

Diagnostic Cells in the Peripheral Blood Smear

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Abstract:

A blood film or peripheral blood smear is a thin layer of blood smeared on a microscope slide. Peripheral blood smear are usually examined to investigate hematological problems and occasionally, to look for parasites within the blood. An examination of the blood smear may be requested by physicians or initiated by laboratory staff. With the development of sophisticated automated blood-cell analyzers, the proportion of blood-count samples that require a blood smear. Nevertheless, the blood smear remains a crucial diagnostic aid. From the peripheral blood smear we can examine the number of white blood cells, platelets and to detect the rouleaux formation, anaemia, platelet clumps and leukocytic clumps and other abnormalities.

Key words : peripheral blood smear, schistocytes, lymphocytosis, thrombocytosis

INTRODUCTION:

A peripheral blood smear is a glass microscope slide coated on one side with a thin layer of venous blood. The slide is stained with a dye, usually Wright's stain, and examined under a microscope [1]. A physician-initiated request for a blood smear is usually a response to perceived clinical features or to an abnormality shown in a previous complete blood count. A laboratory-initiated request for a blood smear is usually the result of an abnormality in the complete blood count or a response to "flags" produced by an automated instrument. The indications for smear review differ according to the age and sex of the patient. Its major roles are in the differential diagnosis of anemia and thrombocytopenia and in the identification and characterization of leukemia and lymphoma [2]. We will discuss about the diagnosis of RBC, WBC and platelets morphologic abnormalities from the peripheral blood smear.

RBC MORPHOLOGICAL ABNORMALITIES :

Normal red blood cells are round to very slightly ovoid cells and a central pale area. Any deviation in size, volume, or shape of red cells which represents an abnormal red blood cell. The main disadvantage of the smear is a non-uniform distribution of red blood cells over the smear, with small crowded red blood cells at the thick edge and large flat red blood cells without central pallor at the feathered edge.

a.) Schistocytes:

The cell shape is the considerable diagnosis importance in the hemolytic anaemia. Some types of hemolytic anemia yield such a distinctive blood smear that the smear is often sufficient for diagnosis.

Microangiopathic hemolytic anemia may indicate pregnancy-associated hypertension, disseminated cancer, chronic disseminated intravascular coagulation, the hemolytic-uremic syndrome, or thrombotic thrombocytopenic purpura. Therefore this type of anemia is of considerable clinical significance. In microangiopathic hemolytic anemia, examination of the blood smear is also important to validate the platelet count, since red-cell fragments and platelets may be of similar size. Blood-smear features similar to those seen in microangiopathic hemolytic anemia are also a feature of mechanical

hemolytic anemia, that provide important evidence of this cause of hemolytic anemia. [3]

b.) Spherocytes :

Spherocytes are small, dense spheroidal RBCs with absence of Central pallor. It may result from hereditary spherocytosis, autoimmune hemolytic anemia or alloimmune hemolytic anemia so it is not diagnostically specific. When compared to Spherocytes, microspherocytes may be present in less number of patients. An osmotic fragility assay, Coombs' test, serum bilirubin, LDH, and haptoglobin, and other laboratory assays may be indicated. [4]

c.) Rouleaux formation:

In rouleaux formation RBCs are arranged in a form of coin stack i.e., linear arrangement. It is due to increase in the blood concentration of fibrinogen, globulin and paraprotein. The associated clinical disorders like multiple myeloma, acute and chronic inflammatory disorder, Waldenstrom's macroglobulinemia. In the absence of acute or chronic inflammatory disease, serum and urine analysis should be performed to determine if a paraprotein is present. [5]

d.) Bite cells :

Bite cells are also known as degmacytes in which RBCs are peripheral single or multiple defect. It can be found in normal individuals receiving large quantities of aromatic drugs which contains amino, nitro or hydroxyl groups. Bite cells can be accompanied by red cells with vacuoles, acanthocytes, schistocytes. Heinz body test, G-6-PD level, and other studies of red blood cell metabolism may be indicated. [6]

e.) Macrocytes:

Oval macrocytes are oval shape red cells with normal MCH. These cells suggest impaired bone marrow DNA synthesis and it may indicate folate or vitamin B12 deficiency. Bone marrow examination may be needed. Round macrocytes are round shape red cells and slightly larger than normal macrocytes. The cell suggests bone marrow impaired DNA synthesis, stress erythropoiesis, or excessive surface membrane. Clinical causes include obstructive jaundice, alcoholism, impaired DNA synthesis from

chemotherapy or inherited diseases, myeloproliferative disorders, myelodysplastic syndromes, or splenectomy thn.[7]

f.) Blister cells

Blister cells are red blood cells with vacuoles or markedly thin areas at periphery of membrane. These cells are characteristic of glucose-6-phosphate dehydrogenase (G- 6-PD) deficiency and other conditions imposing oxidant stress on the erythrocyte.[8]

