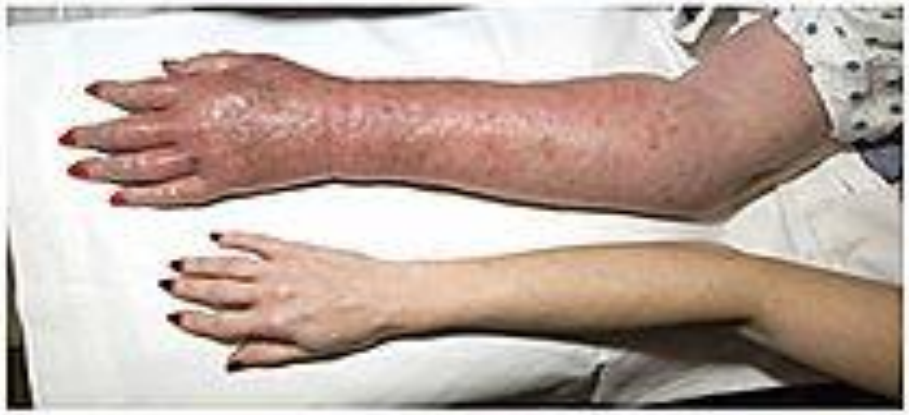
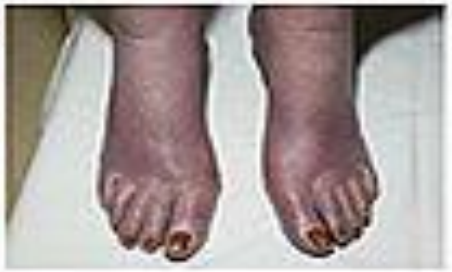




POLICITEMIJA RUBRA VERA LABORATORIJSKI NALAZI

- ⇑ eritrociti, hemoglobin i hematokrit
- ↑↑ masa eritrocita u odnosu na volumen krvi
- Leukociti i trombociti uglavnom ⇑
- Uredna saturacija arterijski krvi kisikom
- Eritropoetin izrazito ↓ ili nemjerljiv
- Pozitivna mutacija JAK 2 gena

POLICITEMIJA RUBRA VERA DIFERENCIJALNA DIJAGNOZA

- SEKUNDARNA ERITROCITOZA

-VME kao u PRV, ali eritropoetin ↑↑uslijed

a) hipoksije (kronična plućna bolest, alveolarna hipoventilacija ili srčani D-L spoj)

b) abnormalnog hemoglobina (↑↑ afinitet za O_2)

c) tm bubrega, jetre, malog mozga ili maternice

- RELATIVNA ILI LAŽNA ERITROCITOZA

- ↓ volumen krvi uz uredan VME, te bez znakova mijeloproliferacije uslijed;

