THE ROLE OF CBC IN THE INVESTIGATION OF BLOOD DISEASES

Olga-Alina, RADA[1], Mihaela Liana FERICEAN[1], Mihaela OSTAN[1]
[1]Banat University of Agricultural Sciences and Veterinary Medicine "King Michael I of Romania" from Timisoara, Timisoara, 300645, Romania
Corresponding author:olga_rada@usab-tm.ro

Abstract. *Anaemia indicates a pathological state, since it is the result of a disruption in the homeostatic balance between the production of erythrocytes and the loss/ destruction of erythrocytes. Running a complete blood count is the primary investigation that must follow the discovery of anaemia. In certain cases, after analysing the results of the CBC, the laboratory doctor may decide to run additional tests, such as a peripheral blood smear, which examines each type of cell in order to find anomalies in their quantity and quality and establish what type of anaemia the patient suffers from. The CBC and blood smears analysed for the purpose of this study were obtained from patients with pathological blood modifications or who were suspected of having blood diseases. The automatic CBCs were analysed with the automatic Sysmex XN-1000 haematology analyser. The blood smears, obtained through May-Grünwald-Giemsa staining procedure, were examined under the microscope and photographed. The numerical data obtained were stored and analysed statistically, and the link between the variables was established with the use of Pearson's correlation coefficient. Processing the CBCs revealed that the average number of erythrocytes was 3.28 ± 0.71/100^3/µL, which represents 72.88% of the minimum biological reference value. Statistically, this parameter has a positive correlation with the quantity of haemoglobin (r=0.694), whose average was 9.52±1.11 g/dL, meaning 73.22% of the minimum biological reference value, and with the haematocrit (r=0.759), whose average value was 29.57±3.90%, which is 73.92% of the minimum reference value. All these low values indicate the presence of anaemia. The values of the erythrocyte numbers pointed towards normocytic anaemia, despite some individual variations. The average numbers of white cells and platelets were slightly higher than the biological values, suggesting the correlated modification of the other types of blood cells, that comes with anaemia. The microscope examination of the blood smears revealed some modifications in the form of the erythrocytes (some that look like a stack of coins, knizocytes, sickle cells, degmacytes and ovalocytes), changes in erythrocyte diameter (mainly macrocytosis, indicating hypochromic anaemia, through a deficit of haemoglobin synthesis) and changes in the haemoglobin load (hypochromic erythrocytes, which indicate hypochromic anaemia).*

Keywords*: Anaemia, complete blood count (CBC), peripheral blood smear, erythrocytes, haemoglobin*

INTRODUCTION

Anemias are diseases of the erythrocyte, defined by decreased values of erythrocyte parameters. A person suffers from anemia if their hemoglobin (Hb) goes below 12 g% in women and below 13 g% in men (the main criterion), even if the normal distribution of hemoglobin varies not only according to the gender but also ethnicity and physiological status (CAPPELLINI, 2015). Additionally, the hematocrit (Ht) reaches values below 35% in women and 42% in men and the red cell count in peripheral blood is low. The homeostasis of the erythrocyte number is maintained through the balance between the production and destruction of red blood cells. Anemia occurs when the spinal cord is no longer able to produce enough red blood cells to cover the erythrocyte loss/destruction (https://kupdf.net/download/hematologie-clinica-studenti_595d71f7dc0d60f139e1ce2f_pdf). The blood loses its capacity to supply oxygen to the tissues (OGEDEGBE et al., 2004).

