From Erythroblasts to Mature Red Blood Cells

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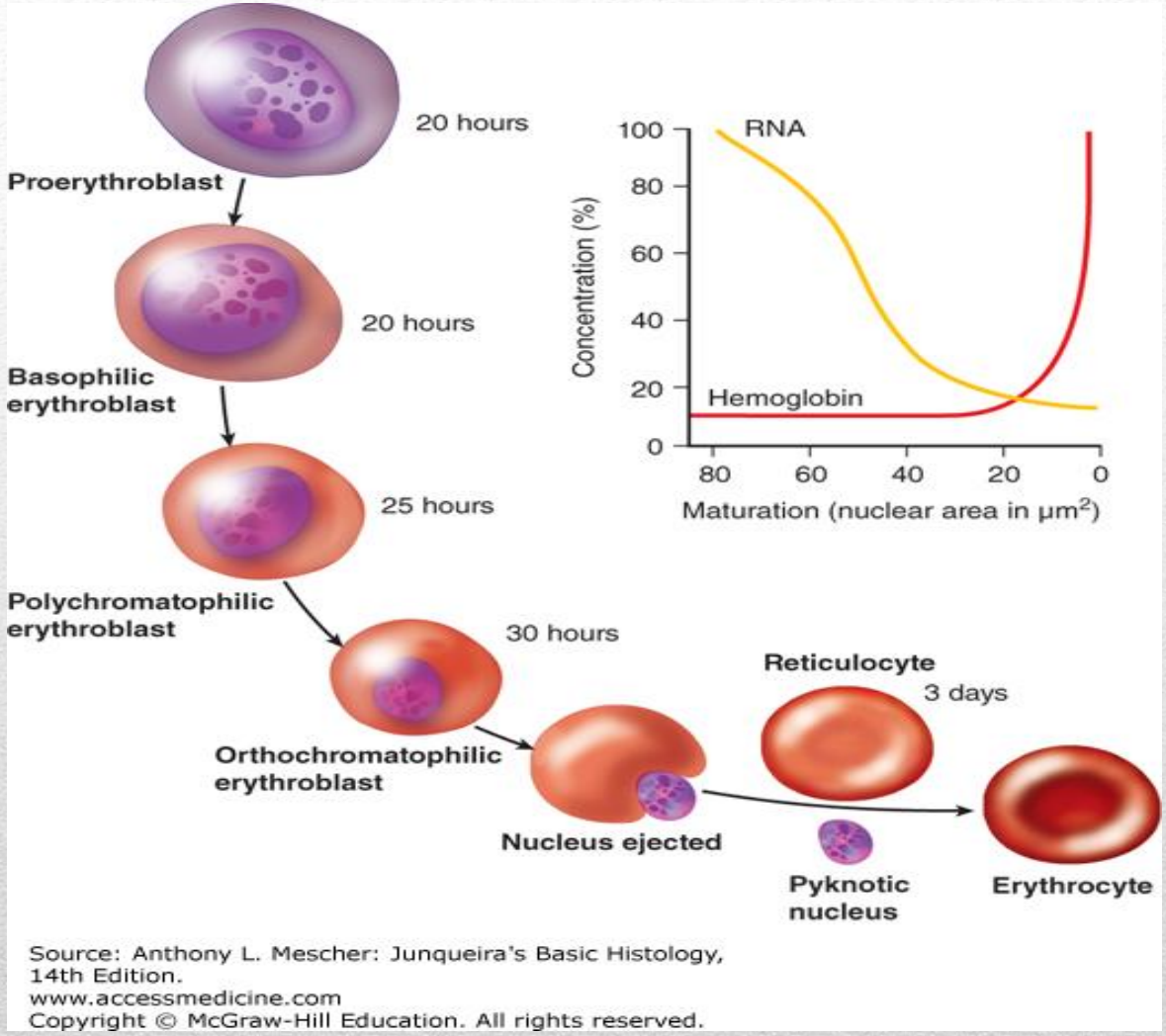
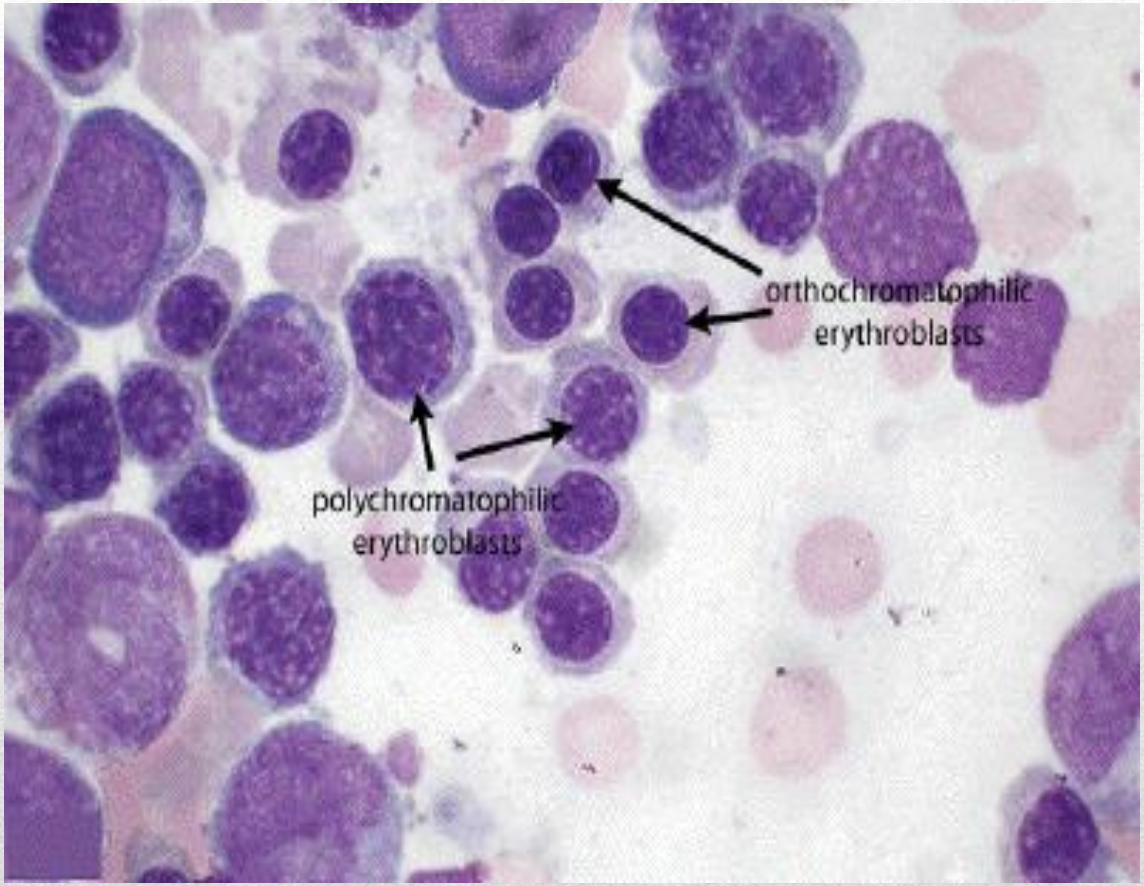
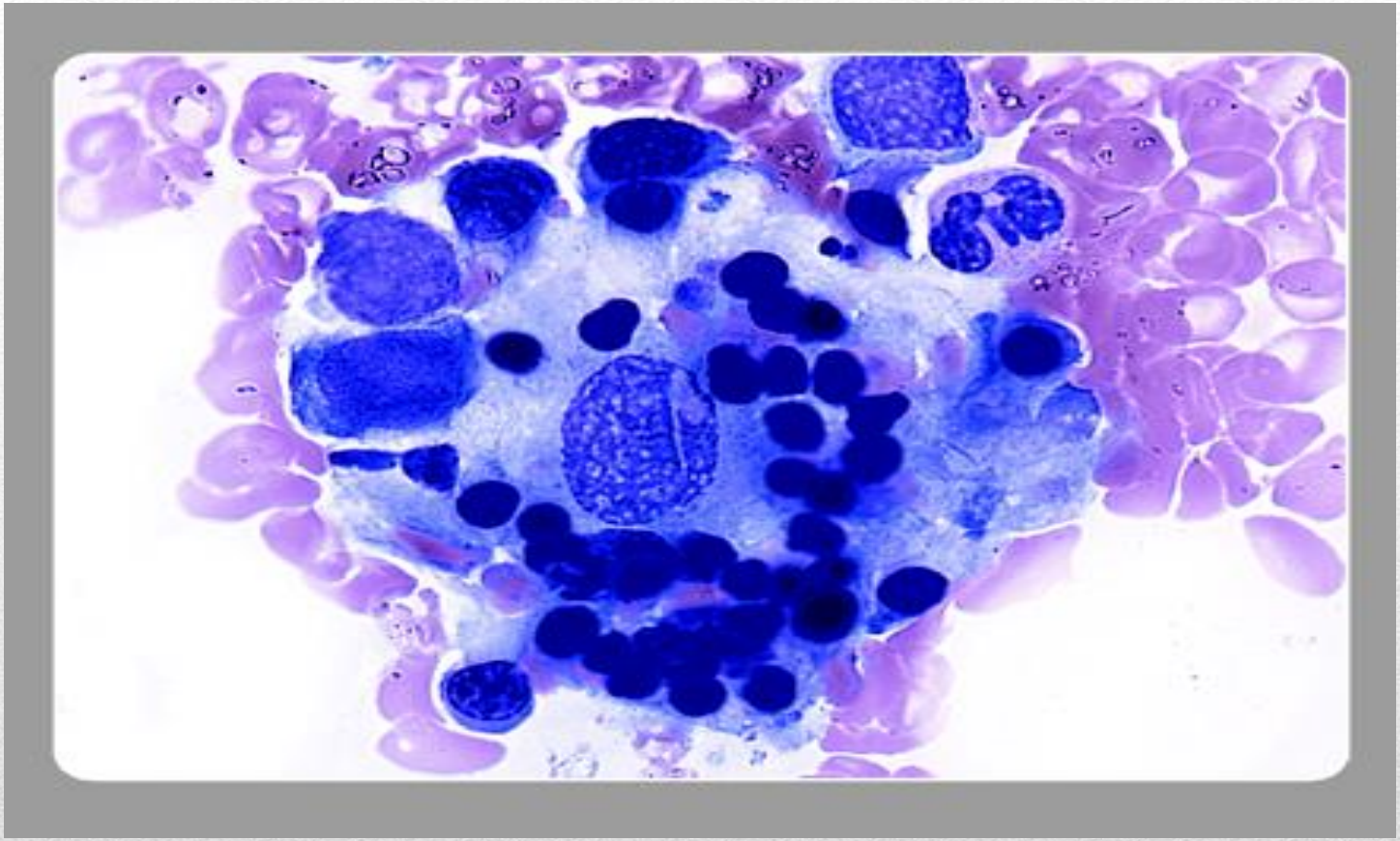
Mature red blood cells (RBCs) result from a finely regulated process called erythropoiesis that produces 2 million RBCs every second in healthy human adults. The standard model of erythropoiesis starts with hematopoietic stem cells (HSCs) in the bone marrow (BM), giving rise to multipotent progenitors that go on to erythroid-committed precursors to mature RBC. Maturation from erythroid-committed precursors is called terminal erythropoiesis and occurs in the BM within erythroblastic islands, which consist of a central macrophage surrounded by erythroblasts, and ends in the blood stream where reticulocytes complete their maturation within 1–2 days. During this phase, proerythroblasts (Pro-E) undergo morphological changes, such as cell size reduction and chromatin condensation, produce specific proteins, such as hemoglobin, and exhibit a reduced proliferative capacity to give rise to basophilic (Baso-E), polychromatophilic (Poly-E) and orthochromatophilic (Ortho-E) erythroblasts, successively. Even though several growth factors are known to regulate erythropoiesis, Epo is the main regulator of erythropoiesis driving RBC precursor proliferation and differentiation, preventing erythroblast apoptosis .The macrophage-erythroblast interaction in the BM is essential since macrophages facilitate proliferation and differentiation and provide iron to the erythroblasts.

