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FER–VITAMINSB9ANDB12–HEMOGLOBINOPATHIES

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1. Iron and its pathology

The body contains 4 to 5 g of iron in the form of heme compounds, that is to say comprising heme (hemoglobin, myoglobin, cytochromes, peroxidases and catalases), or non-heme compounds: siderophilin or transferrin, ferritin , hemosiderin. These latter compounds represent the transport and storage forms.

1.1. Physiology

1.1.1. Losses, intakes, absorption and transport of Iron

Daily iron losses are normally extremely low, of the order of 1 mg per day (through sweat, cellular desquamation, skin appendages, urine and feces). In menstruating women, iron losses are higher (2 to 3 mg per day on average). To compensate for losses, the body depends on iron provided by food. The iron richness of foods varies greatly, the richest being lentils, meat, spinach, chocolate, dried fruits and red wine. The normal diet of an adult living in a developed country includes 10 to 25 mg of iron each day. However,**only 5 to 10% of dietary iron is absorbed, or approximately 1 mg/day, which compensates for daily losses**.

L'**iron absorption**takes place in the intestine,**especially in the duodenum**and essentially**in the form of ferrous iron**. To be absorbed, iron must be released from the food proteins that contain it. This explains that in the event of a change in gastric secretion, a change in iron absorption can be observed. In the duodenum and more incidentally the jejunum, iron passes from the intestinal pole to the blood pole of the intestinal cell. Ferric iron is first reduced to ferrous iron which is then**transported into the cytoplasm of the enterocyte by the transporter DMT-1**(Divalent metal carrier-1),**then released to the blood pole of the cell by another transporter, ferroportin under the control of hepcidin**, a regulatory protein (figure 1). From there, ferrous iron is reoxidized to ferric iron by a ferroxidase called hephaestin.**In plasma, serum iron is not free, but fixed**on a protein,**transferrin** who will transport it to its place of use. Two ferric iron atoms can be transported by one transferrin molecule. Transferrin is normally only saturated to one third of its capacity.

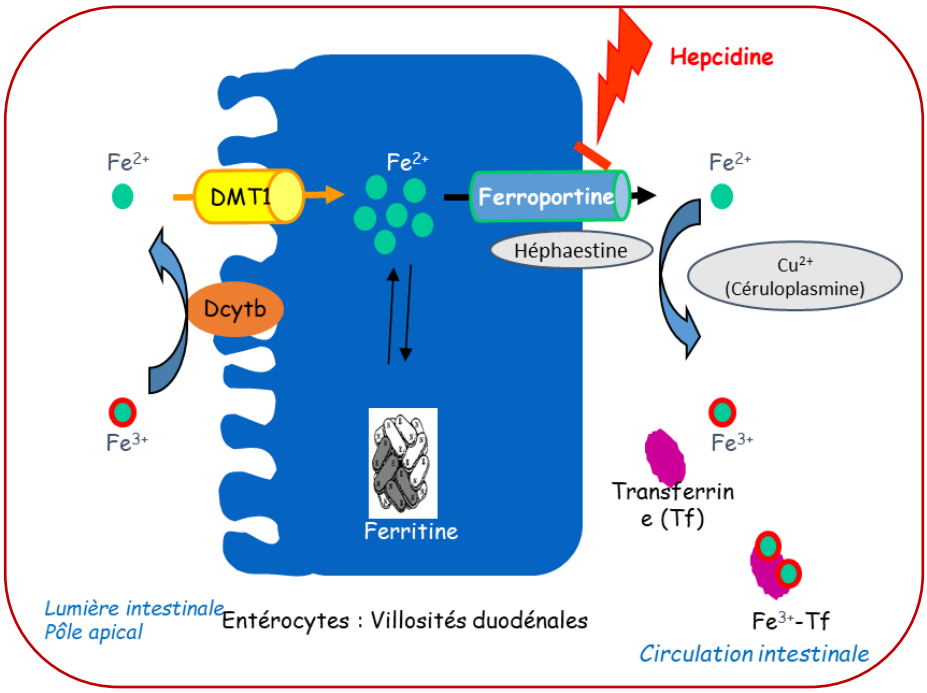

Figure 1. Iron absorption by the enterocyte

1.1.2. Iron and erythropoiesis

Quantitatively, the most important part of the body's iron is contained in RBCs. Each day, approximately 10 to 30 mg of iron is released during physiological hemolysis, and the same amount of iron is “recycled” into new red blood cells (Figure 2). There is thus a**almost closed circuit**iron from erythropoiesis, the vast majority of which is reused for erythropoiesis.