In most cases, anemia is not a diagnostic in itself, but rather a manifestation of a condition. There are multiple causes for anemia: decreased reserves of iron in the body, leading to an impossibility to produce hemoglobin (iron deficiency anemia) (WORLD HEALTH

ORGANIZATION, 2001; YATES et al., 2004; LIU & KAFFES, 2012; SHORT, DOMAGALSKI, 2013; HERSHKO & CAMASCHELLA, 2014; GELAW et al., 2019; SUNDARARAJAN & RABE, 2020); deficiency of certain vitamins involved in the synthesis of hemoglobin (folate, vitamin B_{12} - megaloblastic anemia) (ANKAR & KUMAR, 2020); acute or chronic inflammations (cancer, AIDS, kidney diseases, rheumatoid arthritis, Crohn's disease) (FRITZ et al., 2019), red bone marrow disfunctions (MOORE, 2020), autoimmune diseases (HILL et al., 2019; MICHALAK et al., 2020), peripheral erythrocyte destruction (hemolytic anemia), abnormal shapes of erythrocytes (falciform anemia) (HOWARD et al., 2015; TANABE et al., 2019; https://www.mayoclinic.org/diseases-conditions/anemia/symptoms-causes/syc-20351360)

Running a complete blood count is the mandatory primary investigation. In certain cases (established by the laboratory doctor according to the aspect of the CBC), additional tests are done, such as a peripheral blood smear (MIHĂESCU RODICA et al., 2006). A complete CBC (which in modern laboratories is done in 1 to 5 minutes, thanks to high performance equipment) contains the parameters that in many cases give valuable information warning about the cause of anemia. But after anemia has been confirmed by the determination of the values of hemoglobin, hematocrit and erythrocytes, a blood smear becomes a valuable instrument in the diagnose and assessment of anemia, respectively its aetiology can be clarified (WINTROBE'S CLINICAL HEMATOLOGY, 2003).

MATERIAL AND METHODS

In order to perform CBCs, blood samples were collected from patients by venous puncture, in vacutainer tubes, with EDTA anticoagulant, without prior centrifugation or by puncturing the pulp of the finger (peripheral blood). Automatic hemoleukograms were analyzed with the automatic Sysmex XN-1000 hematology analyzer. This device runs a complete CBC, measuring a number of hematologic parameters of venous whole blood collected on K_2EDTA as anticoagulant, in the ratio blood: anticoagulant, 9:1 (SR EN ISO 15189, 2013).

Peripheral blood smear examines each type of cell for quantitative and qualitative anomalies and for establishing the type of anemia. In our study, the smears were performed in compliance with the usual procedure, stained with May-Grümwald-Giemsa panoptic staining and examined under the OLYMPUS CX 41 microscope. The QUICKPHOTO MICRO 2.2 capture and statistical measurement software of the OLYMPUS CX 41 microscope was used for photography. The samples underwent microscopic examination, initially at low power (with a 20x objective) to assess the coloration and cell distribution and to estimate the number of erythrocytes as well as the presence of other blood cells. The smear was then examined with a 100x immersion objective, each cell type being evaluated for quantitative and qualitative anomalies. The numerical data thus obtained were stored and analyzed statistically, and the link between the variables was established by Pearson's correlation coefficient.

RESULTS AND DISCUSSIONS

The investigation of automated CBCs provides extremely precise and clear indications of the presence of a number of cells outside the biological limit of variation or of the presence of an abnormal distribution or morphology. This situation is signaled, in which case the CBC is followed by performing and examining the peripheral blood smear.

Table 1 presents the automated hemoleukograms of the investigated patients. The analysis of the CBCs revealed the following:

