

ing method of Pappenheim gives very good results. We use a slightly alkaline Giemsa's solution. Short decolorizing of the stained films in 1% NaCO₃ solution brings out the *Bartonella* most distinctly. It is helpful especially if there are only very few parasites present.

The *Bartonella* is first visible free between the red blood cells. It is a small biscuit shaped body composed of two granules kept together by a thin capsule. Later, the bodies become attached to the surface of the erythrocytes. They now have the shape of a slender rod that stains blue and contains two small purple granules on both ends. We could not convince ourselves that the parasites invade the red cells. Mayer, too, says that they are found on the surface of the cells. As many as 30 microorganisms may stick to a single erythrocyte. In this stage, there is a great variety in the size and shape of the *Bartonellae*. Further studies will have to show whether the different forms are stages of a cycle or are due to degeneration.

The problem of cultivating the *Bartonella muris* is being investigated by us.

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² Lauda, E., *Wiener Med. Wch.*, 1927, lxxvii, 772.

³ Nauck, E. G., *Archiv fur Schiffs-und Tropenhygiene*, 1927, xxxi, 322.

⁴ Haan, E., Lauda, E., and Sorge, G., *Klin. Wch.*, 1927, vi, 2240.

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Origin of the Blood Monocyte.*

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The monocytes of the blood were studied in normal rabbits as well as in those in which a monocytosis was produced. This was accomplished by two methods: one was the intravenous injection of lithium carmine, india ink (Higgins') and saccharated iron oxide in various combinations. The other method consisted of infecting the rabbits with *B. monocytogenes* recently isolated by Murray, Webb and Swan.' Some of these infected rabbits were also injected with lithium carmine while others were splenectomized. Two of the latter were slowly injected with one quarter of a cc. of

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Higgins' india ink in a branch of the portal vein so that the carbon would accumulate in the endothelial cells of the liver sinusoids.

The blood of the ear veins of all of the rabbits was studied by supravital, dry and wet smears. The animals were killed at various intervals after the appearance of the monocytosis. At necropsy, supravital, dry and wet smears were made of the various organs and vessels. Portions of all of the viscera and of the main vessels, including the thoracic duct, doubly ligated, were fixed in formalin-Zenker, embedded in celloidin, serially sectioned and stained with hematoxylin-eosin-azure II.

Great numbers of monocytes are produced in the rabbits by both of the methods used. These monocytes are characteristic of the classical descriptions of this cell type as studied with May-Grünwald-Giemsa dry smears. They also have the neutral red rosette which Simpson² and Sabin, Doan and Cunningham³ have shown to be typical of the monocyte of the rabbit. In all of the vessels of the body, and particularly in those of the spleen and liver, the monocytes are always accompanied by large numbers of lymphocytes and monocytoïd lymphocytes. These latter are so numerous and form such a complete series of transition forms between the lymphocytes and monocytes that the conclusion becomes unavoidable that the monocytes develop by individual transformation of the lymphocytes. The supravital neutral red and janus green stain is the most delicate of the methods used in studying the monocytes. It often shows greatly increased amounts of neutral red in cells which appear to be typical lymphocytes when studied in dry smears and sections of organs.

Particular attention was paid in studying the sections to the relation of the vascular endothelium, reticulum endothelium and fibroblasts to the monocytes. I have found no evidence of any of these fixed cells turning into monocytes. This was clearly shown in the rabbits which were injected with vital dyes either with or without concomitant infection with *B. monocytogenes*. In the carminized animals the monocytes do not contain any of this material while the macrophages contain much of it. The development of the fixed histiocytes into free macrophages can be followed in the sections. Although the monocytes do not contain carmin, they can develop into carmin storing macrophages, (Maximow,⁴ Downey and Weidenreich,⁵ Jordan,⁶ Carrel and Ebeling,⁷ Lewis and Lewis⁸).

The evidence of my experiments shows that the monocytes develop from lymphocytes within the blood vessels all over the body and that the sinuses of the spleen are richest in the cells showing this transformation. The numerous mitoses in the lymphocytes in the

lymphoid tissue are sufficient to account for the numbers of new cells necessary to produce the monocytosis; there is no such increase in numbers in any of the fixed cells. When the spleen is removed and india ink injected into the portal vein—these procedures eliminating much of the defense of the rabbit against the intravenous injection of the bacteria—the lymphocytes in the lymph nodes and in the bone marrow (hemocytoblasts) develop neutral red rosettes and, in sections, have markedly constructed nuclei. In this case the lymphocytes show evidences of turning into monocytes before they leave the organs in which they are made and before reaching the blood stream.

I am indebted to Dr. Murray for his kindness in sending me several strains of *B. monocytogenes*.

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² Simpson, M. E., *J. Med. Res.*, 1922, xliii, 77.

³ Sabin, F., Doan, C. A., and Cunningham, R. S., *Carnegie Inst. Contrib. to Embryol.*, 1925, xvi, 125.

⁴ Maximow, A., *Beitr. z. path. Anat. u. z. allg. Path.*, Suppl. v, 1902; *Klin. Wochenschr.*, 1925, iv, 1486; 1926, v, 2193; *Bindegewebe und Blutbildende Gewebe. Handb. der mikr. Anat. of v. Moellendorf*, J. Springer, Berlin, 1927.

⁵ Downey, Hal, and Weidenreich, F., *Arch. f. mikr. Anat.*, 1912, lxxx, 306.

⁶ Jordan, H. E., *Am. J. Anat.*, 1925, xxxv, 105.

⁷ Carrel, A., and Ebeling, A., *J. Exp. Med.*, 1922, xxxvi, 365.

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Preparation of Blood-Free Tissue Proteins.

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It is well known that perfusion and other methods of extraction have hitherto failed to free organs from blood, and for that reason it has been impossible to study the specific proteins in the various tissues or to prepare antisera for them which were not also specific in high dilutions to blood proteins.

In a previous report¹ it was shown that under certain conditions blood-free proteins could be found in the urine which gave precipitation reactions in high dilutions with antisera prepared against liver proteins. It was also suggested² that in the early stages of certain nephritides, the liver leaked its proteins into the blood stream and