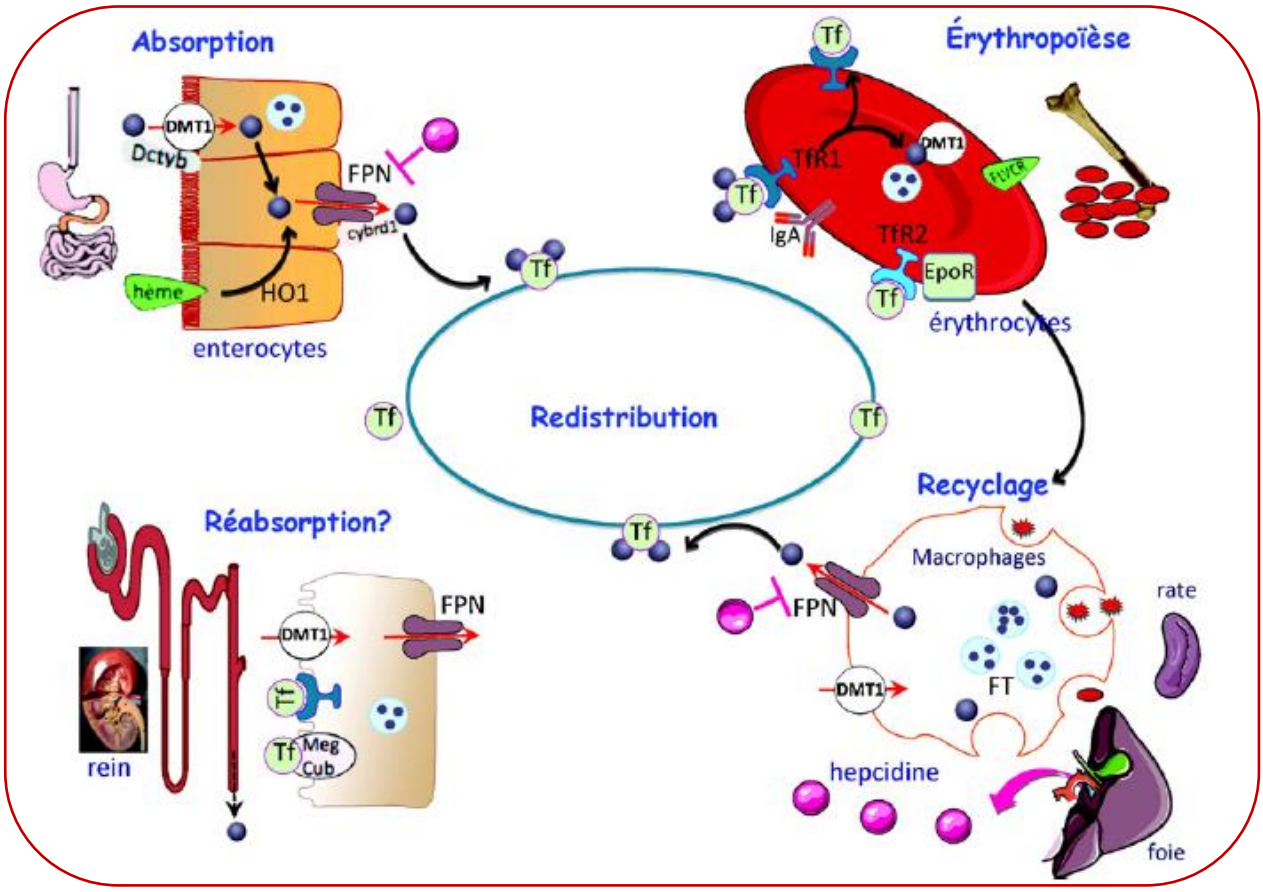

Figure 2. Iron metabolism and closed iron cycle

1.1.3. Iron reserves

Approximately 20% of the body's iron (0.6 to 1.2 g of iron) is stored in the**macrophages**and the **hepatocyte cells**. These reserves are essentially stored**in the form of ferritin**, a large molecule that can hold up to 4,000 atoms. Reserves are generally lower in women (≈600 mg) than in men (≈1,200 mg), which partly explains the greater frequency of iron deficiencies in women.