Table 1

The study of automated hemoleukograms of the investigated patients

Peripheral blood smear number	Age (years)	Sex (M/F)	RBC	HGB (g/dl)	HCT (%)	VEM (MCV) (fL)	HEM (MCH) (pg)	CHEM (MCHC) (g/dl)	WBC	PLT
1	41	F	1.93	8.1	22.6	117.1	42	35.8	80.49	148
2	71	F	3.03	9.9	28.3	93.4	32.7	35	19.03	22
3	80	M	2.79	8.3	28.4	101.8	29.7	29.2	4.64	122
4	61	M	2.60	7.5	22.8	87.7	28.8	32.9	0.53	14
5	42	M	3.45	9.7	31.3	90.7	28.1	31.0	13.74	10
6	36	F	3.93	10.8	31.8	80.9	27.4	33.9	37.70	89
7	43	M	3.38	9.6	31.6	93.5	28.5	30.5	80.20	171
8	41	F	4.65	9.9	31.2	69.3	21.3	30.7	62.30	287
9	73	M	3.95	11.2	36.6	92.7	28.3	30.5	82.10	239
10	77	F	2.92	10.4	31.5	108.0	35.8	33.3	2.08	158
11	72	F	3.36	9.0	28.3	84.0	26.7	31.8	71.90	192
12	64	F	3.47	9.9	30.5	87.9	28.4	32.3	106.20	193
Average			**3.28**	**9.52**	**29.57**	**92.25**	**29.80**	**32.24**	**46.74**	**137.08**
Standard deviation			**0.71**	**1.11**	**3.90**	**12.52**	**5.14**	**2.00**	**37.85**	**89.33**
% of normal minimumm			**72.88 %**	**73.23 %**	**73.92 %**	**115.31 %**	**110.37 %**	**102.34 %**	**116.85%**	**91.38%**
***Biological reference range/ unit**			**4.5-5.5/*10^3/µL**	**13-17 g/dL**	**40-50/%**	**80-100/fL Microcytes: < 80 Macrocytes: > 100**	**27-32/ pg**	**31.5-34.5 g/ dL Normochromic erythrocytes: 32-36 Hypocromic erythrocytes: <32**	**4-10 /*10^3/ µL**	**150-410 /*10^3/µL**

*- Bain, Barbara şi colab., Dacie and Lewis Practical Hematology, 11th edition, London, UK, 2012

below the minimum reference values | above the minimum reference values

The number of erythrocytes had an average of 3.28 ± 0.71/100^3/µL, which is only 72.88% of the minimum biological reference value. The gender distribution of blood counts revealed that in women the average number of erythrocytes was 3.32±1.03/100^3/µL (73.77% of the minimum normal value), while in men, the average erythrocyte number was 3.23±0.54/100^3/µL, which represents 67.77% of the minimum normal value. The erythrocyte number had a positive correlation with the quantity of hemoglobin which had an average of 9.52±1.11 g/dL, that is 73.22% of the minimum reference value (Table 1). It is worth noting that the values were below the normal biological interval in all analyzed patients, with no exceptions. Associated with the low number of erythrocytes, the diagnosis can be oriented towards anemia. The analysis in relation to the genders proved that the same distribution was kept: in women, the average hemoglobin value was 9.71±0.90 g/dL (74.69% of the minimum reference value); in men, the average hemoglobin value was 9.26±1.42 g/dL (71.23% of the minimum reference value);

The hematocrit measures the ratio of erythrocyte volume to total blood volume. In all patients, the hematocrit value was below the normal physiological value, in correlation with the number of erythrocytes and the quantity of hemoglobin. In female patients, the hematocrit value was 29.17±3.23/% (72.92% of the minimum reference value) and in male patients it was 3.14±5.05/% (75.3% of the minimum reference value) (Table 2).

Erythrocyte indices revealed the following aspects: the average value of the medium erythrocyte volume VEM (MCV) was 92.25±12.52/fL (in women the average was 91.51±16.35/fL and in men, 93.28±5.26/fL), which places erythrocytes in the category of normocytes. Still, there were exceptions: one CBC indicated microcytosis (1 woman) and 3 CBCs (2 women and a man) indicated macrocytosis (Table 2). Mean corpuscular hemoglobin (HEM) value was 29.80±5.14/pg (110.37% of the minimum reference value), where the mean value was 30.61±6.80/pg (113.37% of the minimum reference value) in women and

28.68±0.62/pg (106.22% of the minimum reference value), in men. Similar to the parameter previously analyzed, there were individual variations; thus, the values of 3 CBCs for women were under the minimum physiological limit, and the values of other 3 CBCs, also for women, were above the maximum physiological limit (Table 2).