g.) Elliptocytes

Elliptocytes are cells with an elliptical shape, while ovalocytes have an oval shape. Severe elliptocytosis is characteristic of hereditary elliptocytosis, but can be prominent in thalassemia, sickle cell trait, and Hb C trait. Rare elliptocytes occur in normal peripheral blood smears. Other diseases where elliptocytosis occurs include iron deficiency anemia, megaloblastic anemia, myelophthisic anemia, and mechanical trauma.[9]

h.) Nucleated red blood cells

Nucleated red blood cells are immature red blood cells. the presence of NRBCs indicates markedly accelerated erythropoiesis or severe bone marrow stress in an adult. The presence of NRBCs in the peripheral blood of an adult always indicates a significant disease process. NRBCs in the peripheral blood of an infant indicates significant stress. Clinical conditions associated with peripheral normoblastosis include acute bleeding, severe hemolysis, myelofibrosis, leukemia, myelophthisis, and [10]

i.) Keratocytes

Keratocytes are damaged red blood cells. Such damage characteristic occurs from fibrin deposits ,microangiopathic hemolytic anemia, thrombotic thrombocytopenic purpura (TTP), prosthetic heart valves, severe valvular stenosis, malignant hypertension, or march hemoglobinuria. Keratocytes occur in normal newborns with bleeding peptic ulcer, aplastic anemia, pyruvate kinase deficiency, vasculitis, glomerulonephritis, renal graft rejection, severe burns, iron deficiency, thalassemia, myelofibrosis with myeloid metaplasia, hypersplenism and post- splenectomy. These cells are pathologic and should never be ignored.[11]

j.) Microcytes :

Microcytes are small red blood cells with less amounts of hemoglobin. This is due to iron deficiency and defective hemoglobin synthesis, imbalance of globin chains, or defective porphyrin synthesis. Microcytes are usually present, and the mean corpuscular volume is decreased. Clinical causes are iron deficiency anemia, thalassemia, the anemia of chronic disease, lead poisoning, and sideroblastic anemias.[12]

k.) Hypochromia

Hypochromia is a decreased amount and concentration of hemoglobin in red blood cells. Hypochromic cells have an expanded central

zone of pallor in the peripheral blood smear. Microcytosis and hypochromia are characteristic of iron deficiency anemia and other microcytic, hypochromic anemias [anemia of chronic disease, hereditary hemoglobinopathies with diminished globin synthesis, red blood cell enzyme deficiencies . Serum iron studies, erythrocyte sedimentation rate (ESR), hemoglobin electrophoresis, bone marrow examination, and serum and urine lead quantitation are other laboratory studies may be indicated.[13]

l.) Hyperchromia

Hyperchromia is an increase in the red blood cell hemoglobin concentration. Since it is usually associated with spherocytosis, peripheral smear examination reveals many spherocytes and microspherocytes. Heinz body hemolytic anemia, hereditary pyropoikilocytosis, and severe burns. If indicated, an osmotic fragility assay, Coombs' test, serum bilirubin, LDH, and haptoglobin, and other laboratory assays may be indicated.[14]

m.) Polychromasia

Polychromasia is the occurrence of slightly immature red blood cells, which are larger than normal and have a blue-gray coloration. Polychromasia is due to the presence of ribosomal protein in immature red blood cells, which pick up the basophilic component of the Wright-Giemsa stain. Small numbers of these cells (0.5 - 2%) are normally present in the peripheral blood and signify the presence of erythropoietic activity in the bone marrow. The MCV may increase slightly in response to significant polychromasia. Decreased polychromasia is seen with hypoproliferative marrow states.[15]

n.) Howell-Jolly bodies

Howell-Jolly bodies are small dense, perfectly round basophilic red cell. It represent nuclear material derived from nuclear fragmentation or incomplete nuclear expulsion during normoblastic maturation. Howell-Jolly bodies are identified in splenectomized patients . It may also seen in smaller numbers in patients with megaloblastic anemia, severe hemolytic processes, hyposplenism, and myelophthisitic anemia.[16]

o.) Acanthocytes

Acanthocytes are spheroid RBCs with a few large spiny (thorny) projections. Occasional acanthocytes can be seen after splenectomy, in patients with alcoholic cirrhosis, and in hemolytic anemias caused by pyruvate kinase (PK) deficiency. microangiopathic hemolytic anemia, autoimmune hemolytic anemia, sideroblastic anemia, thalassemia, severe burns, renal disease. The majority of erythrocytes form acanthocytosis in the rare disease abetalipoproteinemia.[17]

WBC MORPHOLOGIC ABNORMALITIES :

The WBC is of great importance in the diagnosis and management of patients with hematologic and infectious diseases. White blood cells are classified according to their

functions namely neutrophils , lymphocytes , basophils , monocytes ,eosinophils. The specific morphologic abnormalities of leukocytes occur, and can provide evidence of disease processes.

a.) Neutrophilic hyper granulation:

