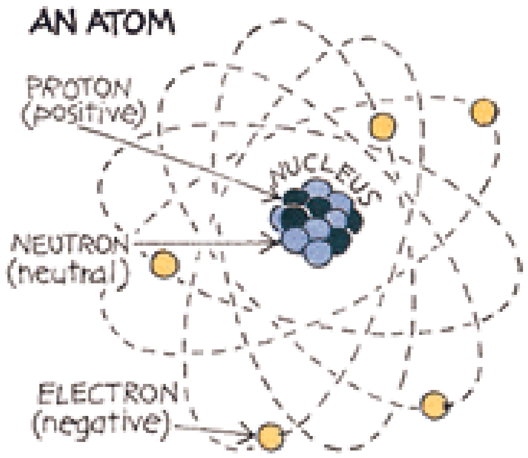
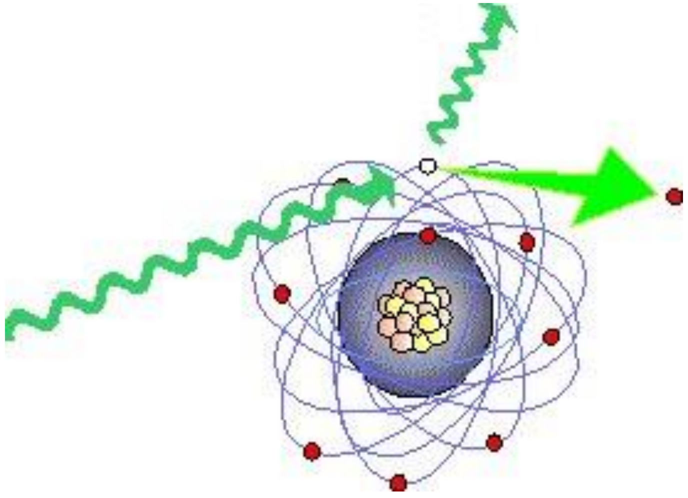
Experimentally induced acute radiation syndrome in experimental animal

What is ionizing radiation?

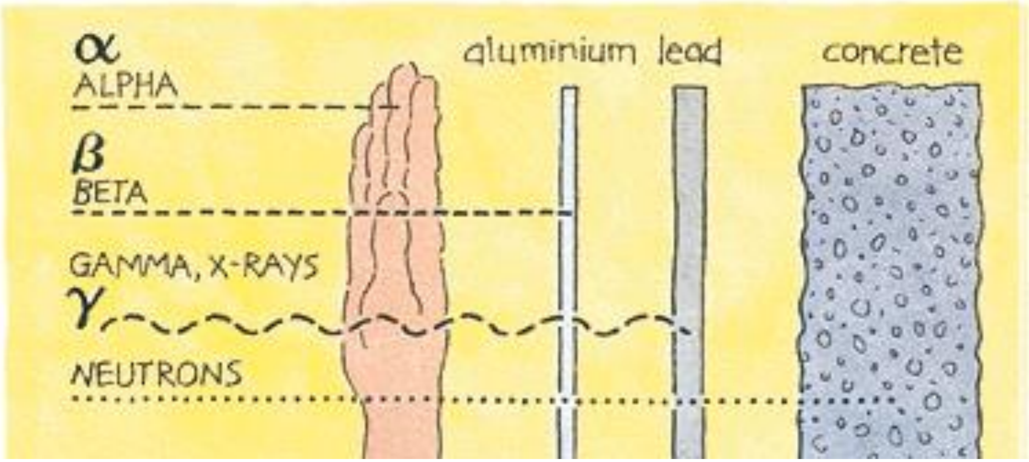
particles or **electromagnetic radiation**, where the particles / photons carry enough energy to ionize atoms and molecules (by removing the electron from their orbit).
It produces electrically charged particles (= ions)
Ionization is biologically very important in macromolecules that are encompassed within the human body.

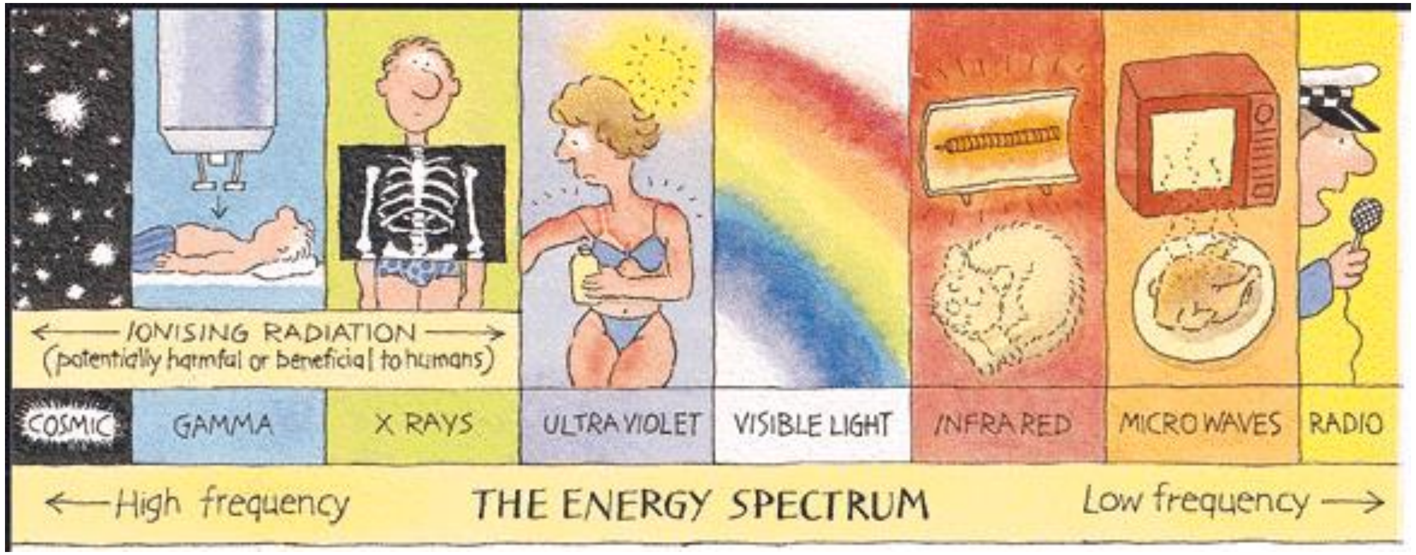




Types of ionizing radiation?

α = α–particles (atoms of helium
β = electornes or positrones
γ = electromagnetic radiation (photons)
neutrons





Units?

the dose of ionizing radiation received by a person is expressed as absorbed energy, the unit being gray (Gy)
1Gy = 1 J / kg (formerly rad)
same dose in Gy of different types of radiation causes different biological effect (1Gy α -radiation has greater effect than 1Gy β -radiation) $\rightarrow$ radiation effect is expressed as effective dose, unit is sievert (Sv)
irrespective of the type of radiation, 1 Sv leads to the same biological effect
example: 1Gy = 1Sv for β - or γ radiation, 1Gy = 10Sv for neutrons and 1Gy = 20Sv for α-radiation
rate of radioactive decay of radioactive material expressed by units of becquerel (Bq)
1 Bq = 1 atomic decay / s

What is radioactivity?

most atoms are stable: carbon-12 or oxygen-16
some have an excess of internal energy and decay spontaneously to form new elements = "radioactive decay"
in decay, excess internal energy is released as γ-radiation or particles

Sources of ionizing radiation?

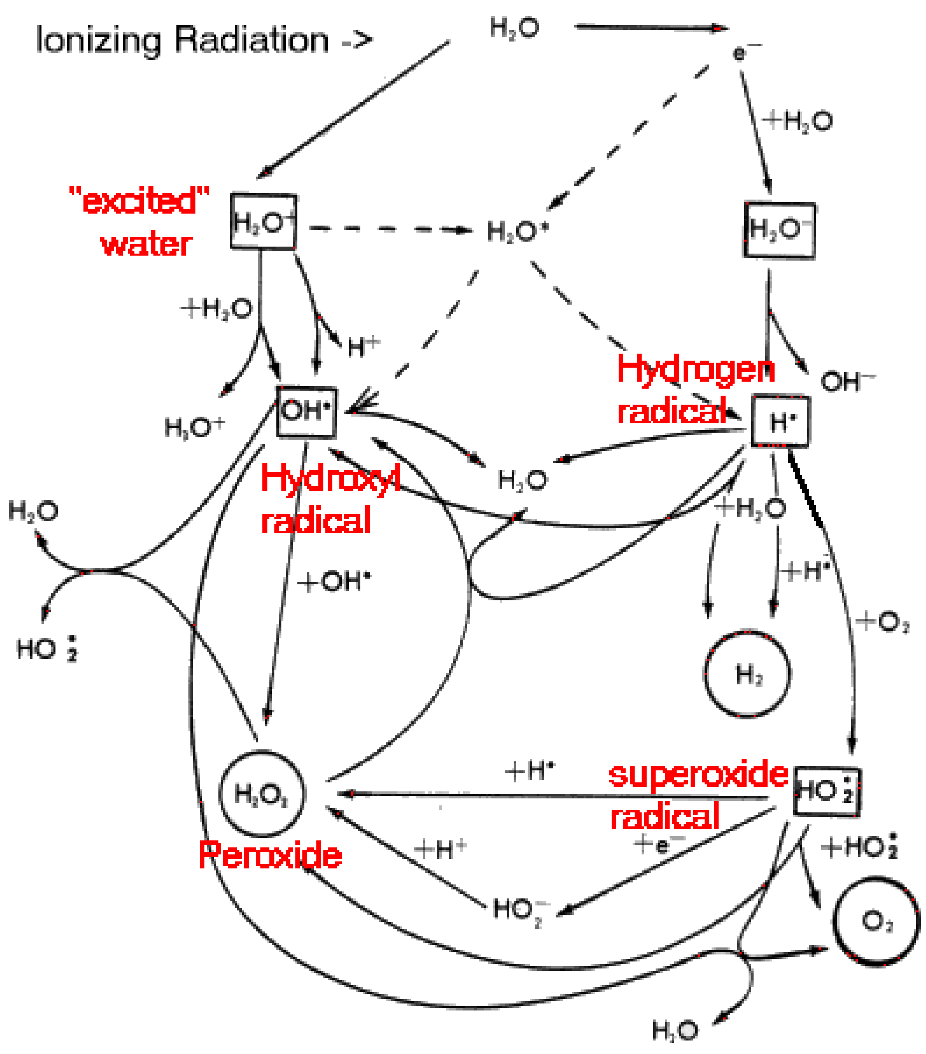
Natural
cosmic
exposure increases with altitude
solar
especially γ-radiation
terrestrial resources
radioactive decay of natural radioisotopes (soil and rock)
Radon
gas, formed by decay of Radio-226 (from uranium)
has the largest share of the total. dose of ionizing radiation

Artificial
medicine
diagnostics, therapy, sterilization
industry
nuclear energy production
agriculture

Biological effects and consequences of ionizing radiation?