1.2. Iron deficiency

1.2.1. Pathophysiology

Iron being a major constituent of heme, any iron deficiency has an impact on erythropoiesis.
In a situation of iron deficiency, the body first draws on its reserves, gradually leading to a reduction in ferritinemia. In parallel, a compensatory mechanism, regulated by**hepcidin**, gets ready**to increase iron absorption at the level**from the apical pole**of the enterocyte and increase the production of transferrin**. Once the reserves are exhausted, we observe a decrease in the bioavailability of iron with a**decreased serum iron**. The reduction in bioavailable iron leads to a disorder of hemoglobin synthesis with a**microcytosis**(continuation of cell divisions as long as the target intra-erythrocyte iron concentration is not reached) and a**anemia**(rarefaction of hemoglobin in red blood cells) accompanied by a**drop in CCMH (hypochromia).**

1.2.2. Biological diagnosis

Iron deficiency is characterized by**hypochromic aregenerative and hyposideremic microcytic anemia**. We thus find a reduction in the hemoglobin level, the MCV and the CCMH as well as

than the level of reticulocytes which it is useless to measure because microcytic anemias are commonly of central origin and are regenerative.**Anemia is classically isolated**without anomaly of other lines **with the exception of frequent reactive thrombocytosis**the mechanism of which is not elucidated. We also find biochemical signs of iron deficiency with a**decreased serum iron**associated with a reaction increase in the total binding capacity (CTF), a decrease in the transferrin saturation coefficient (CST) and above all a**drop in ferritinemia**true reflection of iron reserves.

1.2.3. Clinical

Iron deficiency presents clinically with the usual signs of**anemic syndrome**in the foreground, namely**pallor, asthenia, tachycardia, dyspnea**. Of the**more specific signs of iron deficiency** can be found with in particular: cracks in the corners of the lip, changes in the nails, glossitis (poorly papillated or depapilated tongue), a possible change in food taste (“Pica”) with an attraction to substances such as earth (geophagia). , plaster... Can be associated with **signs pointing towards etiology**as signs of digestive or gynecological hemorrhage.

1.2.4. Etiologies

Iron deficiency can result from multiple and varied etiologies. In developing countries the main cause is**deficiency of intake**while in developed countries, the**chronic bleeding**is the most common cause whose origin is**most often gynecological in women and digestive in men**. There are other causes such as**intake deficiency**(infant, specific diets low in iron),**repeated pregnancies**, there**digestive malabsorption**(e.g. celiac disease).

1.2.5. Therapeutic approach

The standard treatment consists of**martial supplementation*per os***. It is imperative that he be associated with a **etiological treatment**.

1.3. Inflammatory anemia

1.3.1. Pathophysiology

Inflammatory anemia is complex and combines different mechanisms which are partly linked to an increase in pro-inflammatory cytokines (IL1, IL6, TNFα). There is a**direct inhibitory effect of these cytokines on erythropoiesis**directly contributing to anemia. Furthermore, there is also a phenomenon of**macrophage sequestration of iron under the effect of hepcidin**. The iron is thus sequestered **in its reserve form, ferritin**, and therefore not usable for erythropoiesis.

1.3.2. Biological diagnosis

The biological assessment will find stigmata very similar to iron deficiency with a**microcytic, aregenerative, hypochromic and hyposideremic anemia**. The NFS is therefore in all respects superimposable with reduction of hemoglobin, MCV and CCMH, and also of reticulocytes (which it is unnecessary to measure). It is the martial assessment which will make it possible to discriminate between the two etiologies. Inflammatory anemia will in fact be accompanied by**decrease in serum iron**,**but**unlike iron deficiency, the CTF will be reduced and the**very increased ferritinemia**, a reflection of iron sequestration. The dosage of inflammatory proteins (CRP, protein electrophoresis, fibrinogen) and possibly the sedimentation rate (ESR) will also help to confirm the diagnosis.

1.3.3. Clinical

This is the clinic associated with the etiology of the inflammatory syndrome.

1.3.4. Etiologies

All inflammatory syndromes(rheumatic diseases, systemic lupus erythematosus, febrile infections, cancers, Hodgkin's disease, etc.) lead to severe inflammatory anemia (<8 g/dL).