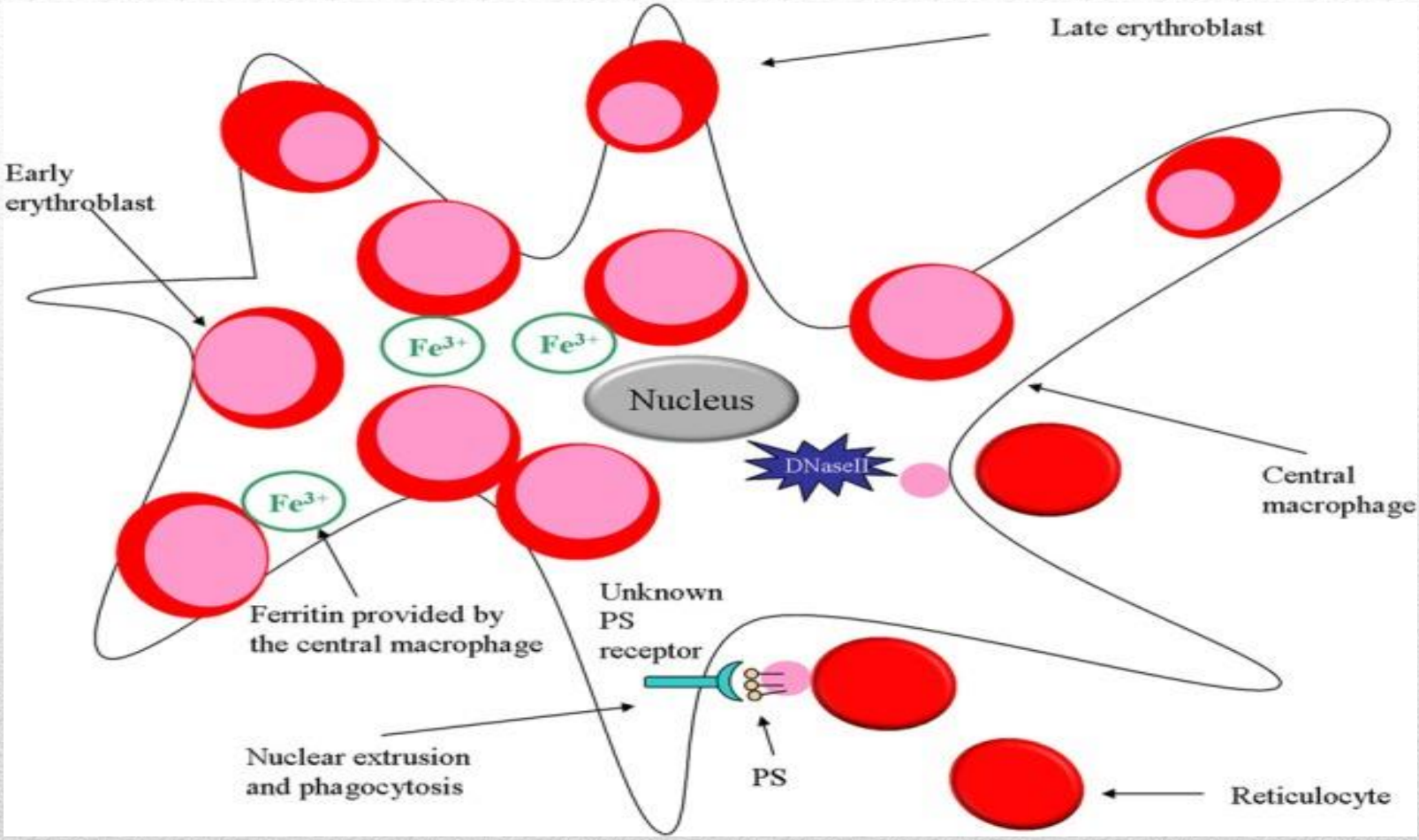


Source: Anthony L. Mescher: Junqueira's Basic Histology, 14th Edition.
www.accessmedicine.com