Small dark blue granules resembling primary granules. It can be accompanied by a "shift to the left" in the neutrophilic population, and by the presence of vacuolations in the cytoplasm. It appear in the cytoplasm of methmyelocytes, bands, and segmented neutrophils during inflammatory states, burns, and trauma, and upon exposure to hematopoietic growth factors such as granulocyte-colony stimulating factor. It is also known as toxic granulation [18]

b.) Leukocytosis and lymphocytosis :

Blood smears should be examined when there is unexplained leukocytosis, lymphocytosis, or monocytosis .The role of the blood smear in the diagnosis of leukemia and lymphoma is to suggest a likely diagnosis or range of diagnoses, to indicate which additional tests should be performed, and to provide a morphologic context and sophisticated investigations cannot be interpreted.Blood smear facilitates rapid diagnosis and specific treatment for two conditions Burkitt's lymphoma and acute promyelocytic leukemia .[19]

c.) Dohle bodies :

Dohle bodies are blue or grayish-blue cytoplasmic inclusions .it can be various size and shapes and usually found near the periphery of the cell. Dohle bodies are lamellar aggregates of rough endoplasmic reticulum, which appear in the neutrophils, bands, and metamyelocytes of patients with infection, burns, uncomplicated pregnancy, toxic states, or during treatment with hematologic growth factors such as G-CSF.[20]

d.) Alder-Reilly granules:

Alder-Reilly granules are large, coarse, dark purple, azurophilic granules that occur in the cytoplasm of most granulocytes. These are characteristically found in the Alder- Reilly anomaly and in patients with mucopolysaccharidoses. [21]

e.) Neutrophilic hypersegmentation:

Increased lobulation of granulocyte nuclei is a characteristic finding in megaloblastic anemia, but can also be seen as an inherited autosomal dominant trait.

f.) Neutrophilic hyposegmentation:

Single or bi-lobed neutrophils can be inherited or acquired in patients with malignant myeloproliferative disorders and infections or tumors which have metastasized to the bone marrow.Large, purple or dark-blue azurophilic granules in the cytoplasm of neutrophils, bands, and metamyelocytes are characteristically seen in patients with severe infection, septicemia, toxic states, and chemical poisoning. Cytoplasmic vacuolation is also seen . [22]

PLATELET MORPHOLOGIC ABNORMALITIES :

The platelet count is one of several laboratory assays of importance in the functional evaluation of the hemostatic system.Platelet defects can be classified by their location in the three phases of clot formation: initiation, extension, and cohesion or aggregation or based on their particular structural or functional deficiency[23]. Light microscopy is of greatest value in confirming the automated platelet count .

a.) Platelet hypogranularity :

There is small, reddish-purple granules are present in the cytoplasm of the platelet. These granules are vary in size and shape, represent dense bodies, alpha- bodies, and lysosomes. These granules may be decreased in number or absent in patients with myeloproliferative diseases and myelodysplastic syndromes. Platelet hypogranulation is usually accompanied by abnormalities in platelet size and shape, anemia, leukocytosis or leukopenia, and leukocyte morphology.

b.) Platelet satellitism :

Normal platelets adhere to the surface of neutrophils, or, rarely monocytes, to form "platelet rosettes". Platelet satellitism may cause spurious thrombocytopenia, since the cell-bound platelets are not counted with the platelet fraction of the blood specimen. It may be associated with blood specimens anticoagulated with EDTA, and disappears when heparin-anticoagulated blood is collected from the same patient.[24]

c.) Thrombocytopenia and thrombocytosis:

Decrease in the platelet count may be the result of thrombocytopenia. Fibrin strands indicate that thrombocytopenia . Thrombocytopenia whether inherited or acquired will impact all three phases to varying degrees based on the severity of the platelet deficiency. Underlying causes that may be re- vealed by the blood smear include the May-Hegglin anomaly , microangiopathic thrombopathies, and leukemias and lymphomas.

Thrombocytosis or thrombocythemia is the presence of high platelet counts in the blood, and can be either primary or secondary. Examination of thrombocytosis is for evidence of a myeloproliferative disorder, such as giant platelets, or an increase in the basophil count. If the cause for the high platelet count remains unclear, bone marrow biopsy is often undertaken, to differentiate whether the high platelet count is reactive or essential.[25]

d.) Large and giant platelets :

Normally platelets are 1.5 to 3 microns in diameter. But the large platelets are 3 to 7 microns , while giant platelets are larger than red blood cells. Morphology may appear normal or abnormal. Platelet size can increase with increased platelet turnover from bleeding or stress, and in the myeloproliferative and myelodysplastic disorders.[26]

CONCLUSION :

The blood smear can have an important part in the speedy diagnosis of certain specific infections. Members of the laboratory staff should make and examine a blood smear whenever the results of the complete blood count. Physicians should request a blood smear when there are clinical indications for it. The blood smear is essential for the validation or the further elucidation of a detected abnormality. All laboratories should have a protocol for the examination of a laboratory-initiated blood smear, which can reasonably be based on the criteria of the International Society for Laboratory Hematology.

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