Table 2

The study of automated hemoleukograms distributed by sex

Nr.crt.	RBC		HGB (dL)		HCT (%)		VEM (MCV) (fL)		HEM (MCH) (pg)		CHEM (MCHC) (g/dl)		WBC		PLT	
	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M
1	1.93	2.79	8.1	8.3	22.6	28.4	117.1	101.8	42	29.7	35.8	29.2	80.49	4.64	148	122
2	3.03	2.6	9.9	7.5	28.3	22.8	93.4	87.7	32.7	28.8	35	32.9	19.03	0.53	22	14
3	3.93	3.45	10.8	9.7	31.8	31.3	80.9	90.7	27.4	28.1	33.9	31	37.70	13.74	89	10
4	4.65	3.38	9.9	9.6	31.2	31.6	69.3	93.5	21.3	28.5	30.7	30.5	62.30	80.20	287	171
5	2.92	3.95	10.4	11.2	31.5	36.6	108	92.7	35.8	28.3	33.3	30.5	2.08	82.10	158	239
6	3.36		9		28.3		84		26.7		31.8		71.90		192	
7	3.47		9.9		30.5		87.9		28.4		32.3		106.20		193	
Average	**3.32**	**3.23**	**9.71**	**9.26**	**29.17**	**30.14**	**91.51**	**93.28**	**30.61**	**28.68**	**33.25**	**30.82**	**54.24**	**36.24**	**155.57**	**111.2**
Standard deviation	**1.03**	**0.54**	**0.90**	**1.42**	**3.23**	**5.05**	**16.35**	**5.26**	**6.80**	**0.62**	**1.80**	**1.34**	**36.51**	**41.27**	**84**	**99.64**
% of normal minimum	**73.77**	**67.77**	**74.69**	**71.23**	**72.92**	**75.3**	**114.38**	**116.72**	**113.37**	**106.22**	**105.55**	**97.84**	**135.6**	**90.6**	**103.71**	**74.13**
***Biological reference range/ unit**	**4.5-5.5/*10^3/µL**		**13-17 g/dL**		**40-50/%**		**80-100/fL Microcytes:< 80 Macrocytes: > 100**		**27-32/ pg**		**31.5-34.5 g/ dL Normochromic erythrocytes: 32-36 Hypocromic erythrocytes: <32**		**4-10 /*10^3/ µL**		**150-410 /*10^3/µL**	

*- Bain, Barbara și colab., Dacie and Lewis Practical Hematology, 11th edition, London, UK, 2012

The mean corpuscular erythrocyte concentration (CHEM) had an average value of 32.24±2.00 g/ dL (102.34% of the minimum reference value), 33.25±1.80 g/ dL (105.55% of the minimum reference value) in women, while in men it was 30.82±1.34 g/ dL (97.84% of the minimum reference value). The values between 32-36 g/ dL suggest normochromic erythrocytes, while the values below 32 g/ dL point to hypochromic erythrocytes. In 28.57% of the total of CBCs analyzed, erythrocytes are within the range of hypochromia (2 CBCs from women and 4 CBCs from men) (Table 2).

White blood cell count was 46.74±37.85 thousand (116.85%), slightly higher than the biological values; the increase was evident in women (135.6%), while in men there was a slight decrease (90.6%), when compared to the minimum biological value (Table 2). The average platelet count behaved similarly to white cells, with an increased value in women (108.71%) and a visible decrease in men (74.13% of the minimum biological value), suggesting the correlated modification of the other types of blood cells, which accompanies anemia.

Table 3 present the descriptive statistical processing of the data obtained by processing the investigated CBCs, while Pearson's index highlights the correlation between the parameters. According to the latter, the number of erythrocytes shows strong positive correlation with the hemoglobin content (r=0.694) and hematocrit (r=0.759) and a negative

correlation with erythrocyte indices (VEM-mean corpuscular volume, HEM-mean corpuscular hemoglobin).