orthochromatophilic
erythroblasts
polychromatophilic
erythroblasts



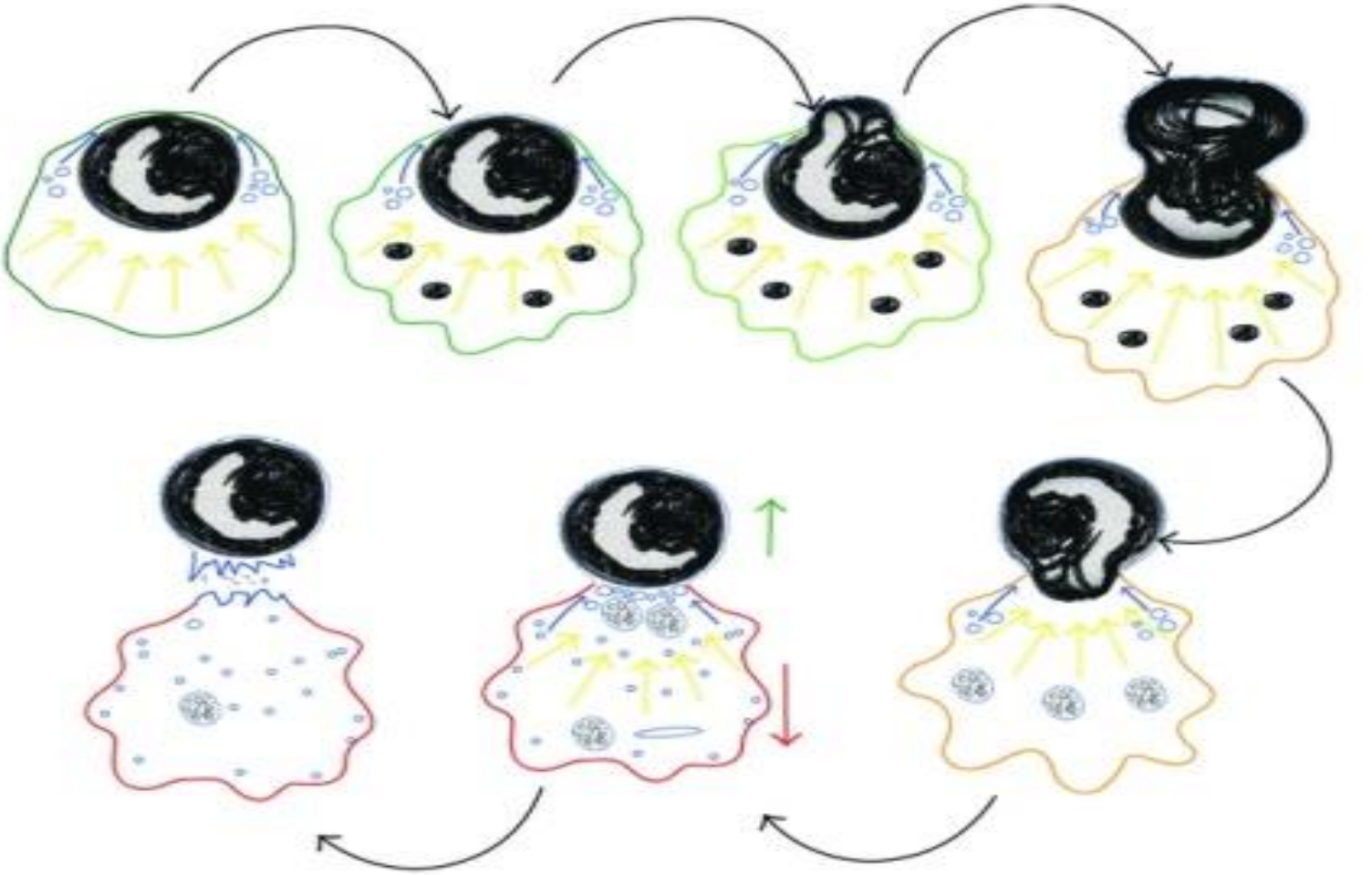
erythroblastic islands, which consist of a central macrophage surrounded by erythroblasts


