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/Following is a translation of an article by F. B. Yermakova of the Department of Pathological Anatomy of the 1st Leningrad Medical Institute imeni Academician I. P. Pavlov. The article appeared in Arkhiv Anatomi, Gistologii i Embriologii (Archives of Anatomy, Histology and Embriology), Vol. XXXVIII, No. 5, Moscow, 1960, pages 56-59./

No data on the features of hemopoietic organs of mice with age were found in the literature available to us. Detailed information exists only on quantitative and qualitative composition of blood under normal (Klinenberger and Carl, 1927; Downey, 1938; Kock, 1931; Hoffman, 1956) and under pathological conditions (Furth and Strumia, 1931; as well as papers on cellualr composition of their bone marrow (Jaffe, 1931; P. P. Sakharov, 1952).

A knowledge of the formation of hemopoietic organs is important as a control in a number of investigations concerning mice, especially in the study of questions concerning disease of hemopoietic organs, especially leucosis and the pre-leucosis state. In this investigation the morphological features of hemopoiesis in mice were studied for different growth periods.

A total of 150 mice were employed in this study, of these, 130 were normal and 20 were of the high leucosis strain Afb at an age of 1 day to 5 months. The mice were killed with chloroform. In dissection, blood was taken from the heart for calculation of the number of erythrocytes and leucocytes; sections of bone marrow, or femur, chest, liver and spleen were subjected to histological examination. In a number of instances impressions of organs and smears of peripheral blood were made. Organs were fixed in Bouin's fluid, less often in 10% formalin, and formed in paraffin-celloidin. Staining was done with hematoxylin-eosin, Van Gleson's stain and azure-eosin.

Data from the literature on the composition of peripheral blood of mice is very contradictory. It is known that blood composition is affected by the site of withdrawal. According to Klinenberger and Carl, Downey, Petri (1932), formed elements are considerably more numerous in peripheral blood taken from the tail of a mouse than in the same blood taken from vessels of the femur (erythrocytes by 500 thousand and leucoytes by 4 thousand). Blood taken from the heart gives more constant figures. There are somewhat more erythrocytes in males than in females. There are less erythrocytes in young mice than in adults.

The data obtained by us on the number of erythrocytes and leucocytes in peripheral blood taken from the heart of mice of various ages are presented in the table.

Age group of mice	Normal mice		
	Number of mice	Average number of erythrocytes (millions)	Average number of leucocytes (thousands)
1-3 days	30	3.0	3.5
15-16 days	25	7.5	5.1
1 month	30	7.9	8.2
1½-2 mos.	25	7.1	7.8
5 months	20	7.5	8.5
Age group of mice	High leucosis strain Afb		
	Number of mice	Average number of erythrocytes (millions)	Average number of leucocytes (thousands)
1-3 days	5	3.0	3.8
14-16 days	5	7.1	
1 month	5	7.5	7.7
1½-2 mos.	5	7.8	8.3

The increase in the number of blood elements in mice of the high leucosis strain Afb occurs in approximately the same way as in normal mice of corresponding age groups. According to the data of A. M. Ginzburg in examinint the blood of 38 mice of high leucosis strain Afb, the average number of leucocytes in the age 2-8 months is 11,200.

In examining the blood smears of strain and normal mice we discovered large and small lymphocytes, neutrophil leucocytes, eosinophils, at times monocytes. Petri indicates the presence of basophil leucocytes in blood taken from the heart and tail of mice, but we did not succeed in finding them.

It is well known that the marrow of spongy and long bones of mice functions throughout life. Between the acrtilaginous trabeculae of the marrow cavity of the femur are large light irregularly shaped,

often extended, cells scattered in groups of 5-6 cells. Their cytoplasm acquires a basophil tinge, turning into islets of erythroblasts consisting mostly of round cells with dark violet "velvet" nucleus. At times, however, indifferent light-nucleated cells appear among them.