1.3.5. Therapeutic approach

There is no iron deficiency in inflammatory syndrome. There is therefore no need to administer iron replacement therapy. The treatment consists of the sole treatment of the inflammatory syndrome causing the disorders.

2. Vitamins B9 and B12

2.1. Physiology of vitamin B9 and B12

Vitamins B9(or folate or folic acid)**and B12**(or hydroxycobalamin) are**essential for the synthesis** thymidilic acid, nucleotide base**DNA**. Their defect results in a defect in DNA replication and an elongation of the cell cycle of erythroblasts.
In the event of a vitamin B12 deficiency, the consequences on hematopoiesis are identical to those of a folic acid deficiency. In addition, vitamin B12 plays a role in the nervous system independent of its intervention in DNA synthesis.

2.1.1. Folic acid metabolism

Folic acid is a water-soluble vitamin, pteroylglutamic acid, which exists in nature in various forms, notably in**green vegetables, grains, liver and meats**.**The needs are** relatively important, **of the order of 50 µg per day**, but**covered as part of a balanced diet**.**Absorption**occurs all along the small intestine, but mainly**in the duodenum and jejunum**(like iron), in the form of monoglutamates,**by an active mechanism**. However, when a massive dose of folate is supplied through the diet, passive diffusion through the intestine occurs. This allows effective oral drug delivery, even in cases of moderate malabsorption. **Excretion is fecal and urinary**.**In plasma, folates are bound to proteins**, without specific transport protein. Their level is 5 to 15 ng/mL. **Reserves are distributed throughout all tissues**, but they are weak,**exhaustible in the event of a deficiency in intake in 2 weeks to 4 months**. Red blood cells contain significant amounts of folate in the form of pteroylpolyglutamate. The clinical exploration of folate metabolism essentially concerns**serum folate measurement**.

2.1.2. Vitamin B12 metabolism

Vitamin B12is present in different foods, including**meat, liver**, but very rare in plants.**Daily requirements are 1 to 3 µg**widely**covered by most plans**. In the stomach, the vitamin is separated from the proteins and combined with its own transporter, the**intrinsic factor**(FI) (figure 3). It is a glycoprotein secreted by parietal cells of the fundus and body of the stomach, which dimerizes by fixing vitamin B12 at a specific site and then ensures its transport to the ileum. **Absorption**takes place**in the terminal ileum**thanks to a**specific receptor**cells of the brush border of the mucosa.**Reserves**, basically**hepatic**, are considerable, **sufficient for 4 to 5 years on average**.**Excretion is urinary and biliary**. It exists**an enterohepatic cycle of vitamin B12**.**In the circulation, vitamin B12 is fixed on**specific carriers:**transcobalamins**.

