

Observations on Morphological Changes in Leucocytes of Stored Blood.

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Amoeboid Movement of Leucocytes. There is some doubt whether the leucocytes in stored blood are still alive and functional. After 14 days storage with 2% yeast extract the white cell layer was removed from a test tube of blood (donor E.M.) and mixed with a drop of brilliant cresyl blue, a supra-vital stain that dyes the nucleus. Examined microscopically at about 25° C the damaged cells so evident in smears were not seen; many of the leucocytes showed active amoeboid movement, others appeared dark, granular, and moribund. In blood of this age the polymorphonuclear leucocytes and the medium-sized lymphocytes have about the same size, but can be distinguished by the way they move. The former in old blood have band-like nuclei that change from U-shape to Y and J-shape; a portion may break off and later reunite with the rest of the nucleus. After 2 weeks the few neutrophils left still move with the cytoplasm pushing ahead as described by Kubie and Schultz¹ in the cat. The lymphocytes put out narrow pseudopodia of nuclear material as described by Lewis² and by Bloom.³

Neutrophilic Leucocytes. Sabin and co-workers⁴ found that there is a rapid periodic death of neutrophils "in showers" in the circulating blood, but that this is not true of lymphocytes; about one-fifth of the neutro-

philic leucocytes died every 24 hours. In the yeast extract experiments the neutrophils decreased in the first day by about 25% of their original number in the controls and the total number of lymphocytes was maintained with a loss of only 23% to 28% for over 2 weeks. The polymorphonuclear leucocytes became rounded cells with a band or kidney-shaped nucleus and clear pale cytoplasm as observed by Clark, Clark, and Rex⁵ in lizards and rabbits.

Lymphocytes. Normal size differences among lymphocytes remained throughout a month in stored blood. After one week there were more lymphocytes than neutrophilic leucocytes in all samples (3 different donors). The small lymphocytes with little cytoplasm are highly resistant; they are highly basophilic and resemble mature or senile lymphocytes of circulating blood⁴ and the plasma cells of pathology (Bloom).³ Yeast extract kept the lymphocyte count above that in the controls, with a slight increase in the total count sometimes between the fourth and the sixth day. These increases in leucocyte count may be due to an increment among the lymphocytes by an accretion from changed neutrophils as Lewis² reported. Some of the smallest very dark staining lymphocytes in old stored blood may be naked nuclei such as were seen by Kubie and Schultz.¹

Eosinophiles, as found by other workers, are the most resistant of all the granulocytes. As in the neutrophils, the nucleus becomes rounded or kidney-shaped. The eosinophilic vacuoles enlarge and are very brilliant in blood 2 or 3 weeks old.

¹ Kubie, L. S., and Schultz, G. M., *Johns Hopkins Hosp. Bull.*, 1925, **37**, 91.

² Lewis, W. H., *Johns Hopkins Hosp. Bull.*, 1931, **49**, 29.

³ Bloom, W., in Downey, H., *Handbook of Hematology*, 1938, **1**, 375.

⁴ Sabin, F. R., Cunningham, C. A., Doan, C. A., and Kindwall, J. A., *Johns Hopkins Hosp. Bull.*, 1925, **37**, 14.

⁵ Clark, E. R., Clark, E. L., and Rex, R. O., *Am. J. Anat.*, 1936, **59**, 123.