The single larger cells with reticulated or delicately meshed chromatin distributed in the nucleus and 1-2 nucleoli with delicately colored cytoplasm we call hemocytoblasts. Jaffe and P. P. Sakharov do not distinguish them as a separate group, apparently combining them with myeloblasts. Elements of the myeloid service in this period are separate eosinophil myelocytes and small conglomerations of leucocytes. In addition there are large rounded or irregularly-shaped hyperchromatic cells. In some cells the cytoplasm is in the form of a narrow bluish ring in which small azure-colored granules are visible. If the cell contains many inclusions, then the nucleus is not outlined and the cell presents a compact hyperchromatic mass. Klinenberger, observing such cells in the bone marrow of rats, referred to them as fat cells. We did not observe such cells in adult mice. Their presence in new-born mice can apparently be due to the partial disruption of immature indifferent cells of bone marrow and subsequent phagocytosis of nuclear fragments. Among cells of the bone-marrow cavity undergoing phagocytosis are cells of the reticuloendothelium, therefore we, as well as Lauda and Flaum (cited by Ye. M. Frejfel'd), are incluned to relate these hyperchromatic cells to reticuloendothelium cells, filled with fragments of nuclear substances.

Thrombocytic sprouts are present in this period in an early form - a megacaryoblastic, large cell with large rounded nucleus and a wide field of pro, oplasm.

Liver hemopoiesis in mammals stops at the moment of birth (A. A. Maksimov, 1927), nevertheless in the liver of new-born mice there are always seen foci of embrionic hemopoiesis - up to 800 in the field of vision. In distended intralobular capillaries, between cords of liver cells and connective tissue layers we observed masses of small round cells with rich colored nucleus and a very narrow ring of protoplasm - erythroblasts and single-megacaryocytes.

Liver hemopoiesis decreases in the first days of life, and by the 8th-12th day in the life of mice the foci of hemopoiesis in the liver are not seen.

In the spleen of the new-born the rounded large follicles and fine layers of red pulp are clearly visible. The follicles consist of light cells of hemoblast type, a follicular artery is clearly visible in each follicle. In the fine layers of red pulp are located reticular cells with large light nuclei, at times rod-nucleated leucocytes can be observed. Cells are encountered in all stages of mitosis.

By the 15th day of life bone marrow hemopoiesis is intensified with a clear division of two sprouts, but erythropoiesis at this age preserves its leading position. Hemocytoblasts, myeloblasts, myelocytes and rod-nucleated leucocytes can be discerned. In each field of

vision no less than one megacaryocyts or megacaryoblast can be seen. The spleen begins to take a more active part in hemopoiesis; megacaryocytes appear in the red pulp. Beneath the capsule small foci of erythropoiesis are revealed. We did not observe foci of hemopoiesis in the red pulp, although, according to the data of Bloom (1926), small foci of myelopoiesis can be seen in the follicles, arising from the small lymphocytes which lie in embryonic centers.

The bone marrow of full-grown mice is distinguished by a diversity of cellular composition. Among the cells are hemocytoblasts, lying in groups are dark small rounded cells of the erythroblast series with "velvet" nucleus and larger light-nucleated cells of the myeloid series. Fields of leucocytes with rod-form and segmented nuclei are encountered in large number. Foci of erythropoiesis are less evident than in immature mice and proerythroblasts and basophil erythroblasts are present, however in places normoblasts of different degrees of maturity are visible.

Rod-nucleated neutrophil leucocytes with bent or segmented nucleus occupy a great area. Encountered in less quantity are the eosinophil myelocytes with large bright red granules in the protoplasm and leucocytes differing from neutrophils only in the red granularity of their protoplasm. In every field of vision 2-3 megacaryocytes can be seen with segmented nuclei and broad field of protoplasm.

Single cells can be seen passing in the liver capillaries of adult mice - lymphocytes and myeloid elements.

Red pulp predominates in the spleen, the follicles are somewhat reduced in volume. Beneath the capsule and in the red pulp, foci of erythropoiesis are visible and mature megacaryocytes are encountered. Small foci of myelopoiesis with differentiation to neutrophil leucocytes can be seen between sinuses and in the vicinity of trabeculae. Basophil leucocytes are encountered in the form of single cells.

It is well known (Downey and Klinenberger) that the hemopoietic activity of the spleen is maintained throughout life. However in mice hemopoiesis of the spleen is not identically evidenced with age. It is absent in the new-born. The first foci of hemopoiesis appear at the close of embryonic hemopoiesis in the liver. Then it grows parallel with the development of medullary hemopoiesis, attaining its maximum development by the period of sexual maturity.

In young mice the lymphatic nodes consist of aggregations of lymphoblasts and lymphocytes. In adult mice the lymphatic nodes have a clear cortical layer, rounded follicles anf fine medullary meshes. There has been no success in finding foci of erythro-and myelopoiesis in the tissue of lymph nodes.