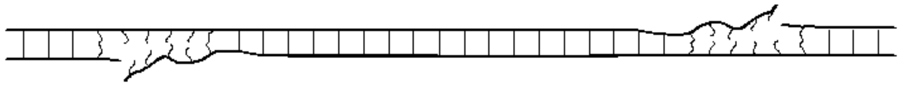
Direct ionization of macromolecules
Indirectly through "radiolysis" of water
free oxygen radicals
Consequences:

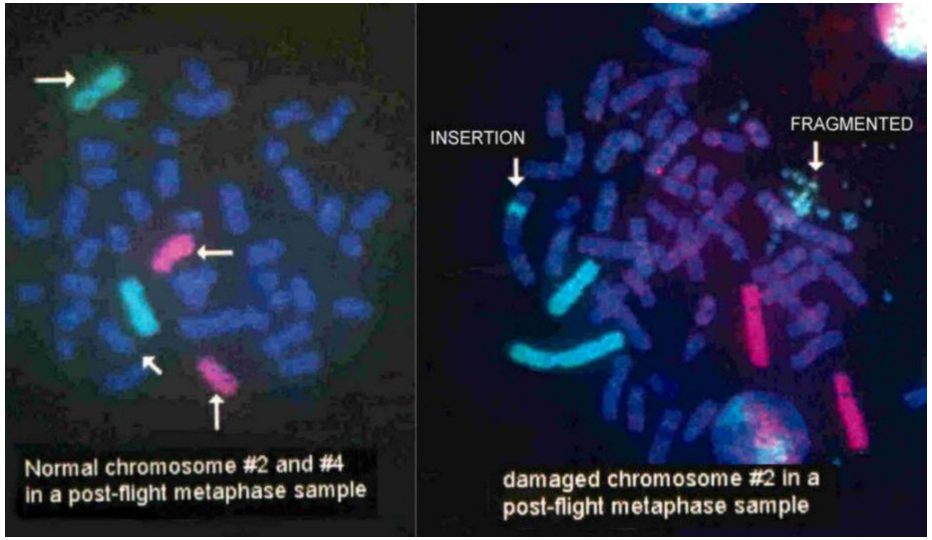
cycle blockade → apoptosis
mitotic or post-mitotic death (proliferating cells)
mutation (gene or chromosome)
reparation
unrepaired change



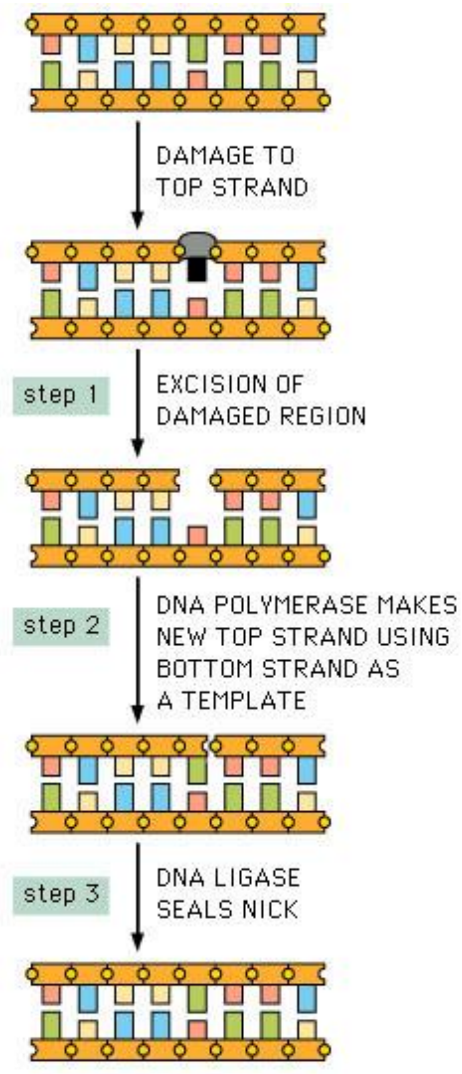
Types and consequences of DNA lesions?

point mutations
DNA repair: mismatch repair
Single Strand Breaks (SSB)
DNA repair: base excision repair
Double Strand Breaks (DSB)
lethal (apoptosis)
DNA repair: homologous recombination
(sometimes)
sometimes non-homologous repair
translocation
insertion





DNA repair



(*in situ* repair)

base excision repair
nucleotide excision repair
mismatch repair

Character of biological effect

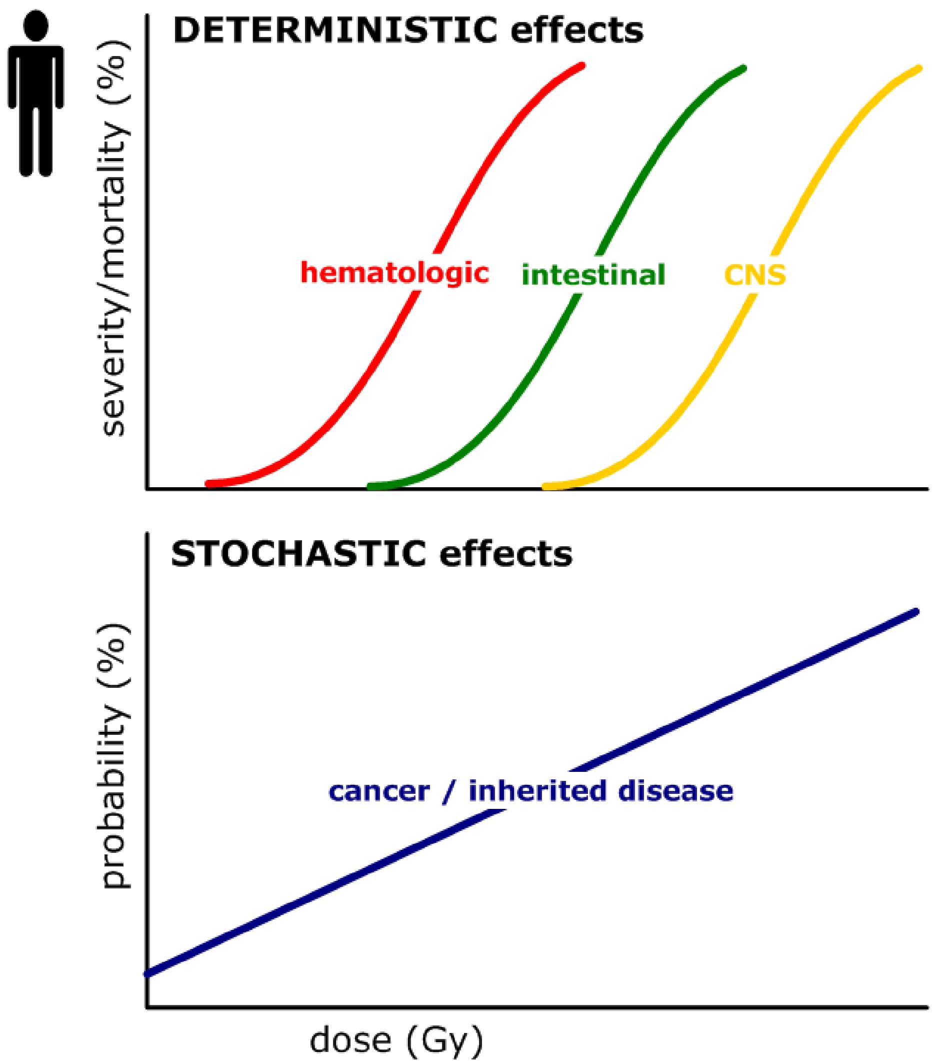
Deterministic

severity depends („is determined by") on the dose
specific manifestation
damage to typical tissues and organs
the effect occurs only when the threshold dose is exceeded
damage is due to the death of a large number of cells
onset of symptoms soon after exposure (short latency)
Types:
acute radiation syndrome (ac. radiation sickness)
whole-body irradiation with a dose> 1Gy
chronic post-radiation syndrome (general or local)
sterility, cataract, radiation dermatitis, alopecia, endarteritis obliterans, pneumonitis,…
fetal damage in utero

Stochastic

probability increases with dose (not severity!)
non - specific manifestation
damage to various tissues and organs
a smooth risk increase without a “safe” threshold dose
single cell damage is sufficient
delayed manifestation (long latency, typically years)
Types:
somatic mutations - tumors
leukemia, št. gland, lungs, ml. gland, skeleton
germinative mutations (oocyte, sperm) - congenital genetic defect

Deterministic × stochastic

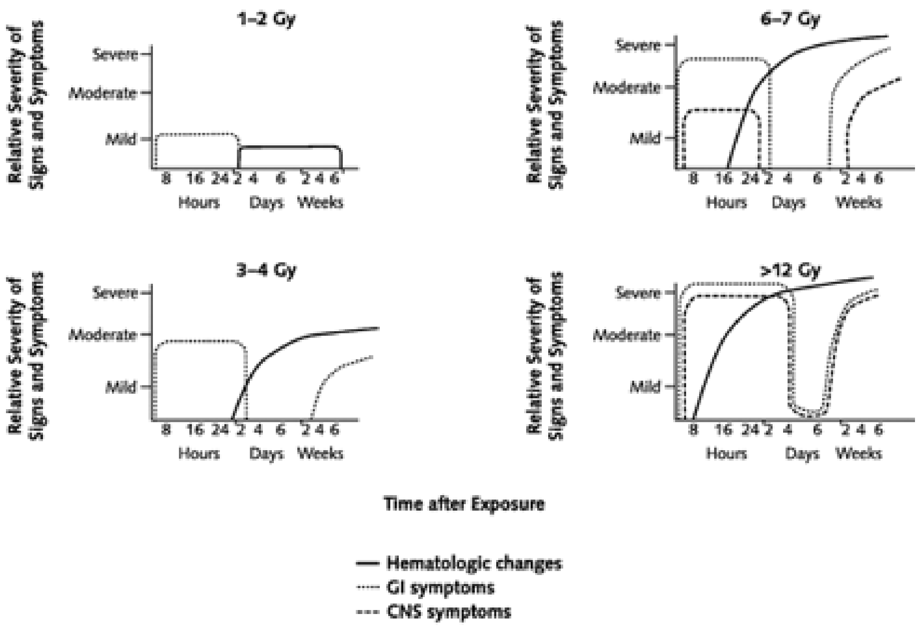


Acute radiation syndrome

it affects the hematopoietic, gastrointestinal and cerebrovascular systems

time course, extent and severity depends by dose – it is a typical deterministic effect !!!