a) stresa i drugih stanja uslijed povišenog tonusa simpatikusa

b) dehidracije

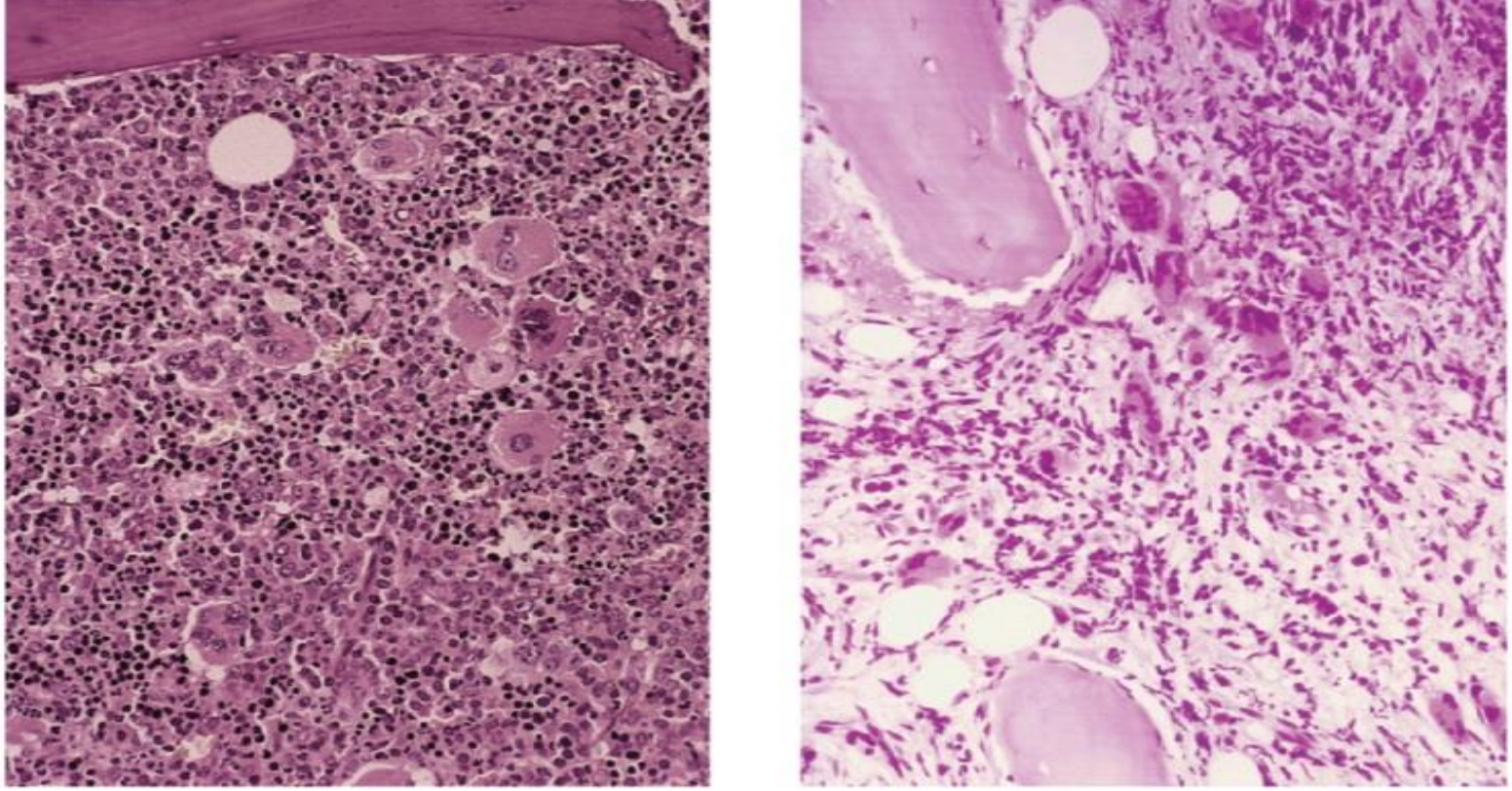
POLICITEMIJA RUBRA VERA TERAPIJA

- CILJ- CITOREDUKCIJA (Htc<0,45)

a) Venepunkcija 350-500ml (oprez- sideropenija)

b) Citostatik- hidroksiurea (oprez- urična dijateza)

c) Antiagregacijska- ASK (oprez- krvarenje)



(a) (b)

Figure 15.7 Iliac crest trephine biopsies. **(a)** Polycythaemia vera: fat spaces are almost completely replaced by hyperplastic haemopoietic tissue. All haemopoietic cell lines are increased with megakaryocytes particularly prominent. **(b)** Primary myelofibrosis: normal marrow architecture is lost and haemopoietic cells are surrounded by increased fibrous tissue and intercellular substance.

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ESENCIJALNA TROMBOCITEMIJA

- ↑megakariocitopoeza s trombocitozom (> $600x10^9$/L često iznad 1 $000x10^9$/L)
- JAK 2 pozitivan u oko 50% bolesnika
- Najčešća mijeloproliferativna bolest-0,6-2,5/100000
- U dobi 20-80 godina (medijan 60g, M:Ž=1:1)
- Moguća krvarenja i/ili tromboembolije