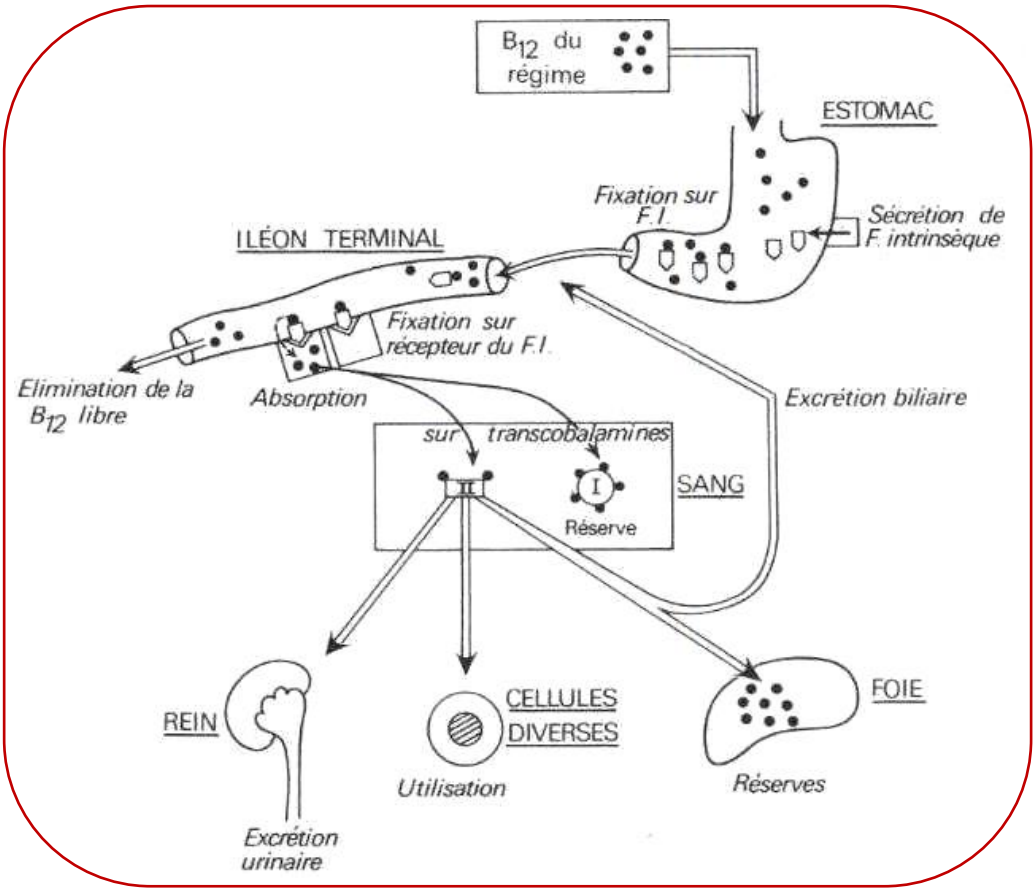

Figure 3. Vitamin B12 metabolism

2.2. Pathophysiology

Any disorder in the metabolism of vitamins B9 and/or B12 will be accompanied by a**disruption of nucleo-cytoplasmic synchronism**. Excessive lengthening of the cell cycle results in the formation of cells with an "immature" appearing nucleus while the cytoplasm continues to differentiate at a normal rate and becomes giant with a content rich in hemoglobin (mature).

2.3. Clinical

Vitamin B9 and/or B12 deficiency manifests itself by clinical signs common to both vitamins: classic symptoms of anemia whose very gradual onset explains why they may not be**discovered only at a stage of profound anemia**, hemorrhagic syndrome in very advanced forms due to severe thrombocytopenia, and atrophy of the digestive, genital and urinary mucous membranes, explained by the extension of the defect in DNA synthesis to other tissues with rapid renewal.
Due to its role in the nervous system, vitamin B12 deficiency can be associated with neurological signs which will not be found in B9 deficiency. Combined spinal cord sclerosis is classically described, which combines posterior cord and pyramidal damage.

2.4. Biological diagnosis

Cytopenias will be found on the CBC with a**macrocytic anemia, normochromic, aregenerative** in the foreground. Depending on the depth of the vitamin deficiency, leuko-neutropenia and/or thrombocytopenia may be associated. The myelogram when performed reveals a megaloblastic marrow, with erythroblastic hyperplasia. Nowadays,**the biological diagnosis is confirmed on serum vitamin dosages**.

2.5. Etiologies

The main etiologies are summarized in the table below. Note that the most common cause of**vitamin B9 deficiency**in developed countries is represented by the**undernutrition**(elderly subjects, anorexia nervosa)**And**by**the vegan diet**And,**for vitamin B12, Biermer's disease**. Biermer's disease

is characterized by autoimmune gastritis leading to a defect in the synthesis of intrinsic factor which protects vitamin B12 in its intestinal transit. This results in a breakdown of vitamin B12 which is not absorbed.

B9 deficiency	B12 deficiency
Intake deficiency - Malnutrition - Anorexia nervosa	Intake deficiency - Strict vegan diet
Absorption defect - Celiac disease - Jejunal resection	Absorption defect - Biermer's disease - Crohn's disease - Gastric, ileal resection
Intestinal consumption - Microbial proliferation	Intestinal consumption - Microbial proliferation - Pests
Folate metabolism disorder - Antifolic medications - Cirrhosis	Drugs : - metformin
Increased needs - Hemolysis - Pregnancy - growth	Genetic diseases (rare) - congenital atranscobalaminemia - transcobalamin receptor defect

2.6. Therapeutic approaches

Treatment will combine replacement with the deficient vitamin and etiological treatment. The route of administration and duration of treatment will depend on the etiology.