from several hours to several months after exposure

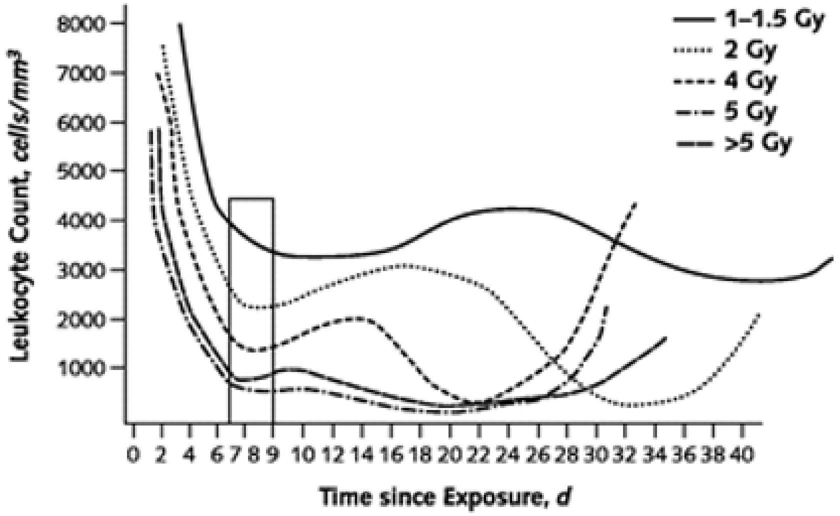


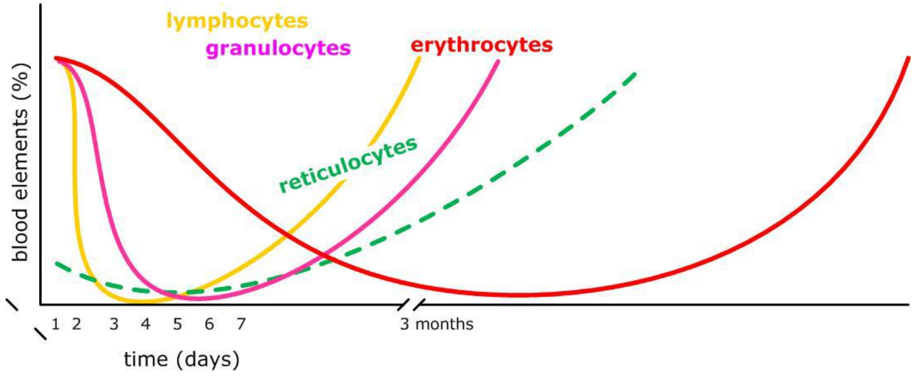
Acute radiation syndrome

Hematopoietic syndrome (> 1Gy)
1) reticulocytopenia, lymphopenia + granulocytosis
2) granulocytopenia (immunodeficiency)
3) thrombocytopenia (bleeding)
4) anemia (hypoxia)
GIT syndrome (> 10Gy)
early (hours) - nausea, vomiting, diarrhea
late (days) - loss of intestinal integrity
malabsorption, dehydration, toxemia / sepsis, ileus, bleeding
Cerebrovascular syndrome (tens of Gy)
headache, cognitive impairment, disorientation, ataxia, convulsions, exhaustion and hypotension
Cutaneous
erythema, burns, edema, impaired wound healing
epilation

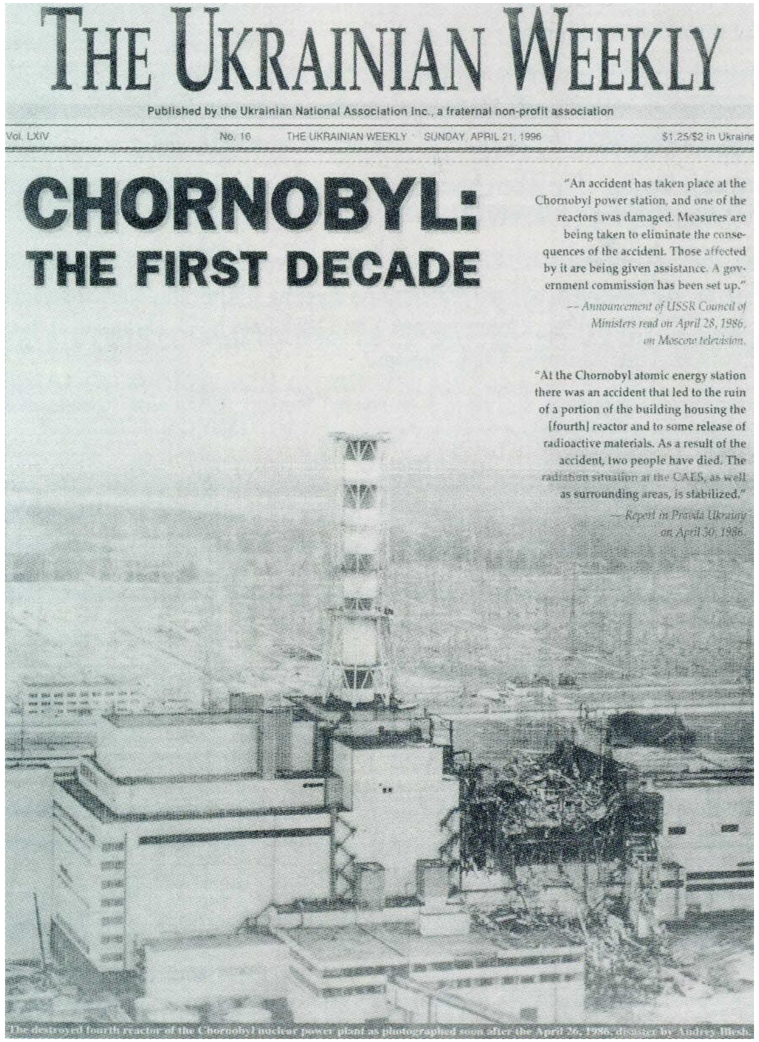
Hematopoietic syndrome

irradiation of bone marrow (> 1Gy) leads to exponential cell death - hematological crisis
hypoplasia to pulp aplasia + peripheral pancytopenia (infection, bleeding)
subpopulation of stem cells. it is selectively more radio-resistant (probably due to the prevalence of bb in the Go phase)
necessary for regeneration
anemia is a late consequence (erythrocytes ~ 120 days)!
massive stress reactions (glucocorticoids) contribute to lymphopenia (cytolytic effect) and paradoxically delay the onset of granulocytopenia (release of stocks of granulocytes from the pulp and spleen)





Exemplární příklad?



THE UKRAINIAN WEEKLY

Published by the Ukrainian National Association Inc., a fraternal non-profit association

Vol. LXIV | No. 16 | THE UKRAINIAN WEEKLY | SUNDAY, APRIL 21, 1996 | $1.25/$2 in Ukraine

CHORNOBYL: THE FIRST DECADE

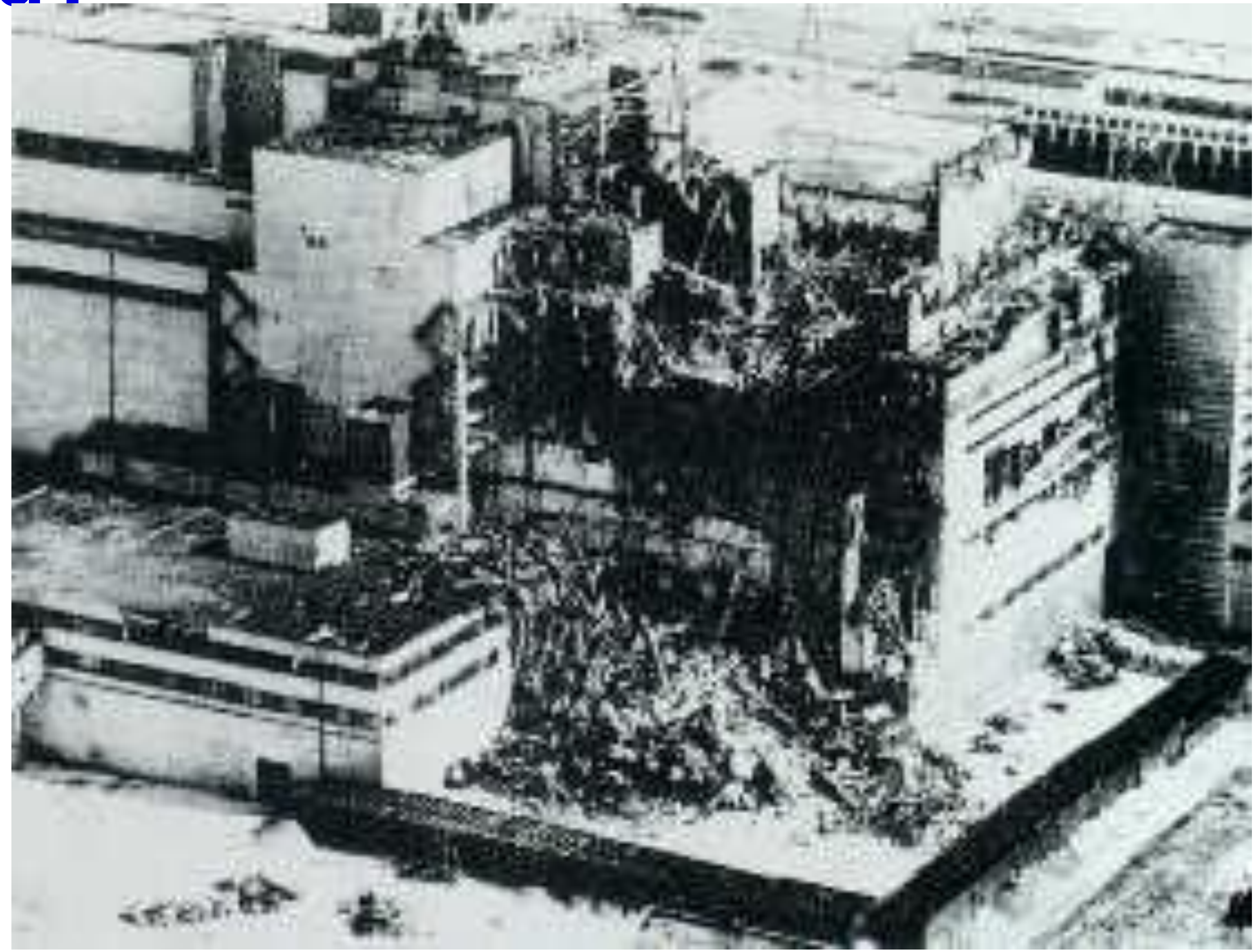
"An accident has taken place at the Chornobyl power station, and one of the reactors was damaged. Measures are being taken to eliminate the consequences of the accident. Those affected by it are being given assistance. A government commission has been set up."

