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Origin and Proliferation of Thrombocytes in Splenectomized Salamanders.

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In normal salamanders (*Triturus viridescens*) thrombocytes are differentiated chiefly in the spleen. In splenectomized salamanders that were kept in excellent condition for one year after total extirpation of the spleen, thrombocytes were noted in all stages of development in the general circulation. The blood smears made from certain of these animals and stained with Wright's stain have afforded exceptionally favorable material for the study of thrombocytopoiesis.

The question of the origin of the thrombocytes has been at issue for many years. There is little unanimity of opinion concerning the type of cell from which the thrombocyte is derived, or whether its locus of differentiation is intra- or extra-vascular. Among the later investigators may be mentioned Sugiyama,¹ who holds that the thrombocyte is a derivative of the megaloblast, a hemoglobin-containing cell; Gordon,² that it is a senile erythrocyte; Maximow,³ that it is a derivative of the lymphocyte; Hartmann,⁴ that it is a cell genetically and structurally comparable to the megacaryocyte of mammals (as suggested by Wright⁵) which furthermore differentiates in the bone marrow in extravascular location only; Jordan and Speidel,⁶ that it is a derivative of the small lymphocyte which has a minimum of cytoplasm.

Our observations on splenectomized salamanders, however, point unmistakably to the large lymphoid hemoblast (hemocytoblast) as the ancestral cell. In our blood smears prepared according to Wright's technic, the young thromboblasts are easily distinguishable from the young cells of the erythrocyte series. The thromboblasts present a characteristic reddish or reddish-violet fine granulation in the cytoplasm, quite different from the color shades of the proeryth-

¹ Sugiyama, S., Carnegie Inst. of Wash., Pub. 363, Contrib. to *Embryol.*, 1926, xviii, 121.

² Gordon, L., *Virchow's Arch. f. Path.*, 1926, ccxxii, 19.

³ Maximow, A., *Arch. f. mikr. Anat.*, 1923, xvii, 623.

⁴ Hartmann, E., *Fol. haematol. Arch.*, 1925, xxxii, 1.

⁵ Wright, J. H., *J. Morph.*, 1910, xxi, 263.

⁶ Jordan, H. E., and Speidel, C. C., *Am. J. Anat.*, 1929, xlvi, 77.

roblasts, the stages between the hemoblast phase and the true erythroblast.

The megaloblast can be eliminated as the progenitor of the thromboblast. Megaloblasts are plentiful but there is no sign of transition stages toward the thrombocyte. No trace of hemoglobin can be seen at any stage in thrombocytopoiesis. Furthermore, in the salamander it is apparent that the thrombocytes cannot be senile or degenerate red cells. Senile red cells occur in large numbers, but all stages in the process of degeneration can readily be distinguished from the cells of the thrombocyte series.

Of special interest and importance is the observation of thromboblasts in mitosis. As far as we can ascertain thromboblasts have not up to this time been seen in mitosis in the circulation, certainly not in adult animals. In fact, they have rarely been seen in mitosis in any location. A small number of unequivocal examples of thromboblast mitosis have been seen by us in blood from different salamanders. These cells in anaphase contain the fine reddish granulation that is quite characteristic of thrombocytes in this species, and are beyond doubt genuine thromboblasts. This observation would seem to cast grave doubt on all theories which regard thrombocytes as degenerate red cells or other types of cells. The thromboblast in salamander appears to be on the same footing as the erythroblast, eosinophilic granuloblast, and neutrophilic granuloblast; all of these being capable of proliferation by mitosis at the stage of development after the appearance of their specific cytoplasmic differentiation.

The plentiful occurrence of young thromboblasts in the circulation, and of various stages in the process of differentiation, would seem to indicate intra-vascular thrombocytopoiesis in this species. The salamander has no bone marrow (the place where extra-vascular thrombocytopoiesis is described by Hartmann in the toad) and in our experiments was deprived of the spleen. In the lympho-granulocytopoietic capsule of the liver, which is partly analogous to the bone marrow of higher forms, there was no sign of extra-vascular thrombocytopoiesis.