Table 3

Statistical processing of the data obtained by processing the investigated CBC

	RBC	HGB	HCT	VEM	HEM	CHEM
RBC	1					
HGB	0.694529044	1				
HCT	0.7597235	0.901207648	1			
VEM	-0.830400576	-0.284258445	-0.330662151	1		
HEM	-0.84108678	-0.270000392	-0.466196359	0.93040854	1	
CHEM	-0.476364283	-0.089742498	-0.503782976	0.345233178	0.661400334	1

The examination of blood smears revealed poikilocytosis, anisocytosis, changes in the hemoglobin load in the erythrocytes, as well as other aspects.

Shape variations of erythrocytes (poikilocytosis): there were erythrocytes that looked like a stack of coins – "short rouleaux", in which 3 to 5 red blood cells appear stacked on top of each other on the smear, which is a common occurrence in anemia (Fig. 1); this aspect can be associated or not with hypochromia or with anisocytosis. There were also erythrocytes with the aspect of intense rouleaux, which formation is considered a sign of malignity (Fig. 2). The investigation also found knizocytes (pinched erythrocytes), erythrocytes from which a fragment got detached, characteristic for Biermer's disease (when erythrocyte fragility increases) or exposing normal erythrocytes to severe trauma (Fig. 3). Drepanocytes or sickle cells from thalassemia (sickle cell anemia) appear after a transformation (which can be reversible or irreversible) of the discocytes in drepanocytes, degmacytes ("bite" cells) – cells which lack a semi-circular area on one side (Heinz bodies removed in the spleen); they appear in the deficit of glucose-6-phosphate dehydrogenase, hemolysis induced by oxidative drugs (Fig. 3) and ovalocytes (oval-shaped erythrocytes) in variable proportion.

Variation in the size (diameter) of erythrocytes (anisocytosis), i.e. the presence of microcytes, with a diameter that is smaller than that of normocytes (below 7.5µm, dry weight), associated with VEM values that are below 80 fL; or the presence of macrocytes, erythrocytes with a very big diameter and VEM values above 100 fL.

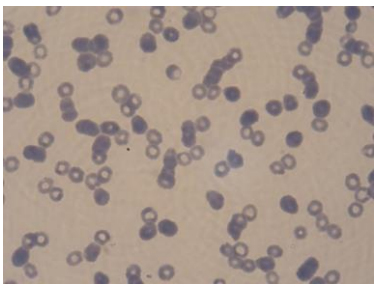

Figure 1. The aspect of erythrocytes displayed in "short rouleaux" associated with erythrocyte hypochromia (x100, original)

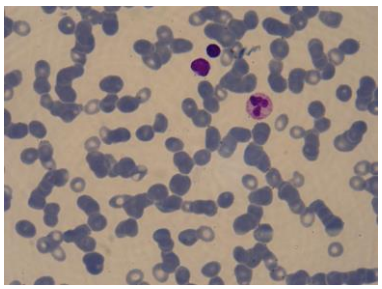

Figure 2. The aspect of erythrocytes displayed in "intense rouleaux" (x100, original)

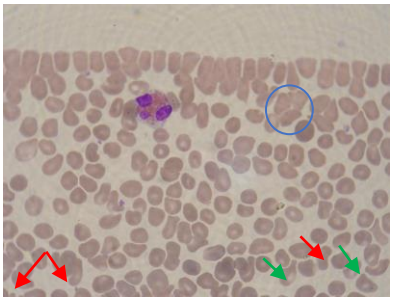

Figure 3. Knizocytes (pinched erythrocytes) (blue circle) associated with macrocytosis; rare drepanocytes (sickle cells) are present (⟶) and degmacytes ("bite" cells) (⟶) (x100, original)

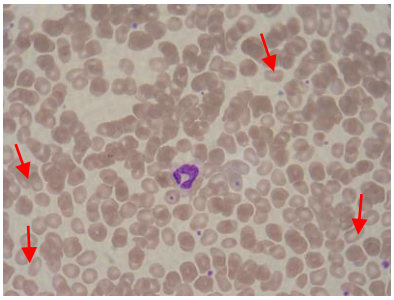

Figure 4. Ovalocytes (oval-shaped erythrocytes) (⟶) associated with anisocytosis (x100, original)