— Announcement of USSR Council of Ministers read on April 28, 1986, on Moscow television.

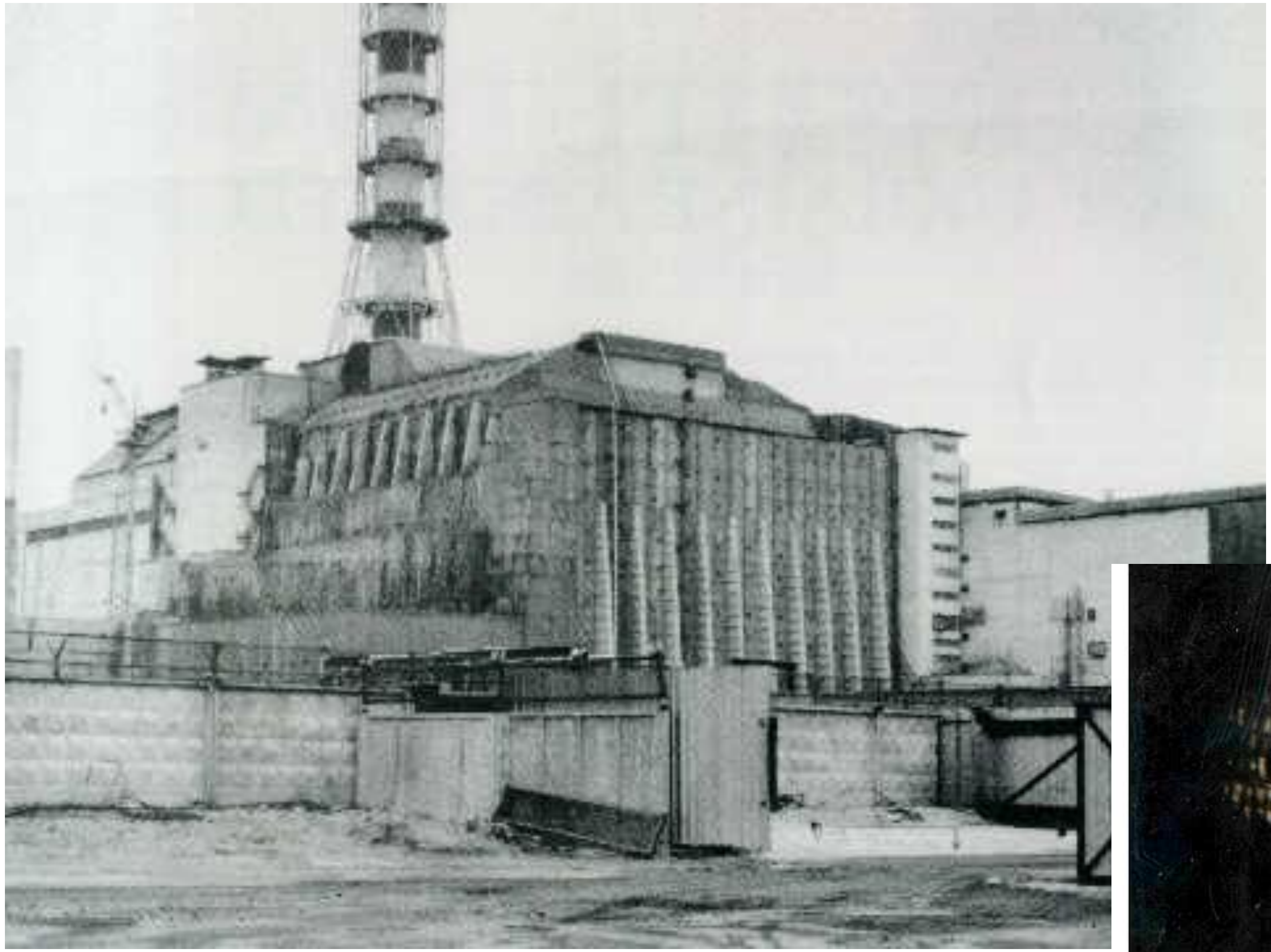
"At the Chornobyl atomic energy station there was an accident that led to the ruin of a portion of the building housing the [fourth] reactor and to some release of radioactive materials. As a result of the accident, two people have died. The radiation situation at the CAES, as well as surrounding areas, is stabilized."

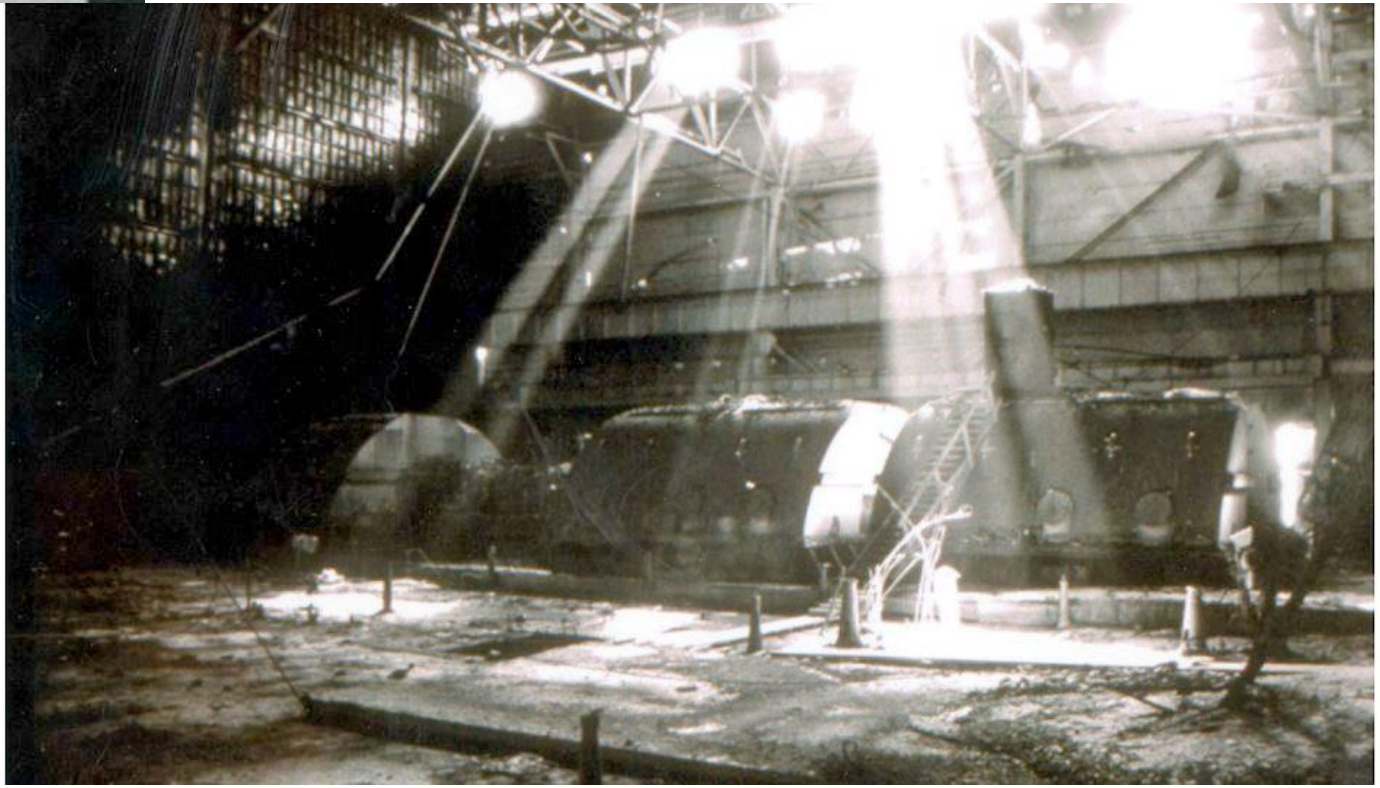
— Report in Pravda Ukrainy on April 30, 1986.

The destroyed fourth reactor of the Chornobyl nuclear power plant as photographed soon after the April 26, 1986, disaster by Andrey Illesh.



Super quick explanation of the what the Chernobyl nuclear disaster and reactor number 4 including it's timeline of events in the seconds and minutes and days following April 26th, 1986 and what radioactivity is






ПАРИКМАХЕ СКАЯ

Bioacumulation

Origin

Originates w/in zone

Originates outside of zone

Relationship w/ territory

Lives on/in ground

Lives above ground

Place w/in Food Web

Higher in web

Lower in web

Gulakov, Andrey Vladimirovich 2014. Rask, Martti 2012.

Practical:

EXPERIMENTALLY INDUCED ACUTE RADIATION SYNDROME IN EXPERIMENTAL ANIMAL

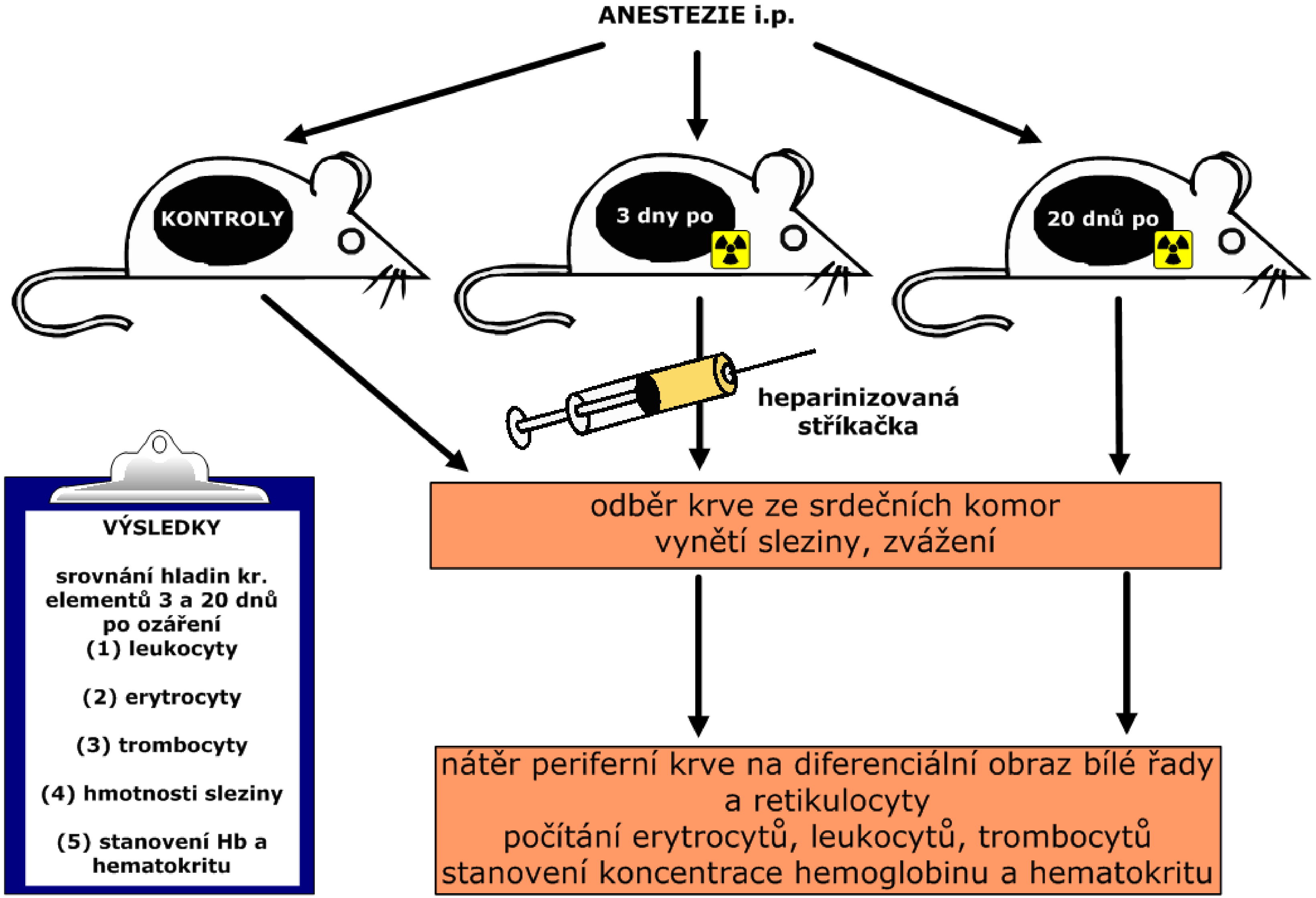
Aim of the practical

to document the deterministic nature of radiation effects on the practical examples

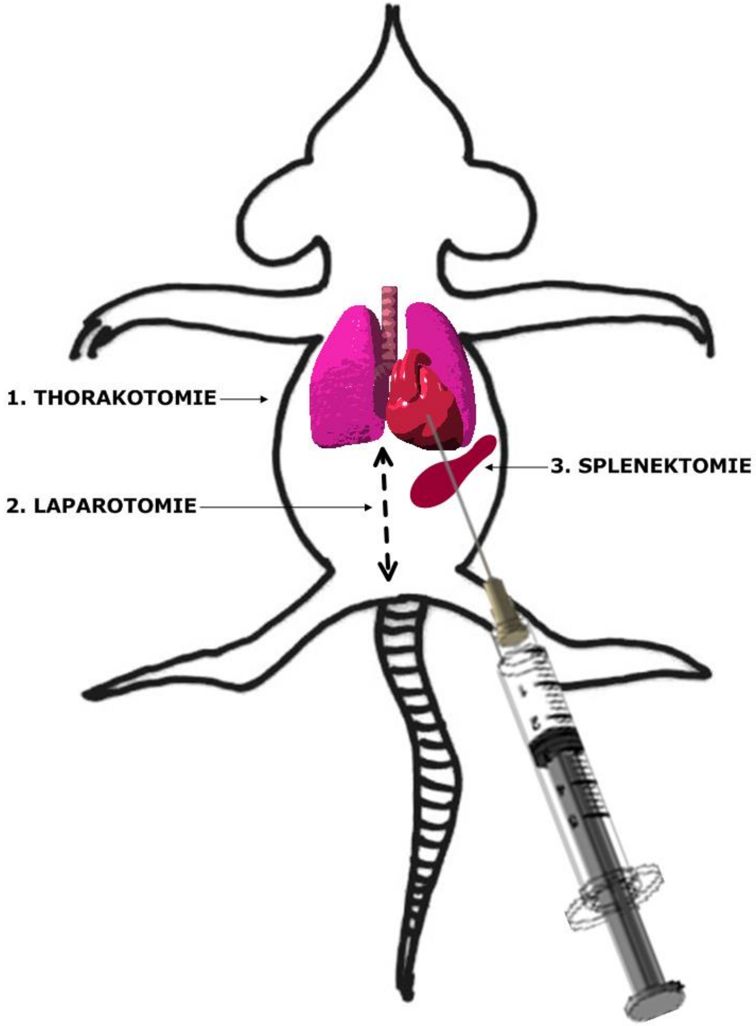
to monitor the dynamics of peripheral blood count changes as a result of the bone marrow

acute radiation syndrome is a model situation on which the principle of hematopoiesis regulation can be demonstrated