Late erythroblast
Early erythroblast
Fe3+
Fe3+
Fe3+
Nucleus
DNaseII
Central macrophage
Ferritin provided by the central macrophage
Unknown PS receptor
Nuclear extrusion and phagocytosis
PS
Reticulocyte

- At the end of the terminal maturation, mammalian erythroblasts expel their nuclei and lose all their organelles, such as the Golgi apparatus, endoplasmic reticulum (ER), mitochondria and ribosomes. After expelling its nucleus, the reticulocyte maturation continues, losing 20–30% of the cell surface and eliminating any remaining membrane-bound cytosolic organelles through an autophagy/exosome-combined pathway.

- The most spectacular aspect of mammalian erythropoiesis is the generation of enucleated cells. Enucleation occurs in orthochromatic erythroblasts producing two kinds of cells, the reticulocyte and the pyrenocyte [the nucleus surrounded by a tiny layer of cytoplasm and the plasma membrane (PM)]. Pyrenocytes are rapidly eliminated by the macrophages of the erythroblastic island, where phosphatidylserine exposure acts as an “eat me” signal. Among the changes occurring during terminal differentiation, cell cycle arrest, chromatin and nuclear condensation and nuclear polarization are important for enucleation. In addition, nucleus expulsion is believed to be dependent on adhesion protein reorganization across the PM and macrophage interactions. The transcription factor KFL1 is required for enucleation. regulating the expression of cell cycle proteins, deacetylases, caspases, and nuclear membrane proteins.

- Nuclear and chromatin condensation is essential for enucleation and is dependent on the acetylation status of histones H3 and H4 under the control of histone acetyl transferases (HATs) and histone deacetylases (HDACs). Accordingly, Gcn5, an HAT protein, is down-regulated, and H3K9 and H4K5 histone acetylation decreases during mouse fetal erythropoiesis. In addition, Gcn5 is up regulated by c-Myc, which is known to decrease during the late phase of the erythropoiesis . With the same model, the role of HDAC2 protein was shown to be essential not only for chromatin condensation but also for the formation of the contractile actin ring (CAR), which is involved in nuclear pyknosis . Moreover, it was recently shown that major histones are released through a nuclear opening that is induced by caspase 3 activity-dependent lamin B cleavage and chromatin condensation.
- Cytoskeletal elements play an important role in erythroblast enucleation. actin filaments (F-actin) condensate behind the extruding nucleus to form the CAR.



- **Nucleated RBC**

Nucleated RBC are usually not seen in the blood of healthy mammals (low numbers may be seen in dogs, particularly Dachshunds and Schnauzers, and camelids, but are rarely normally seen in other species. nucleated RBCs are most often noted in dogs, cats and camelids in the context of strongly regenerative anemia. They can also be observed in llamas with regenerative anemias. They are seen less often in this setting in cattle and rarely seen in horses, even those with a strongly regenerative anemia. Note, that even though nRBCs can be seen in the blood of some animals with a regenerative anemia, alone (without polychromasia or macrocytes in equidae). They can also be seen in other conditions, including lead poisoning, altered or abnormal splenic function, or bone marrow injury (suspected mechanism in heat stroke). . It has long been postulated that a normoblastosis in blood of animals with a regenerative anemia is part of the regenerative response or an expected response to an anemia, however this is not true. In reality, we do not know why we see nRBCs in circulation in some animals with a regenerative anemia. It may be due to the type, severity or cause of anemia, since more nRBCs are seen, for instance, in dogs with a hemolytic anemia (such as due to regenerative variants of immune-mediated hemolytic) than dogs with experimental blood loss. It is possible that nRBCs in blood in animals with regenerative anemia reflects an altered bone marrow microenvironment (e.g. defective pitting function of marrow macrophages, which control, to a certain degree, the enucleation of developing red blood cells or an altered sinusoidal endothelial barrier) or altered splenic function (where presumably circulating nRBC are removed once they are expelled from marrow).