The examination of blood smears under the microscope revealed the presence of microcytes in low numbers but also of macrocytes in big numbers (Fig. 3). According to the data found in the specialized literature, macrocytes appear because of vitamin B12 deficiency and folate deficiency as well: both deficiencies lead to the impossibility to synthesize normal quantities of hemoglobin. Thus, the presence of macrocytes may indicate hypochromic anemia (ANKAR & KUMAR, 2020; RADA et al., 2018). Some aspects of anisocytosis were also observed, both microcytes and macrocytes could be observed on the same smear (Fig. 5).

Variations in hemoglobin load (color), were present in almost all peripheral blood smears examined where the aspect was hypochromic (pale erythrocytes with a lower hemoglobin load than normochromic erythrocytes); percentage-wise, hypochromia was associated with CHEM values, more evident in male patients, specific for hypochromic anemias (Fig. 1). Other aspects observed in blood smear examinations were those that indicate the presence of a large number of lymphocytes - these aspects were observed in two patients, where the suspicion of anemia may be associated with other pathologies, such as leukemias (Fig. 6). Myeloblasts, promyelocytes and myelocytes represent the mitotic compartment, the cells being capable of replication, and metamyelocytes, non-segmented neutrophils and segmented neutrophils represent the postmitotic / differentiation compartment. In the case of strong stimulation aimed to renew the white blood cell series, the metamyelocytes and myelocytes can get into the peripheral blood (Fig. 7). The presence of hypersegmented neutrophils was also detected: these appear in cases of megaloblastic anemia, folate deficiency or in antifolate chemotherapy (Fig. 8).

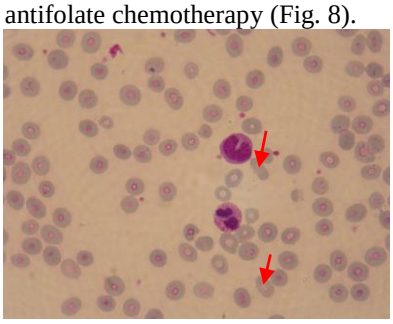

Figure 5. Ovalocytes associated with macrocytosis ( ); note the low density of erythrocytes in the examined microscopic field and anisocytosis (x100, original)

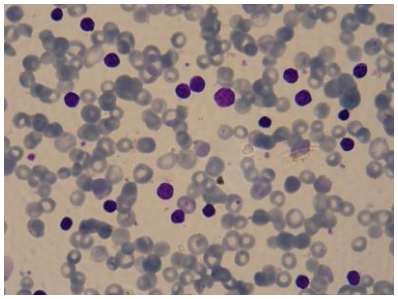

Figure 6. Aspect of anisocytosis and hypochromia; lymphocytes present in very large numbers (x100, original)

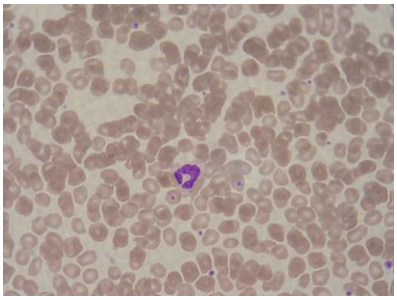

Figure 7. Myelocyte present in the examined microscopic field; normochrome erythrocytes agglutinated in a "short rouleaux" (x100, original)

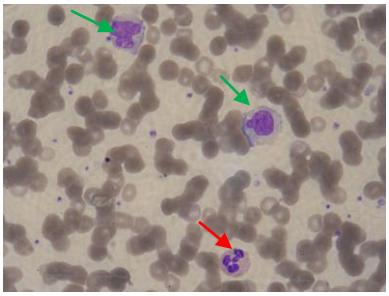

Figure 8. Numerous macrophages ( ); hypersegmented neutrophils ( ); erythrocytes displayed in "intense rouleaux" (x100, original)