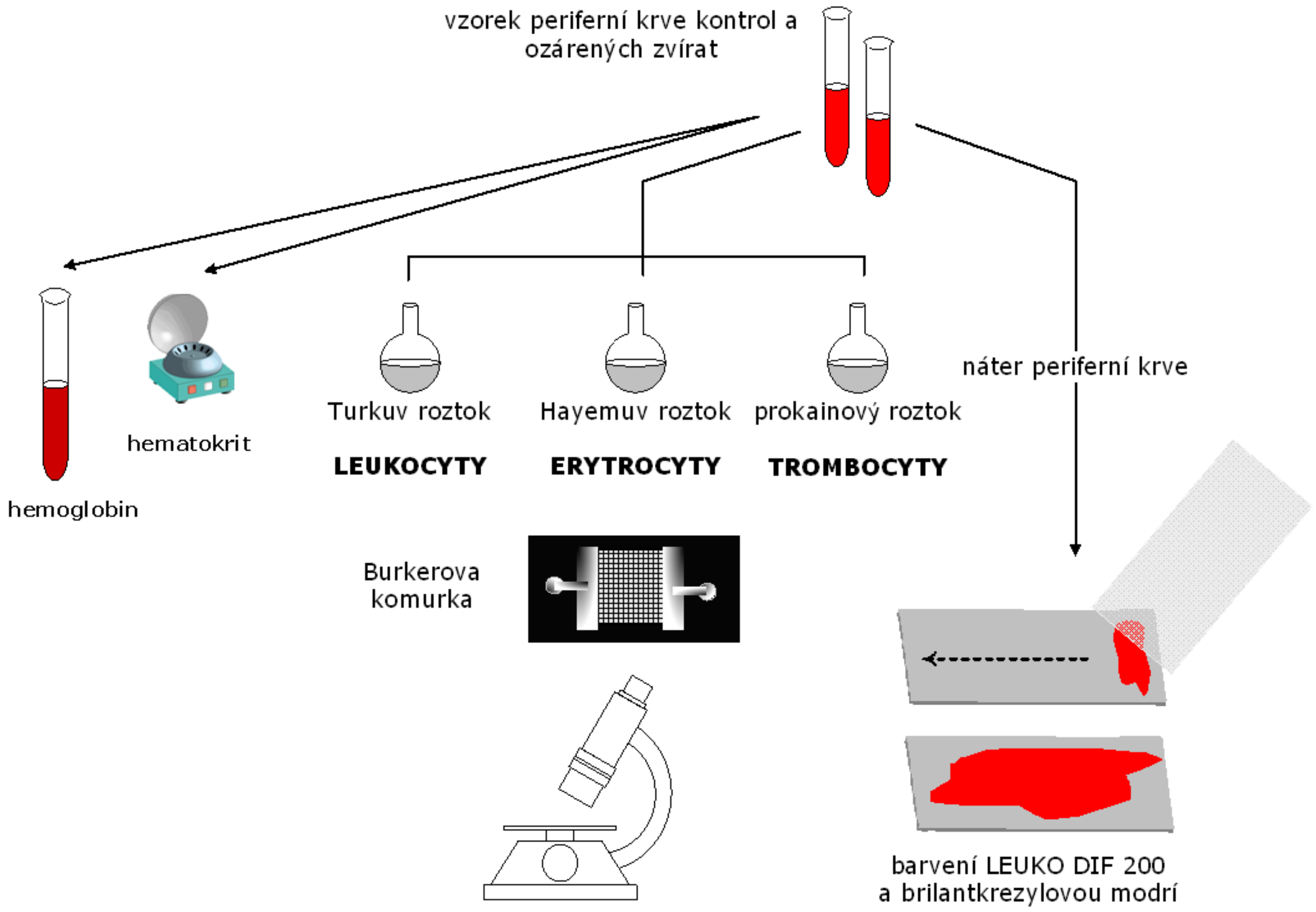
Practical experiment I - design



Practical experiment I – surgery technique

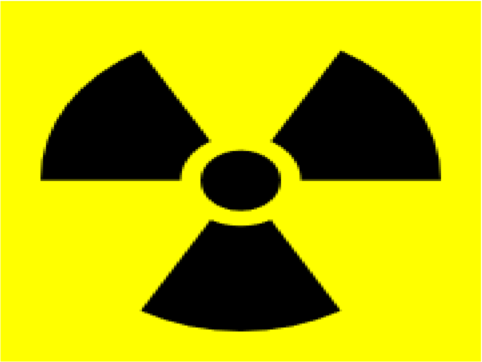


Practical experiment I - evaluation



Effects of ionizing radiation on haematopoietic tissue

Practical experiment II – evaluation of peripheral blood smears



Control questions

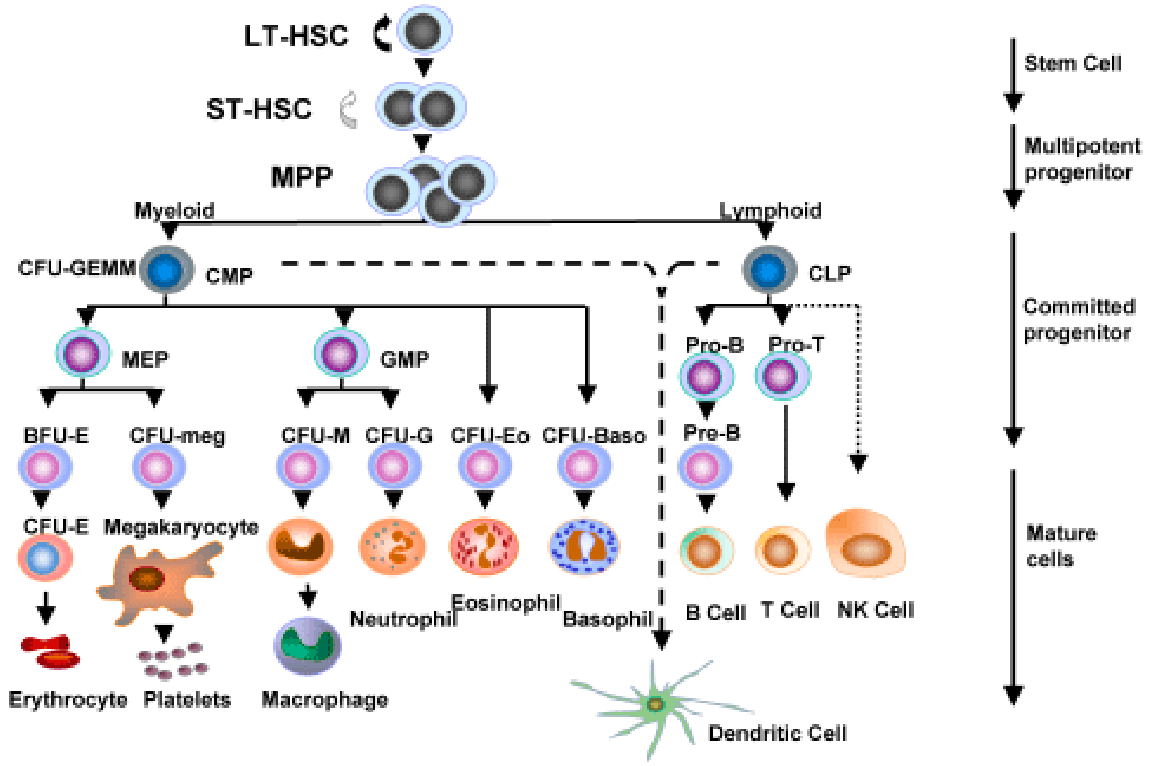
What is ionizing radiation?

What is radioactivity?

How are the biological effects of ionizing radiation mediated?

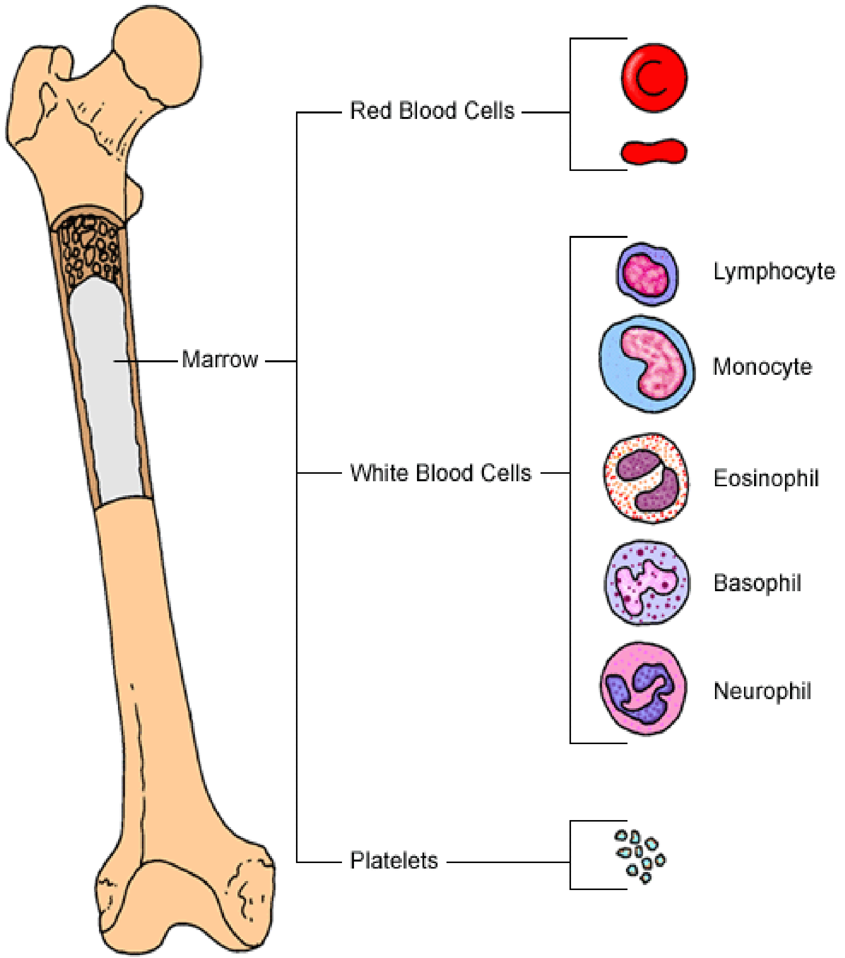
Types of biological effects of radiation + examples?

Hematopoeisis



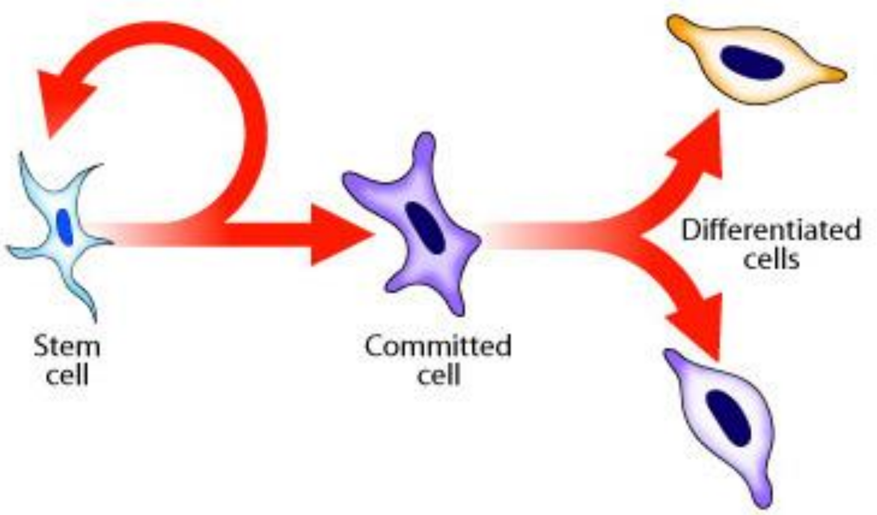
Hematopoiesis = bone marrow

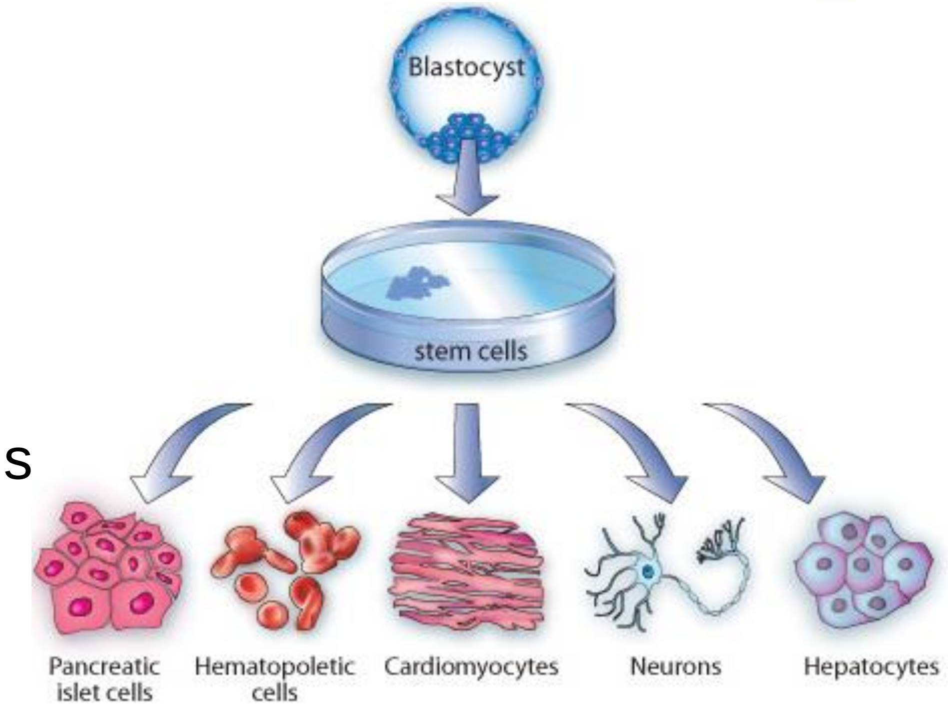
bone marrow
(1) haematopoietic cells.
(2) hematopoietic stroma - essential for normal production of blood cells.
fibroblasts, adipocytes, macrophages, T-lymphocytes
connective tissue, fat
own hematopoietic bb. - tribal bb.
pluripotent hematopoietic stem cell
differentiation into all series + self-renewal !!!
unclear phenotype - antigen classification CD34 +
in the pulp <0.01%
progenitor (determined) stem bb.
do not have long-term self-renewal ability
unclear phenotype - classification according to the ability to form colonies (CFU-E, CFU-M, CFU-G, CFU-Meg,…)
blood precursors bb.
clear phenotype (morphology, histochemistry)
in medulla ~ 90%
proerythroblast - basosile erythroblast - polychromatophilic erythroblast - orthochromic erythroblast - reticulocyte - erythrocyte
myeloblast - promyelocyte - myelocyte - metamyelocyte - granulophyte (rod)
promonocyte - monocyte
megakaryoblast - megakaryocyte
mature elements