Thus in the normal physiological condition of the mouse organism hemopoiesis in different age periods is characterized by a different condition of the hemopoietic organs. In the peripheral blood of healthy mice mature elements of white blood are discarded and only as an exception are single myelocytes encountered.

The following conclusions can be drawn on the basis of the study

made:

1. The bone marrow of new-born mice, predominantly erythropoietic with individual megacaryoblasts, has many indifferent cells. Myelopoiesis is poorly evidenced with differentiation to myelocytes. There is active lymphopoiesis in the spleen and lymph nodes. Embryonic hemopoesis is maintained in the liver in the form of foci of erythropoiesis.

2. By the 15th day of life hemopoiesis in bone marrow increases with differentiation of erythroid and myeloid sprouts of hemopoiesis. In the spleen small foci of erythropoiesis and single megacaryocytes appear. Hemopoiesis in the liver is lost.

3. In sexually immature 1-2 month mice active erythro-, myelo- and thrombocytopoiesis with differentiation to mature forms is observed in bone marrow. Small foci of myelopoiesis are peritrabecularly distributed in the spleen, and beneath the capsule and in intersinus spaces - erythro- and thrombocytopoiesis.

4. In sexually mature mice hemopoiesis does not show notable differences from the hemopoiesis of two-month mice.

5. Foci of erythro- and myelopoiesis are not discerned in the lymph nodes of healthy mice.

6. No peculiarities in the structure and development of the organs of hemopoiesis in mice of the high leucosis strain Afb have been discerned with respect to the normal white mice.

LITERATURE

1. Ginzburg, A. M., "Comparative Study of the Blood Picture of Mice of Different Strains," Voprosy onkologii /Questions of Oncology/, Vol. 1, No 4, 1955.
2. Zavarain, A.A., Ocherki evolyutsionnoj gistologii krovi i coyedinitel'noj tkani, /Outlines of the Evolutionary Histology of Blood and Connective Tissue/, Ed. 1, 1945, Ed. 2, 1947, Medgiz. /Gosudarstvennoye izdatel'stvo meditsinkoj literatury - State Publishing House for Medical Literature/.
3. Maksimov, A.A., "Connective and Hemopoietic Tissue", in the book: Mollendorff, W., Handbuch der mikroscopischen Anatomie des Menschen /Handbook of Human Microscopic Anatomy/, Vol. 2, Part 1, 1927.
4. Nikitin, V. N., Atlas kletok krovi sel'skokhozyajstvennykh i laboratornykh zhivotnykh /Atlas of Blood Cells of Agricultural and Laboratory Animals/, Sel'khozgiz /Gosudarstvennoye izdatel'stvo sel'skokhozyajstvennoj literatury - State Publishing House of Agricultural Literature/, 1949.
5. Sakharov, P.P., Metelkin, A.I., and Gudkova, Ye. I., Laboratornyye zhivotnyye /Laboratory Animals/, Medgiz, 1952.
6. Frejfel'd, Ye.M. Gematologiya /Hematology/, Medgiz, 1947.
7. Bloom, W., "The Hemopoietic Potency of the Small Lymphocyte", Folia haematologiae, Vol. 33, 1926, p. 122.
8. Downey, H., Handbook of Hematology, Vol II, 1936, New York.
9. Furth, J., and Strumia, "Studies on Transmissable Lymphoid Leucemia of Mice," Journal of Experimental Medicine, Vol. 53, 1931.
10. Hoffman, G. and Kurzer, Abriss der Anatomie und Physiologie der Laboratoriumstiers /Outline of the Anatomy and Physiology of Laboratory Animals/, 1956, Jena.
11. Jaffe, R. Anatomie und Pathologie der Spontanerkrankungen der kleinen Laboratoriumstiers /Anatomy and Pathology of Spontaneous Diseases of Small Laboratory Animals/, 1931, Berlin.
12. Klinenberger, K., and Carl, W., Die Blutmorphologie der Laboratoriumstiere /Blood Morphology of Laboratory Animals/, 1927, Leipzig.
13. Kock (1931). Cited by A. A. Zavarzin.
14. Petri, S., Morphologie und Zahl der Blutkorperchen bei 7-30g. schweren normalen Laboratoriumsmausen /Morphology and Number of Blood Corpuscles of Normal Laboratory Mice of 7-30g Weight/, 1932, Copenhagen.

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