CONCLUSIONS

The following aspects were revealed after processing the CBCs:

- the number of erythrocytes had an average of 3.28 ± 0.71/100^3/µL, representing only 72.88% of the minimum reference value (4.5-5.5/*100^3/µL). The gender distribution of CBCs revealed that in women the average erythrocyte count was 3.32±1.03/100^3/µL (73.77% of the minimum normal value), and in men the average erythrocyte count was 3.23±0.54/100^3/µL, which represents 67.77% of the minimum biological value.
- statistically, the low number of erythrocytes had a positive correlation with the hemoglobin load (r=0.694) the average value of which was 9.52±1.11 g/dL, meaning 73.22% of the minimum reference value (13-17 g/dL) and with hematocrit (r=0.759), the average values of which were 29.57±3.90%, meaning 73.92% of the minimum reference value (40-50/%). All these low counts point to anemia.
- together with the erythrocyte count, the hemoglobin and hematocrit load, the erythrocyte indices make it possible to steer the diagnosis towards a specific type of anemia; thus, VEM - the mean corpuscular value was 92.25±12.52/fL, which is 115.31% of the minimum reference value, categorizing anemia as normocytic. However, there were individual variations, one patient presenting values pointing to microcytosis and other three patients, to macrocytosis. These aspects were supported by the microscopic examination of blood smears.
- mean corpuscular hemoglobin (HEM) was 29.80±5.14/pg (110.37% of the minimum reference value) (the normal value being 27-32/pg); mean corpuscular erythrocyte concentration (CHEM) value was 32.24±2.00 g/dL (102.34% of the minimum reference value), meaning that most patients fell into the category of those with normochromic erythrocytes; however, there were individual variations, approximately 28.57% of the analyzed CBCs presented aspects of hypochromia.
- white blood cell count was 46.74±37.85 (116.85%), slightly higher than the biological values, suggesting the correlated modification of the other types of blood cells that comes together with anemia; it is worth noting that there was a single case, of a female patient, where the number of white blood cells was 106,20 thousand, which is ten times higher than the upper limit of the biological values (4-10/*10^3/ µL). This aspect was confirmed by the microscope examination of the peripheral blood smear (Fig. 3.12). In this case, anemia is secondary.
- the average platelet count had an evolution similar to the white blood cells, where there was an increase in the values in women (108.71%) and a marked decrease in men (74.13% of the minimal biological value).
- the microscope examination of blood smears revealed modified erythrocyte shapes (short or intense rouleaux, knizocytes, drepanocytes, degmacytes and ovalocytes), modified erythrocyte

diameter (mainly macrocytosis, indicating hypochromic anemia, caused by hemoglobin synthesis deficiency), as well as variations in the hemoglobin load (hypochromic erythrocytes, which indicates hypochromic anemia).

BIBLIOGRAPHY

ANKAR, A., KUMAR, A., 2020 - Vitamin B12 Deficiency, 2021- In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing. Available from: https://www.ncbi.nlm.nih.gov/books/NBK441923/.

CAPPELLINI, M. DOMENICA, MOTTA, IRENE, 2015 - Anemia in Clinical Practice—Definition and Classification: Does Hemoglobin Change With Aging? Seminars in Hematology, 52(4): 261-269.

FRITZ, J., WALIA, C., ELKADRI, A., PIPKORN, R., DUNN, R.K., SIERACKI, R., GODAY, P.S., CABRERA, J.M., 2019 - A Systematic Review of Micronutrient Deficiencies in Pediatric Inflammatory Bowel Disease. Inflamm Bowel Dis., 25(3):445-459.

GELAW, Y., WOLDU, B., MELKU, M., 2019, The Role of Reticulocyte Hemoglobin Content for Diagnosis of Iron Deficiency and Iron Deficiency Anemia, and Monitoring of Iron Therapy: a Literature Review, Clin Lab., 65(12). doi: 10.7754/Clin.Lab.2019.190315.