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- Why do mammalian red blood cells lack a nucleus?

1:The absence of a nucleus is an adaptation of the red blood cell for its role. It allows the red blood cell to contain more hemoglobin and, therefore, carry more oxygen molecules. It also allows the cell to have its distinctive bi-concave shape which aids diffusion. This shape would not be possible if the cell had a nucleus in the way. Because of the advantages it gives, it is easy to see why evolution would cause this to occur. However, since little is known about the genes the control enucleation, it is still not a fully understood process

2: it lightened the cardiac workload. Each extruded RBC nuclei is approximately 40 picograms. A normal healthy adult individual would produce about 2 million RBC per second. That would be 0.08 milligrams of weight per second are required to be removed.

3: reduce risk of hemolysis when transversing through the microvasculature. In other words, mature RBC can move along tiny blood capillaries by changing their biconcave shape,so that they will not rupture (and die).

- Why do camels have nucleated red blood cells?

1 : What's different from other mammals is not that camels have nucleated red blood cells but that the red blood cells have a different shape. Like most other animals, the red blood cells of camels lose their nuclei as they mature. The camel's red blood cells are more ovoid rather than round, which allows them to expand significantly when a camel drinks water. The oval shape also allows the red blood cells to fit more easily through capillaries when the camel is not consuming water, as there blood is very thick when dehydrated.

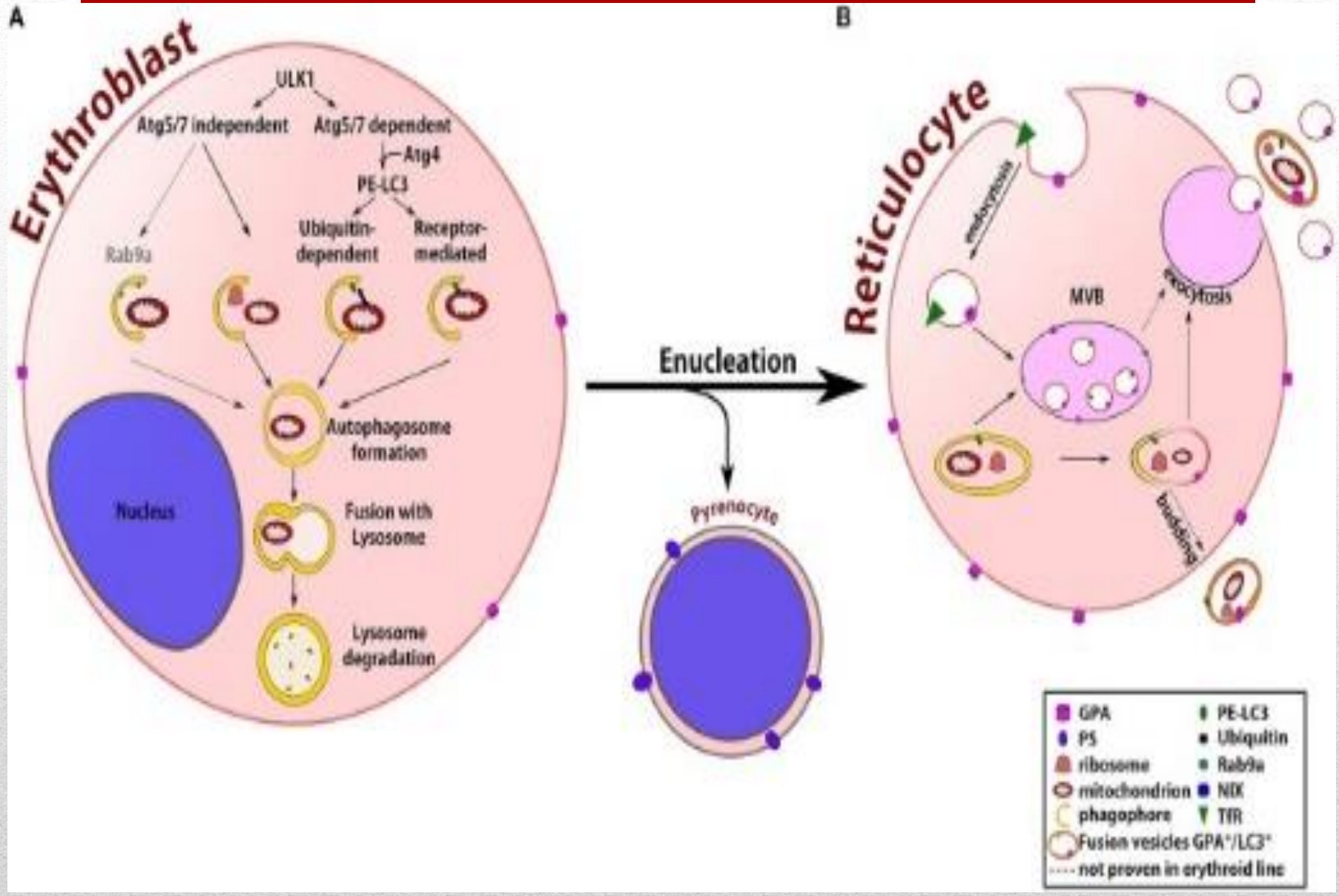
2: Camel RBCs are anucleate . The dark structure seen in microscopic images is not nuclei but a network of microtubules called the marginal bands. Marginal bands cause these RBCs to adopt an ellipsoid shape. The unique shape of these RBCs possibly allows them to survive osmotic stress and is probably advantageous to a camel under extreme dehydration.