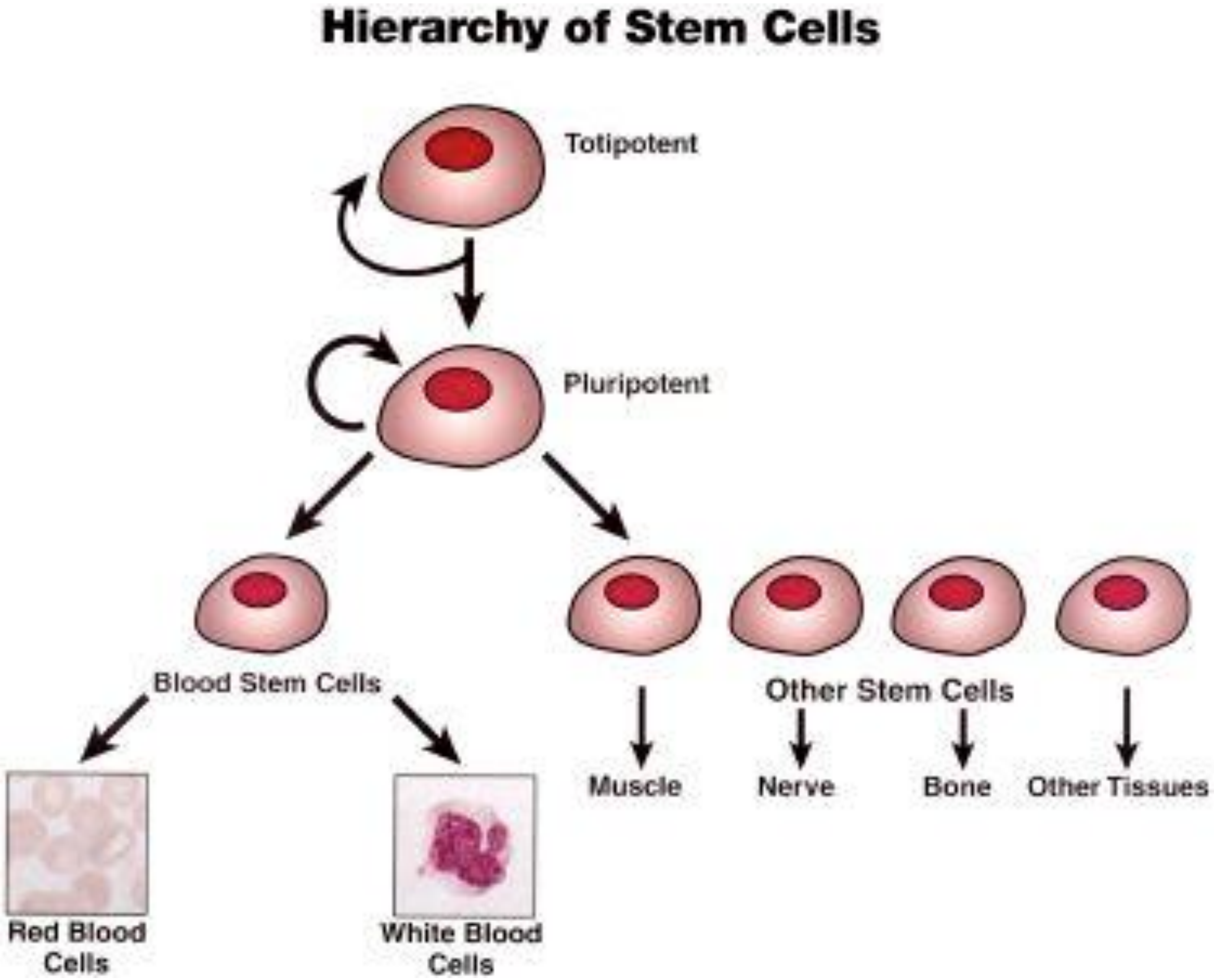
Stem cells

basic properties of KB are
self-renewal
division without differentiation (asymmetric)
production of specialized bb. (tissue regeneration)
KB types
mature KB (pluripotent)
adult, somatic
individual KBs give rise to a limited repertoire of bb.
eg haematopoietic KB, mesenchymal KB,…
early KB (toti- / omnipotent)
embryonic (blastocyst)
they give rise to all types of body cells
they are the only ones that do not need growth factors to stimulate division, in all others the cell cycle is initiated by mitogens





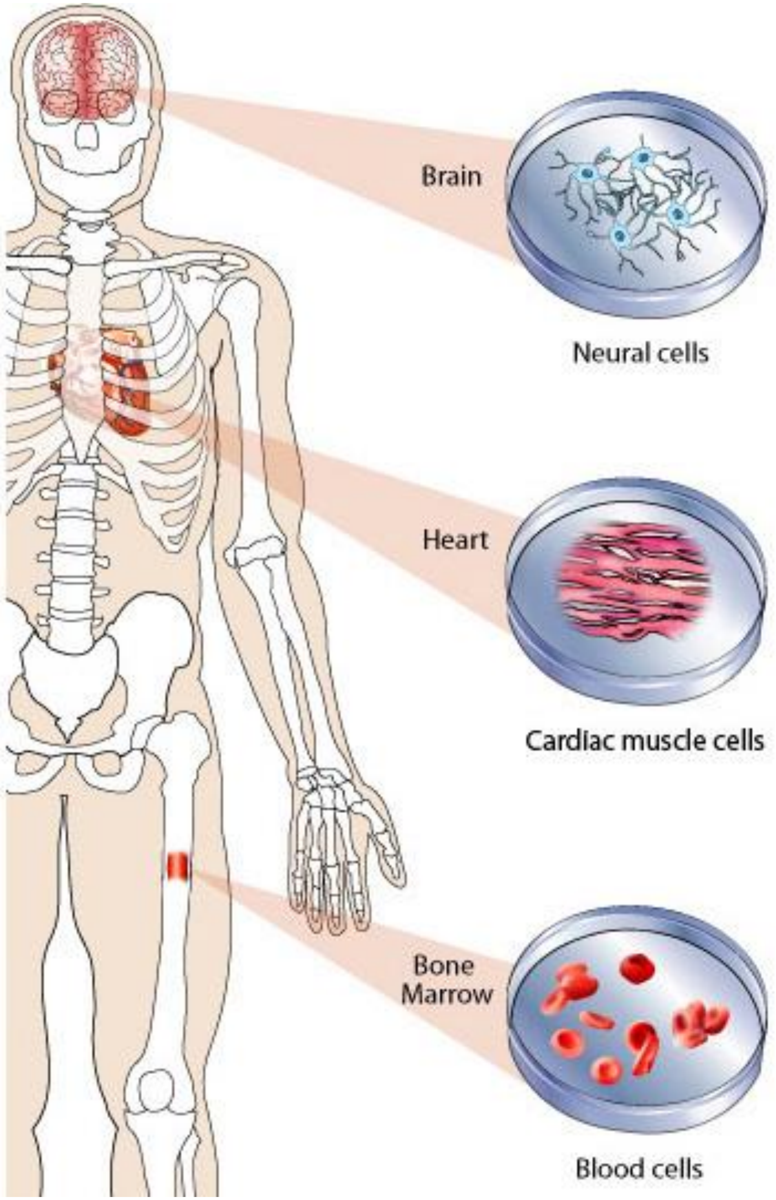
Stem cells



Hierarchy of Stem Cells
Totipotent
Pluripotent
Blood Stem Cells
Other Stem Cells
Red Blood Cells
White Blood Cells
Muscle
Nerve
Bone
Other Tissues

Somatic stem cells

located in most body tissues as a source of cells for constant self-renewal and replacement
they are pluripotent
give rise to all bb. of a specific type of tissue, but not another (this is only the ability of embryonic KB)
however, it appears that some universality is possible



Regulatory factors

interplay of autocrine, paracrine and endocrine factors

endocrine

erythropoietin (kidney)

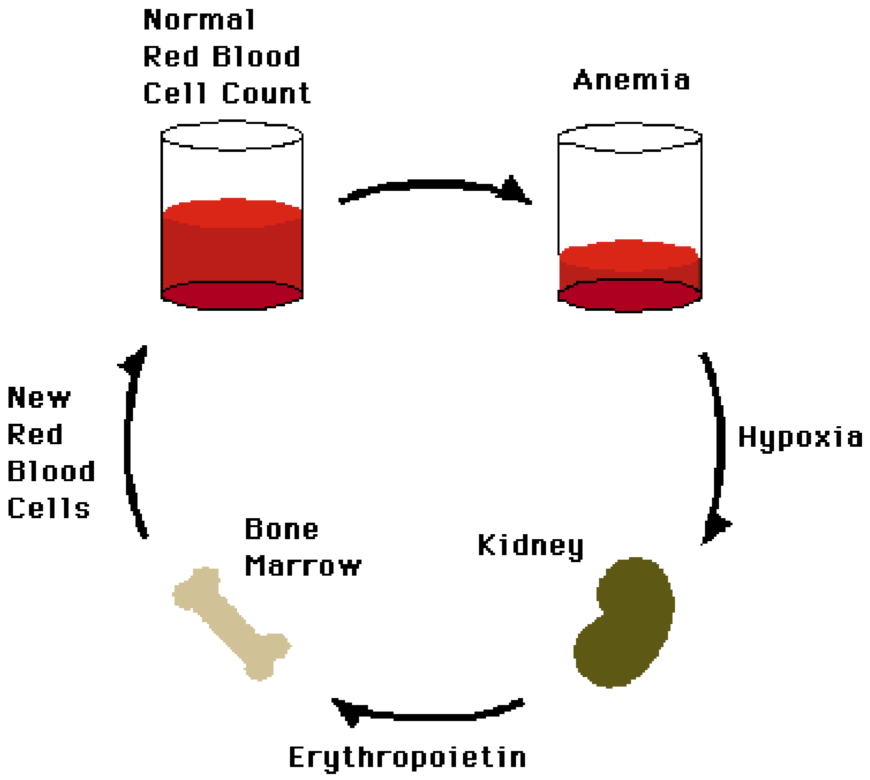
thrombopoietin (liver)

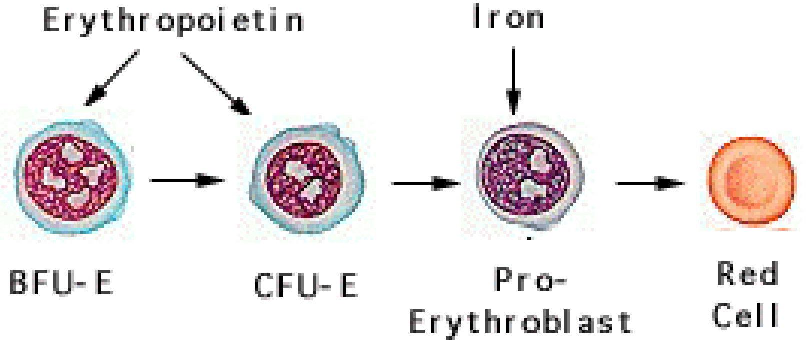
cytokines

para- / autocrine

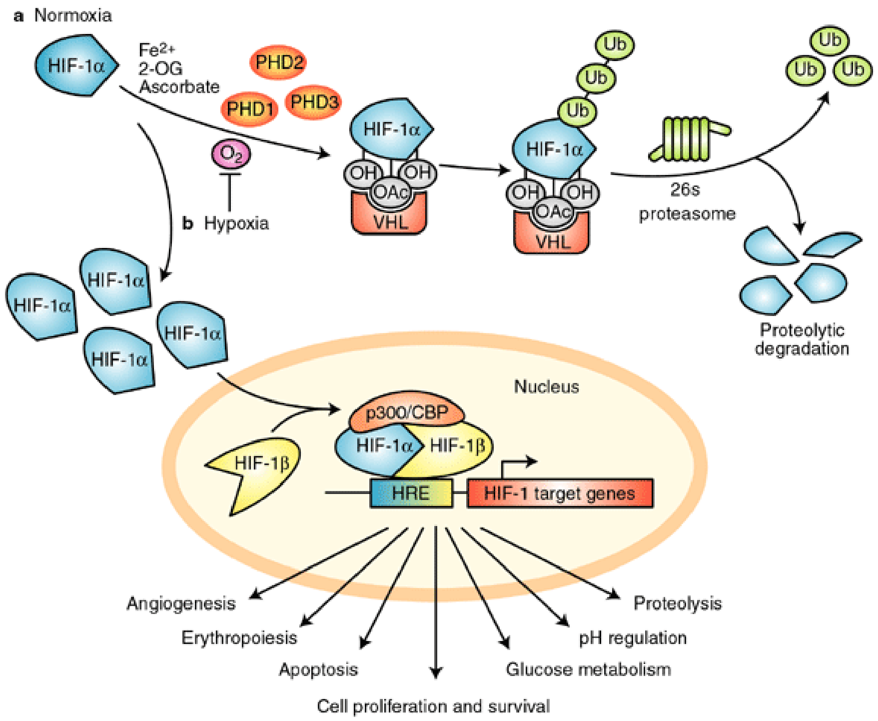
hematopoietic growth factors (cytokines)

produced by stromal cells, eg CSFs (colony-stimulating factors)