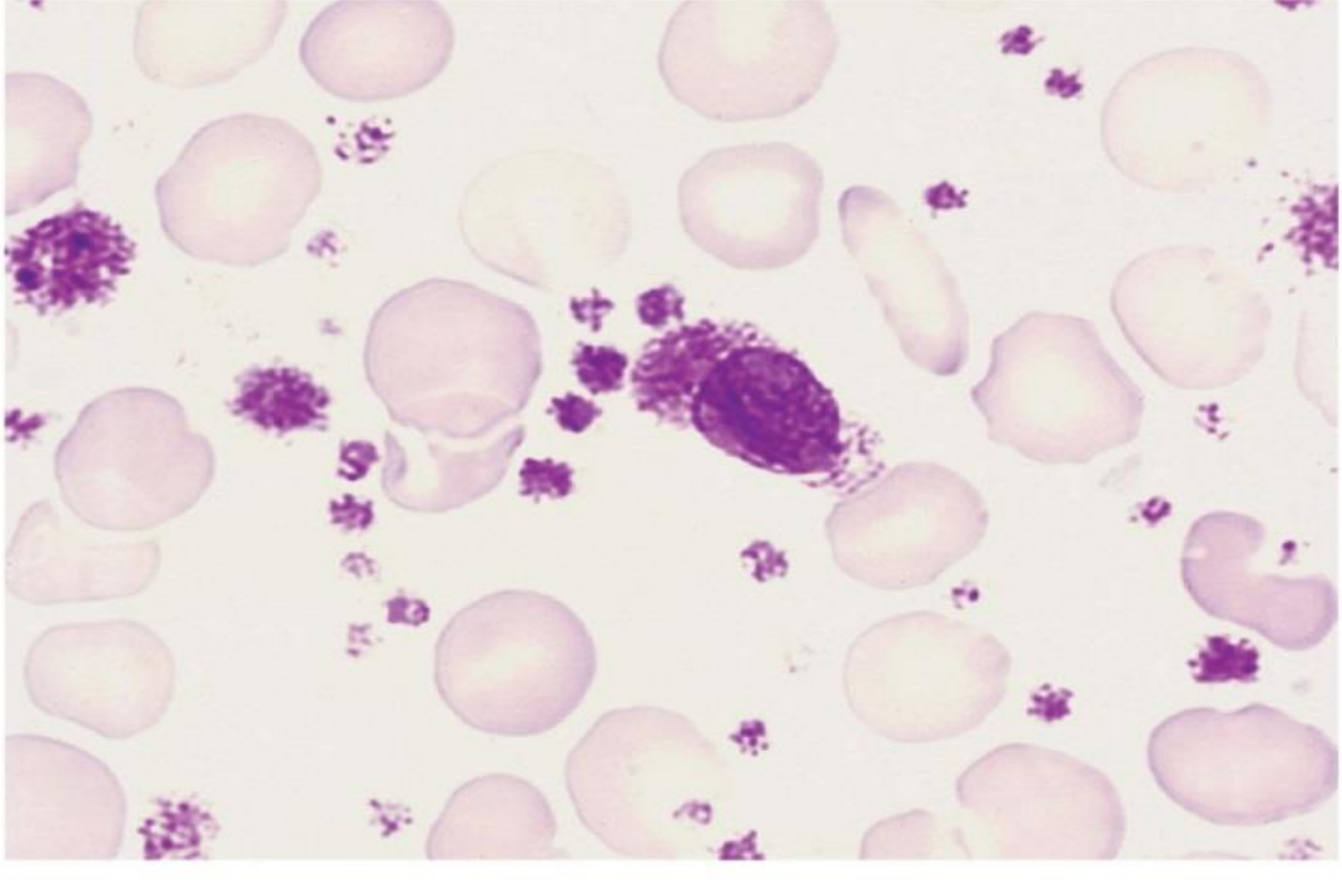

Figure 15.9 Peripheral blood film in essential thrombocythaemia showing increased numbers of platelets and a nucleated megakaryocytic fragment.

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SEKUNDARNE TROMBOCITOZE

- Upale
- Tumori
- Sideropenija
- Hemolitička anemija
- Krvarenje
- Stres
- Splenektomija
- Druge mijeloproliferativne bolesti