HERSHKO, C., CAMASCHELLA CLARA, 2014 - How I treat unexplained refractory iron deficiency anemia, Blood 123 (3): 326–333.

HILL, Q.A., HILL, A., BERENTSEN, S, 2019 - Defining autoimmune hemolytic anemia: a systematic review of the terminology used for diagnosis and treatment. Blood Adv., 3(12):1897–906.

HOWARD, J., HART, N. MARILYN ROBERTS-HAREWOOD, MICHELLE CUMMINS, AWOGBADE M., DAVIS, B., 2015 - Guideline on the management of acute chest syndrome in sickle cell disease Br J Haematol., 169 4 492 505, https://doi.org/10.1111/bjh.13348.

LIU, K., KAFFES, A.J., 2012 - Iron deficiency anaemia: a review of diagnosis, investigation and management. Eur J Gastroenterol Hepatol., 24(2):109–116.

MICHALAK, SYLWIA SULIMIERA, OLEWICZ-GAWLIK, ANNA, RUPA-MATYSEK, JOANNA, WOLNY-ROKICKA, EDYTA, NOWAKOWSKA, ELŻBIETA & GIL, LIDIA, 2020 - Autoimmune hemolytic anemia: current knowledge and perspectives, Immunity & Ageing (2020) 17:38 https://doi.org/10.1186/s12979-020-00208-7.

MIHĂESCU RODICA, IONIȚĂ HORTENSIA, PĂCURAR RODICA MIOARA, NICOLA DOINA**,** 2006 - Bazele teoretice hematologice în laboratorul clinic, LITO UMF, Timișoara.

MOORE, CHRISTINE A.; KOYAMANGALATH KRISHNAN, 2020-Aplastic Anemia, In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing. Available from: https://www.ncbi.nlm.nih.gov/books/NBK534212/.

OGEDEGBE, H.O., CSURY, L., SIMMONS, B. H., 2004 - Anemias: A Clinical Laboratory Perspective, Laboratory Medicine, vol. 3 (35):177-185.

RADA OLGA-ALINA, BREZOVAN DIANA, OSTAN MIHAELA, 2018 - Morphological changes of erythrocytes in hypochromic anaemia, in humans, Research Journal of Agricultural Science, 50 (3):3-9.

SHORT, M. W., DOMAGALSKI, J. E., 2013 - Iron Deficiency Anemia: Evaluation and Management, Am. Fam. Physician., 87(2):98-104.

SR EN ISO 15189, 2013-REGULAMENT SPECIFIC DE ACREDITARE în domeniul acreditării laboratoarelor medicale.

SUNDARARAJAN SRIPRIYA, RABE HEIKE, 2020 - Prevention of iron deficiency anemia in infants and toddlers, Pediatric Research, 89:63–73.

TANABE PAULA, SPRATLING REGENA, SMITH DANA, PEYTON, G., HULIHAN MARY, 2019 - CE: Understanding the Complications of Sickle Cell Disease, American Journal of Nursing, 119 (6):26-35.

WINTROBE'S CLINICAL HEMATOLOGY, 2003 – Editors: John P. Greer, Daniel A. Arber, Bertil Glader, Alan F. List, Robert T. Means, Frixos Paraskevas, George M. Rodgers, John Foerster (Editor Emeritus), Publisher: Lippincott Williams & Wilkins Publishers; 13th edition, Philadelphia, USA.

WORLD HEALTH ORGANIZATION, 2001 - Iron Deficiency Anaemia: Assessment, Prevention, and Control: A Guide for Programme Managers. Geneva, Switzerland: World Health Organization.

YATES, J.M., LOGAN, E.C., STEWART, R.M., 2004 - Iron deficiency anaemia in general practice: clinical outcomes over three years and factors influencing diagnostic investigations. Postgrad Med J., 80(945):405–410.

***https://kupdf.net/download/hematologie-clinica-studenti_595d71f7dc0d60f139e1ce2f_pdf

***https://www.mayoclinic.org/diseases-conditions/anemia/symptoms-causes/syc-20351360