3. Constitutional hemoglobinopathies

Constitutional hemoglobinopathies include genetic diseases affecting hemoglobin. These are mainly sickle cell syndromes, thalassemia syndromes and more rarely porphyrias.

Structure of globin(see course “physiology of the red blood cell”

3.1. Sickle cell syndromes

3.1.1. Pathophysiology

Sickle cell anemia, or hemoglobinosis S, is a genetic pathology mainly affecting black people. The substitution concerns the**6th amino acid of the β chain of globin**(valine instead of glutamic acid). This anomaly favors the formation of**globin polymers in case of deoxygenation or presence of inflammatory cytokines**. These polymers deform the RBC and give it a sickle-shaped appearance (sickle cell or “sickle cell”).**sickle cell**").This abnormally shaped red blood cell tends to get blocked in the small vessels, forming ischemic accidents. In addition, these rigid red blood cells are more easily phagocytosed by macrophages, leading to**hyperhemolysis**. Only homozygous (SS) patients are seriously affected because, in the case of a heterozygous (AS) mutation, the hemoglobin A molecules which constitute approximately 50% of the Hb pool prevent the formation of polymers.

3.1.2. Clinical picture

There**Heterozygous sickle cell disease is asymptomatic**(sickle cell trait) and does not require any treatment apart from genetic counseling to prevent possible homozygous offspring.
There**Homozygous sickle cell disease is symptomatic**, from early childhood (except in the first months of life when physiological HbF production persists). It is characterized by

- A**associated anemic syndrome +/-**Has**signs of hemolysis**
- of the**acute ischemic accidents called “vaso-occlusive”**which can affect all parts of the body (abdominal, bone, thoracic, etc.) and which manifest as intense pain

- of the**acute hemolytic accidents**
- chronic complications linked to repeated ischemic attacks (PAH, growth retardation, blindness, deafness, motor deficits, etc.)

3.1.3. Biological table

Sickle cell disease manifests itself as**normochromic, normocytic, regenerative anemia**with the presence of abnormally shaped red blood cells on the smear,**sickle cells**(form of forgery). He associates himself with**biological stigmata of hemolysis**with an increase in unconjugated bilirubin, a collapse in haptoglobin and an increase in LDH. Hemoglobin electrophoresis makes the diagnosis by revealing the**presence of hemoglobin S**.

3.2. Thalassemic syndromes

3.2.1. Pathophysiology

This is an extremely heterogeneous group of pathologies defined by the**lack of synthesis of one of the α or β hemoglobin chains**. the imbalance between the α and β chains in the erythroblast leads to the precipitation of excess chains leading to its**intramedullary destruction**. There are 2 α genes (therefore 4 alleles) and 1 β gene (2 alleles):**depending on the mutated gene, the number of mutations/deletions**, and the possible association with another Hb pathology (e.g. sickle cell disease),**anemia is more or less severe**.

3.2.2. Clinical picture

Depending on the genotype we witness**anemic tables of precocity and variable depth**ranging from antenatal anemia requiring transfusions*in utero*to a better tolerated anemia developing in early childhood. There is a note**hemolytic**in these patients**with frequent presence of jaundice**. In the event of insufficient care (absent or inadequate transfusions), the appearance of extra-medullary erythropoiesis may be observed, leading to bone deformations.
A number of patients are asymptomatic. They present a**thalassemia trait**, that is to say microcytosis associated with mild anemia,**linked to a heterozygous mutation**affecting 1 to 2 α alleles or 1 β allele.

3.2.3. Biological table

The hemoglobinosynthesis disorder is in the foreground, revealing itself as aregenerative microcytic anemia. Hemoglobin electrophoresis makes the diagnosis by analyzing the distribution of the different physiological hemoglobins.

3.2. Porphyrias

3.2.1. Pathophysiology

It is**very rare diseases affecting heme synthesis**. The most common is represented by Gunther's disease consisting of a deficiency in uroporphyrinogen III-cosynthetase, it is characterized by the accumulation of uroporphyrin I and coproporphyrin I, a red pigment which colors the urine.

3.2.2. Clinical picture

It is marked by**skin manifestations**various, one**splenomegaly**, there**red coloring of teeth**and **acute intermittent hemolytic accidents**.

3.2.3. Biological table

We again find a**aregenerative microcytic normochromic anemia**.