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NUMERICAL ABNORMALITIES OF THE SPLEEN: IMMUNOHEMATOLOGICAL AND ANATOMIC CONSEQUENCES

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Congenital asplenia is a rare disease which occurs predominantly in boys and is associated with severe infections that can influence the long-term survival. Polysplenia disorder, defined as the presence of multiple spleens of almost equal volume, is also a rare disorder, but it mainly affects girls and the long-term survival is often possible. The case report presents the diagnostic difficulties of a 2 years old boy who presented leukocytosis and persistent thrombocytosis, together with the methods of diagnosis which led to the identification of the isolated congenital asplenia and the case of a 4 months-old girl who presented for the evaluation of a tuberous hemangioma at tight, which was later diagnosed with polysplenia syndrome, knowing that these two syndromes are associated with plurimalformative organic disorders. In terms of morphogenesis, the splenic abnormalities are the result of an unknown teratogenic factor which occurs in the embryonic development in the first 5 weeks of gestation. These rare affections represent the result of an early unknown teratogenic insult which came about during the embryogenesis and complex investigations are needed in order to exclude pluriorganic affection. The cases presented here are relevant for the identification of the haematological, immunological and imaging methods and aim to evaluate the function of the spleen, the splenic hypofunction being also encountered in other chronic disorders in children.

Key words: asplenia, polysplenia, hyposplenism, thrombocytosis, leukocytosis.

INTRODUCTION

The numerical abnormalities of the spleen together with malpositions and malformations – especially the cardiovascular ones – have been reported more than 60 years ago, when the first case of congenital asplenia was diagnosed by Myerson and Koelle (1956)[1].

The 12th day of the embryogenesis represents the initial moment of the spleen formation, which coincide with the initial formation process of the left-right asymmetric axis.

In terms of morphogenesis, the splenic abnormalities are the result of an unknown teratogenic factor which occurs in the embryonic development in the first 5 weeks of gestation[2].

Congenital asplenia is a rare disease, more frequent to boys, encumbered with a high risk of severe infections which can influence long-term survival. Asplenia is associated with great vessel transposition, pulmonary stenosis and anomalous pulmonary venous drainage.

Polysplenia disorder, defined as the presence of multiple spleens of almost equal volume, is a rare disorder, but it mainly affects girls. Long-term survival is often possible. Polysplenia is more frequently associated with an abnormality of the inferior vena cava which drains via Azygos system[3].

Through its unique mode of organization, the spleen plays an essential role in the immunological response both innate and adaptive.

The spleen structure makes it act like a filter which participates in the destructions of the three aged lines or malformed erythrocytes and thrombocytes and also of the micro-organisms arrived in the circulation. Throughout the associations of these two functions, the spleen becomes the most important organ involved in the immunological antifungal and antibacterial reactivity.

The red pulp of the spleen has an anatomical structure which consists in a venous sinuses system which plays the role of a filtering system and also

has a critical role in the body's ability to recycle iron and globine resulting from the removal of the red cells.

In addition, these macrophages are involved in the removal of bacteria from blood circulation by interfering with the intracellular iron transport mechanism.

The white pulp of the spleen is organized as a lymphoid organ, having the B and T cells disposed around an arterial ramification. This layout makes it resemble with the structure of the lymph node[4].

The white pulp contains germinal centers with lymphocytes and macrophages and plays an essential role in both innate and adaptive immunity. The white pulp is also a reservoir for B and T lymphocytes and plays an active role in the IgM and compliment, both involved in the opsonisation of the bacteria for their destruction and removal.

The spleen is a complex organ which combines in an efficient way the erythrocytes phagocytosis with the iron recycling as well as the capture and destruction of the pathogen agents with the initiation of the immune response.

These functions are being combined in a unique mode in a single organ which is compartmented in different zones, situation which does not exist in any other lymphoid organ.

MATERIALS AND METHODS

The quantifying methods of the splenic function are based on the haematological, immunological and scintigraphic images.

The haematological parameters indicate the spleen filtering function. The presence of the aberrant erythrocytes, thrombocytosis and leucocytosis indicate hypo/asplenia.

The immunological parameters are based on the role of the spleen in CD4 + CD8 + T cell activation. Scintigraphic parameters indicate the capacity of the spleen to filter the blood of cells and particles to measure its activities.

The suggestion of the functional hyposplenism is raised by the presence of the Howell-Jolly bodies (clusters of DNA in erythrocytes) on the peripheral blood smear.