- **Why do chickens have a nucleus in their red blood cells?**

We have extremely narrow blood vessels, called capillaries, in our tissues and organs where the blood supplies oxygen and nutrients. If our red blood cells had a nucleus, they wouldn’t be able to squeeze through. Birds have slightly wider blood vessels.

Or maybe..

Getting rid of the nucleus means that the whole of the cell is cytoplasm and can be filled up with haemaglobin to carry more oxygen per cell. So our hearts don’t have to work so hard. Have you ever held a bird and felt how fast their hearts beat.


A
Erythroblast
ULK1
Atg5/7 independent
Atg5/7 dependent
Atg4
PE-LC3
Rab9a
Ubiquitin-dependent
Receptor-mediated
Autophagosome formation
Nucleus
Fusion with Lysosome
Lysosome degradation
Enucleation
pyrenocyte
B
Reticulocyte
endocytosis
MVB
exocytosis
budding
GPA
PS
ribosome
mitochondrion
phagophore
Fusion vesicles GPA+/LC3+
not proven in erythroid line
PE-LC3
Ubiquitin
Rab9a
NIX
TfR

- Mitochondrial clearance Ribosomes and other organelles

The main mechanism for mitochondrial clearance is mitophagy, a selective type of autophagy that allows the degradation of damaged mitochondria. The importance of this process is highlighted by knowing that an impairment in mitochondrial function triggers an increase in reactive oxygen species production, which can in turn cause damage to cellular components (proteins, nucleic acid, and lipids) and trigger cell death .
During regular autophagy processes, stress or nutrient deprivation activates APM-activated protein kinase (AMPK), triggering two ubiquitin-dependent pathways. One of these allows the assembly of the phagophore and involves several autophagy-related proteins (Atg), such as Atg5 and Atg7. The other aims to activate and lipidate LC3 (MAPLC3, microtubule-associated protein 1 light channel 3) by Atg4, a redox regulated protein. Atg4 and Atg7 cooperate to conjugate LC3 onto phosphatidylethanolamine in the lipid bilayer of the membrane originated from the ER-mitochondria contact site. The elongated phagophore is then recruited to engulf targets via adaptor proteins, containing an LC3-interacting region (LIR) that forms a double-membrane autophagosome, which will fuse with a lysosome, initiating the degradation of the autophagosome components. Upon mitochondria damage or depolarization, the mitochondrial membrane proteins are exposed and act as a beacon to recruit the phagophore membranes.

- Refrences

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