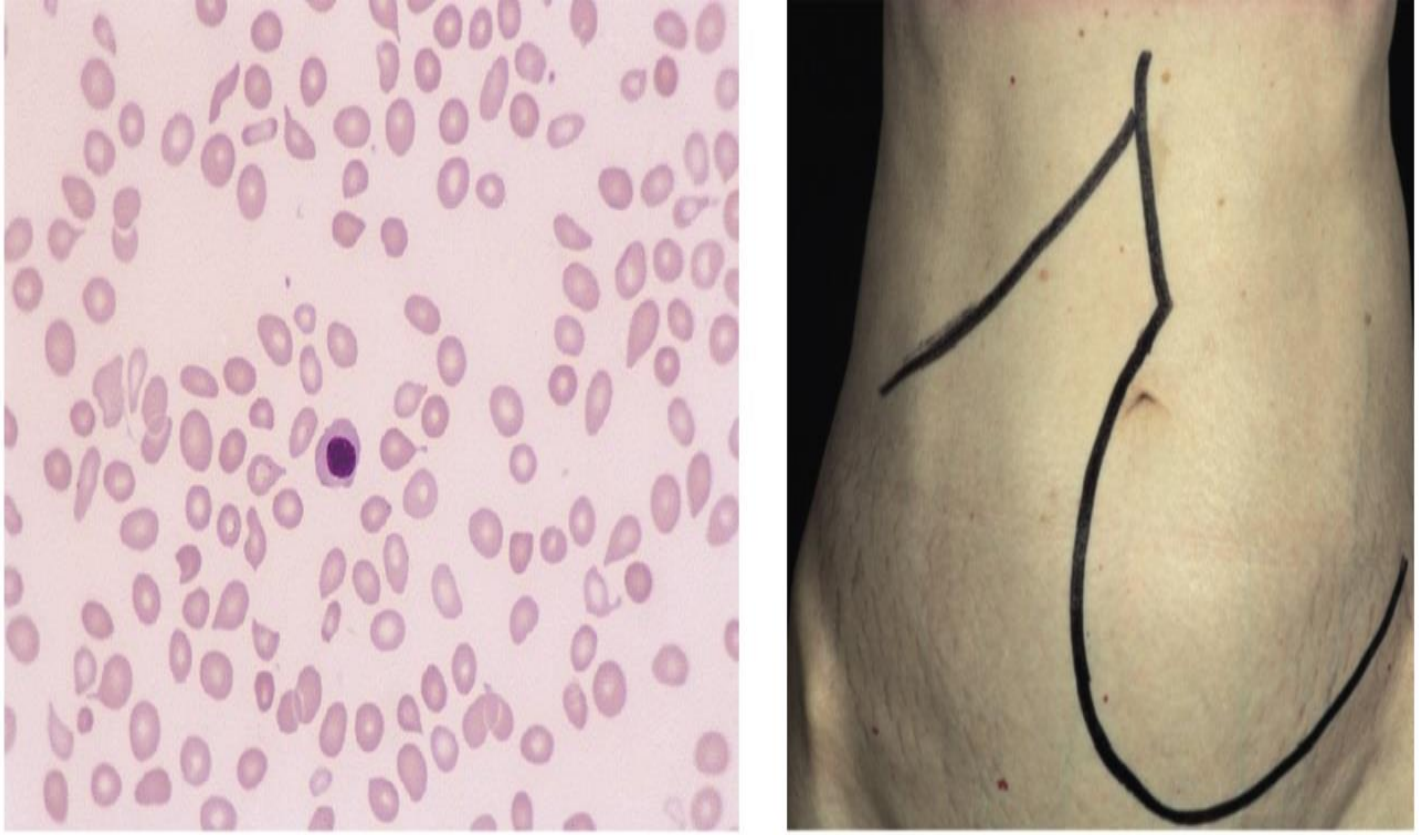
ESENCIJALNA TROMBOCITEMIJA TERAPIJA

Cilj-citoredukcija TRB na 450-600x10^9/L

a) citostatik- hidroksiurea
b) α interferon
c) Anagrelid
d) Antiagregacijska- ASK

IDIOPATSKA MIJELOFIBROZA

- 1-2/100 000 stanovnika (M:Ž=1:1)
- U starijoj populaciji (od 50g)
- Fibroza koštane srži
- Splenomegalija- ekstramedularna hematopoeza
- Leukoeritroblastoza+dakriociti
- Suportivna terapija, splenektomija, zračenje slezene
- Medijan preživljenja 4-5 godina



(a) (b)

Figure 15.10 (a) Peripheral blood film in primary myelofibrosis. Leucoerythroblastic change with 'tear-drop' cells and an erythroblast. **(b)** Massive splenomegaly in a patient with myelofibrosis.

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