Erythropoetin (EPO)



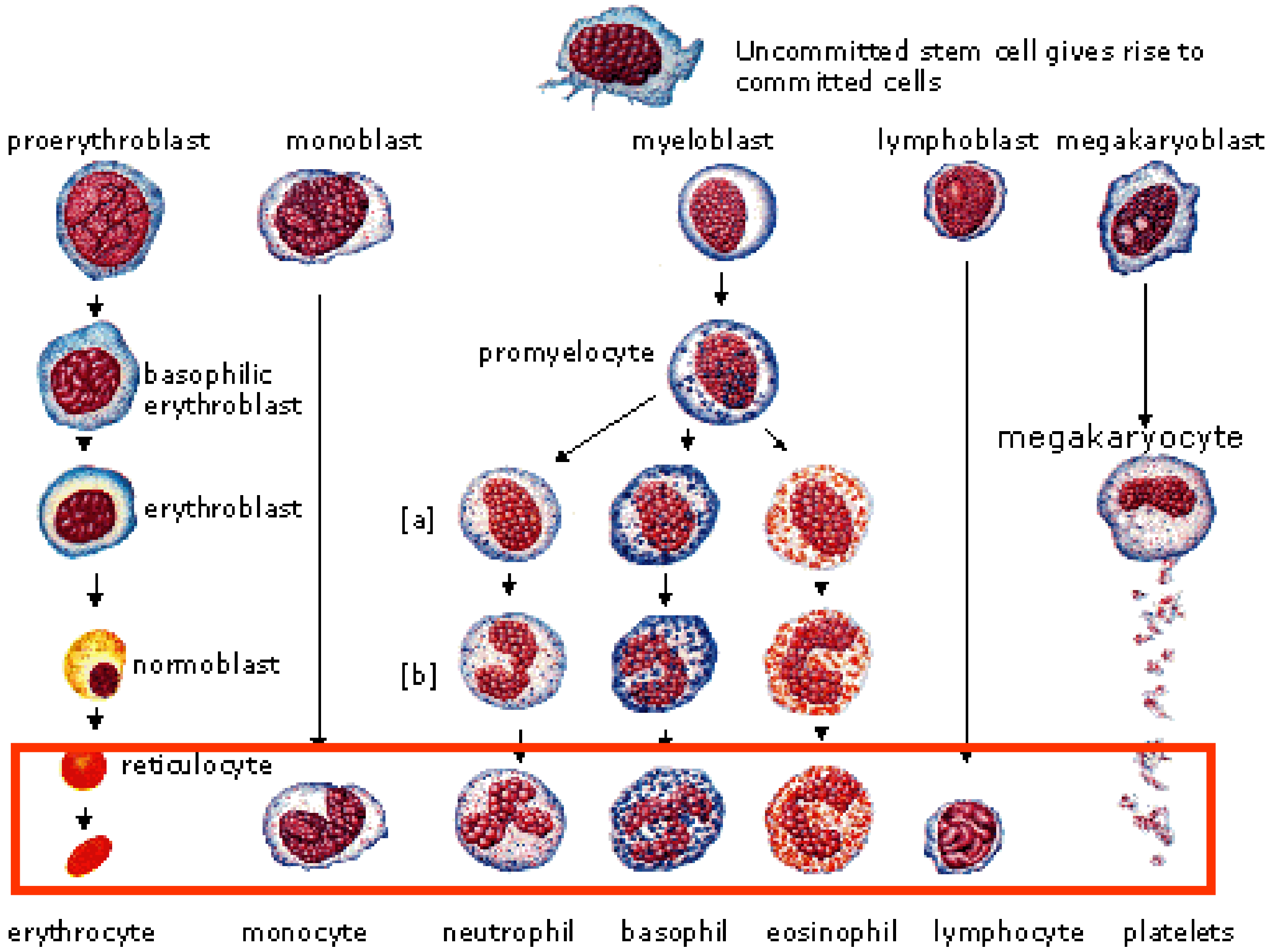
HIF-1α regulation by proline hydroxylation

90% oxygen in the body is used for ox. phosphorylation - ATP production
oxygen is relatively insoluble in water - Hb allows 100- more oxygen to be transported by blood than would be possible only in physically dissolved form
EPO is hl. regulator conc. Hb and hence oxygen availability
1893 - alpine environment leads to increase in Hb in humans – adaptation to hypoxia!
1950 - A humoral factor produced by the kidneys stimulating erythropoiesis
bilateral nephrectomy in rats led to anemia
1977 purification of EPO from the urine of a patient with aplastic anemia
1983 cloning of EPO gene - recombinant EPO production (epoetin)
long-term treatment of renal failure and some anemia
EPO production - peritubular fibroblasts of kidney (deep in cortex and outer cortex)
why kidney?
phylogenesis - in lower organisms, the kidney is a haemopoietic organ
more sensitive sensing of the actual Hb content and hence oxygen (after separation of plasma and circulating elements) at glom. filtration

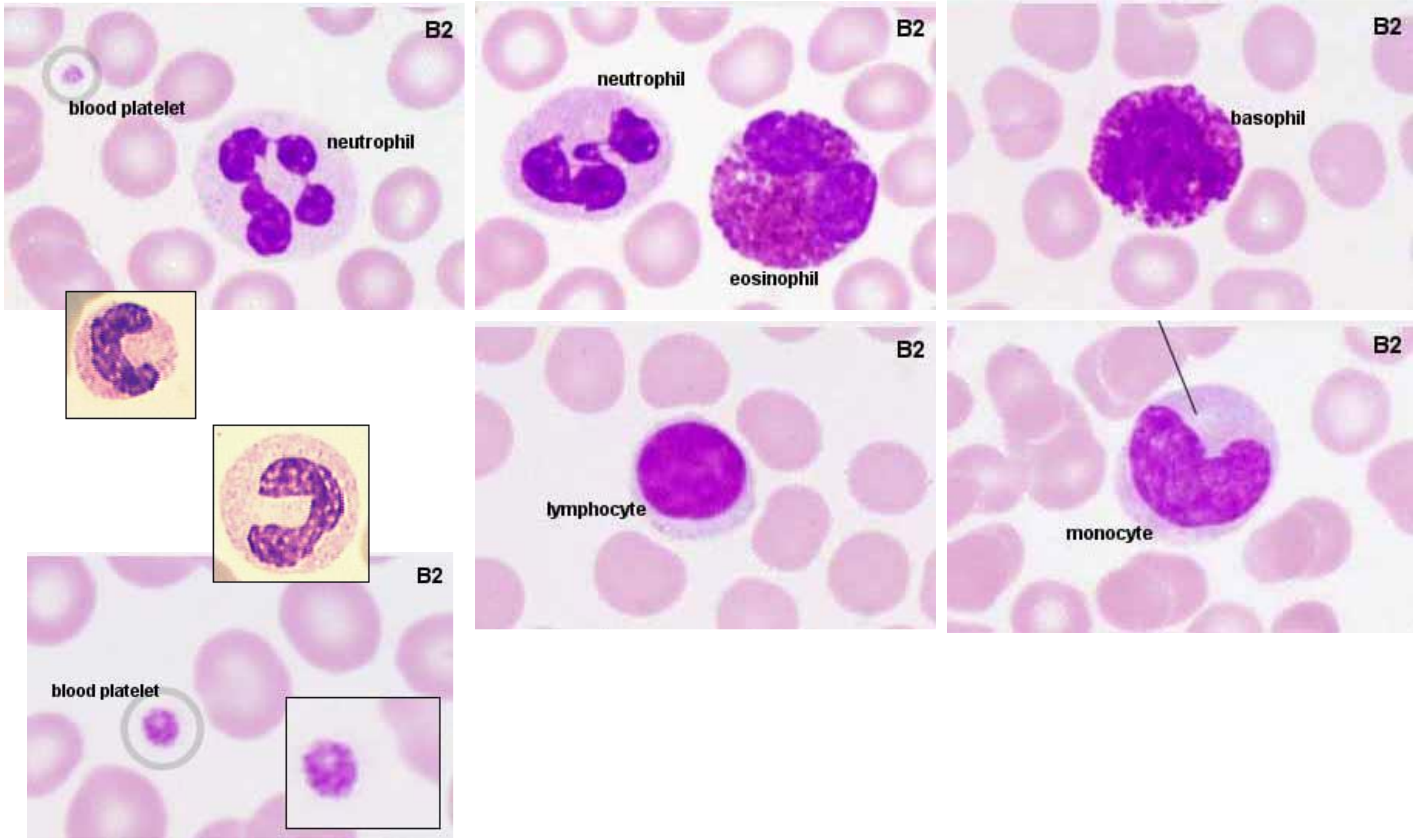
Blood count – reference values

	Muži	Ženy
Ery [RBC] ($\times 10^{12}$/l)	4.2 – 5.8	3.8 – 5.2
Leu [WBC] ($\times 10^{9}$/l)	5 – 10	
Tromb ($\times 10^{9}$/l)	150 - 400	
hematocrit (%)	0.38 - 0.49	0.35 – 0.46
hemoglobin (g/l)	135 – 175	120 - 168
Mean volume Ery [MCV] (fl)	80 - 95	80 - 95
Average Hb content in ERY [MCH] (pg) *MCH = Hb × 10/RBC*	27 - 32	27 - 32
Average concentration of Hb [MCHC] *MCHC = Hb × 100/hematocrit*	0.32 – 0.37	0.32 – 0.37
Red Cell Distribution Width [RDW] (%)	11 - 15	

Peripheral blood cells



Differential white blood count



Practical experiment II