The quantification of changes appeared on the erythrocyte membrane represents the gold standard for the splenic hypofunction diagnosis.

With the electron microscope large vacuoles named "pitts" are visible on the erythrocyte membrane. The normal splenic functions are considered according to the number of vacuoles relative to the number of circulating erythrocyte: 0–4% of erythrocyte with these changes – normal; > 4% – hyposplenism and between 15–70% – asplenia[4].

The ^{99m}Tc sulphur colloid scintigraphy is being used to visualize the liver and the phagocytic function of the spleen.

Yet, the latest recommendation is to use the scintigraphy with the use of heat-damaged Technetium-99m labeled erythrocytes because this is not influenced by the liver uptake and also allows the estimation of the functional splenic volume.

The immunological response can be estimated through the immune response after vaccination. There is a positive correlation between the IgM CD27 + level and the functional splenic volume[6].

The imagistic methods are necessary to establish the anomalies in the number of spleen and possible associated malformations.

This report presents two cases of numerical abnormalities of the spleen diagnosed during one year at INSMC Alessandrescu - Rusescu, using the methods listed above.

FIRST CASE REPORT

The I.A. male patient is hospitalized at the age of 1 year and 9 months with respiratory symptoms and high fever. He received a treatment with broncho-dilators and anti-inflammatory drugs administrated in aerosols. These were later associated with a treatment with antibiotics, which improved the general state, but didn't annihilate the fever completely. As soon as the antibiotic therapy was interrupt the fever reappeared.

The laboratory investigations show a significant leukocytosis due mostly to the growing number of the neutrophils, in association with an increased number of basophils as well as in the number of monocytes. The increase number of the platelets is very important as well. This modification in the complete blood count (CBC) remains unchanged during the entire hospitalization, despite the given treatment with antibiotics.

The patient medical history tells us that after repeated CBC investigations, done during the hospitalization periods which happened between the age of 4 months and 1 year and 7 months, the same changes were registered: leukocytosis with lympho-

monocytosis, associated with marked thrombocytosis.

The peripheral blood smear was indicating poikilocytosis with ovalocytes, schistocyte, erythrocytes in drops, anysocytosis with microcytosis.

The haematological investigation excludes the probability of a haematological malignancy and it is considered that the haematological alterations are probably due to an acute respiratory infection.

In January 2015 the patient returned at the clinic with fever, coughing, rhinorrhea. The CBC has the same alterations. By correlating the clinical results (frequent respiratory infections) with the laboratory results, the idea of a differential diagnoses with different causes of leukocytosis is being raised.

The number of leukocytes is the result of the production and release in circulation on one hand and the destruction and removal from the circulation on the other hand. Therefore the alteration of any component of this balance leads to a modification in the number of leukocytes in the peripheral blood. The decrease of the removal in the circulation can appear as a consequence of the administration of corticosteroids, of splenectomy or of the leukocyte adhesion molecule deficiency.

From a clinic point of view the leukocytosis can be caused by infections, inflammations, allergic reactions, malignancies or other causes such as splenectomy.

In case of leukocytosis associated with alterations of the erythrocytes from the blood smear (spherocytosis, acanthocytosis, Howell-Jolly bodies) accompanied by an important thrombocytosis the functionality of the spleen can be put to discussion.

In order to determine the functionality of the spleen we used two of the presented methods, the haematological and the immunological methods, to which it adds the imagistic method.

A new peripheral blood smear was made, but this time besides all the changes mentioned earlier – poikilocytosis, with rare ovalocytes, schistocyte, and rare spur cells – the new test showed the presence of erythrocytes with Jolly bodies.

The spleen has the capability to phagocytes abnormal erythrocytes, but in case of a dysfunction, this ability is affected, resulting in a growth in circulation of the abnormal erythrocytes. In the same time, the growth in number of the circulating thrombocytes represents an indicative of the hyposplenism. One of the first methods of evaluation of a splenic function was the detection of erythrocytes, which contain Howell-Jolly bodies.

Other abnormalities can be visible in the peripheral blood smear in patients with an absent or lowered splenic function- acanthocytosis, target cells, Heinz bodies, and siderocytes.

In our case the immunophenotyping showed a decrease in the CD3+CD4+ T cells with CD3+CD8+ at normal.

Among the imaging methods, abdominal ultrasound examination is the first to come, often enough for splenic pathology and having the same diagnostic accuracy as the CT.

In our case, repeated abdominal ultrasound examination at different times and by different examiners doesn't visualize the spleen. This, together with the erythrocytes modifications on the smear, raised suspicion for a congenital asplenia.

The CT did not detect any parenchymal intra-abdominal structure with the characteristic spleen dynamics. Therefore the spleen is not visible on native sequences or with contrast agents.

Other syndromes associated with asplenia were being excluded, syndromes such as: Ivemark syndrome (in which visceral heterotaxy is present with bilateral right-sidedness. The right-sided organs are duplicated, and organs that are normally present on the left side are absent. This syndrome is accompanied with complex malformations, Pearson syndrome (pancreatic insufficiency, sideroblastic anemia), Stormoken syndrome (thrombocytopenia and miosis), Smith-Fineman-Myers syndrome (mental retardation, short stature, cryptorchidism)

Corroborating: the leukocytosis, the thrombocytosis, the modification of the blood smear, the immunofenotyping, the undetectable spleen on the US, the absence of the spleen on CT, the definitive diagnosis of isolated congenital asplenia is being settled.

SECOND CASE REPORT

A 4 months old female patient with irrelevant family history was presented for evaluation of a cavernous hemangioma on the left thigh. The physical examination did not reveal any other abnormality.

The thrombocytosis on the CBC count (768000/μl) are associated with the red cells

abnormalities in the peripheral smear including an increased presence of Pappenheimer bodies, poikilocytosis, target cells and the presence of Howell-Jolly bodies.

All this suggested splenic dysfunction therefore a abdominal ultrasonography was taken, which showed 8 spleens of almost identical size. The diagnosis of polysplenia was established.

Knowing that morphogenetic congenital anomalies of the spleen are the result of a teratogenic factor which occurs in the first 5 weeks of gestation and that polysplenia is often associated with impaired inferior vena cava, an echo-cardiography was performed. This revealed the existence of an ambiguous situs with an interruption of the inferior vena cava with drainage via the hemiazygos system. The suprahepatic veins drain directly in the right atrium.

The diagnostic of heterotaxic syndrome with polysplenia is being fixed.

In this case the immunophenotyping was normal. The immunological response will be evaluated using the level of IgM and IgG after vaccinations.

RESULTS

These two cases overlap with the existing data from the speciality literature referring on the numerical abnormalities of the spleen; asplenia is a rare disease which occurs predominantly in boys and is associated with severe infections that can influence the long-term survival. Medical care involves antibiotic prophylaxis, appropriate immunization, aggressive management of suspected infection and parent education.

Polysplenia is more frequent to girls and it is associated with abnormalities of the inferior vena cava which drains via the Azygos system.

CONCLUSIONS

Many medical conditions are being associated with the splenic hypofunction:

Hematologic-neoplastic disorders: sickle cell disease, bone marrow transplantation, acute leukemias, non-Hodgkin's lymphoma, advanced breast cancer.

Hepatic disorders: alcoholic liver disease, chronic active hepatitis, liver cirrhosis and portal hypertension, primary biliary cirrhosis.

Autoimmune disorders: systemic lupus erythematosus, antiphospholipid syndrome, vasculitis, rheumatoid arthritis, Hashimoto's thyroiditis.

Gastrointestinal diseases: celiac disease, Crohn's disease, ulcerative colitis, Whipple's disease.

Infectious diseases: acquired immunodeficiency syndrome.

Circulatory disorders: splenic/celiac artery thrombosis, splenic vein thrombosis.

Miscellaneous: amyloidosis, sarcoidosis, primary pulmonary hypertension, total parenteral nutrition, splenic irradiation, high dose corticosteroid administration[4].

The hypofunction of the spleen is due on one hand to the decrease of the splenic function during the activity of the disease and can be improved with treatment. On the other hand it's due to the splenic atrophy, which is irreversible and is caused by the loss of the splenic volume, which has as a consequence the function loss.

The functional hyposplenism is associated with overwhelming infections, due to gram positive and capsulated organisms, which implies antibiotic prophylaxis and aggressive management of suspected infection.

The haematological abnormalities such as leukocytosis and/or monocytosis as well as thrombocytosis are frequent in patients suffering from hyposplenism. Therefore the risk for thrombotic accidents increases.

The congenital asplenia and polysplenia are rare diseases, but important ones, which are missing in the statistics.

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