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No Platelet-Destroying Action in Extracts of the Spleen and Urine of Patients with Chronic Thrombopenic Purpura.*

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The spleen of patients with chronic or "essential" thrombopenic purpura is said to contain a substance which destroys blood platelets. By acetone treatment of such spleens, Troland and Lee[1] have obtained an extract which, when injected into the circulation of rabbits, produced a thrombopenia. Extraction by the same method of other organs, or of spleens of patients with diseases other than thrombopenic purpura, yielded negative results.

The spleens of 6 patients under our observation were extracted by the method as outlined by Troland and Lee. Three of these patients had chronic thrombopenic purpura (with characteristic findings in the sternal marrow), one had portal cirrhosis with congestive splenomegaly, and 2 had the splenohepatic syndrome (Banti's Disease) in an advanced form. The spleens were ground immediately after removal from the body, and the grindings were placed in a volume of reagent acetone equivalent to 3 times the weight of the spleen. Extraction was allowed to proceed for at least 2 months. At the end of this time, the acetone was distilled off by heat and suction, and the residue suspended in distilled water. In 2 instances, the acetone extract was divided into 2 portions; in the first portion the acetone was driven off by heat, in the second portion it was removed by vacuum distillation alone.

In one instance, the oily residue after evaporation of the acetone was extracted with ether and the material remaining after evaporation of the ether suspended in distilled water. An amount of water equivalent in volume to about one-half the weight of the extracted organ was used to take up the residue. In addition to splenic extracts, the urine of patients in the acute stage of chronic thrombopenic purpura was injected intravenously into rabbits, either as whole unmodified urine in 10 cc amounts or as chloroform extracts of the entire 24-hour output. Such extracts were prepared by acidifying the urine with 1% hydrochloric acid to pH 3.0, using Congo Red as indicator and extracting with an equal volume of chloroform in a reflux condenser for about 10 hours. The chloroform was then

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1 Troland, C. E., and Lee, F. C., *J. A. M. A.*, 1938, **111**, 221.

evaporated off and the residue suspended in water or cotton seed oil. The cotton seed oil suspension was injected subcutaneously. Twenty-four-hour specimens of urine after evaporation to dryness were also extracted with acetone or ether or 95% alcohol, and after the solvent was driven off, each of the respective residues was suspended in water and injected intravenously. Chinchilla rabbits of about the same age and weight were used in all experiments. Duplicate platelet,[2] red and white cell counts were performed at various intervals, 6 times on the first day of an injection, twice or more times on the second day, and daily thereafter. A few animals received one single injection of the extract, others received as many as 6 injections of 10 cc each, one each day consecutively.

Results. There were no significant changes in the number of platelets following the injection of the extracts of any of the spleens. A slight leukocytosis, lasting approximately 12 hours was the only change. The injection of ether, acetone or chloroform extracts of the urine was followed likewise by no significant fluctuation in the number of platelets. The alcohol extracts of the evaporated urine when injected in saline suspension, produced necrosis of the tissue around the injection site, convulsions and occasionally death. In none of the surviving rabbits were any significant changes in the number of platelets detected.

Summary. Extraction by various methods of the spleen and urine of patients with "essential" thrombopenic purpura, and injection of these extracts into rabbits, failed to disclose the presence of platelet-destroying substances in the spleen and urine of these patients.

2 Tocantins, L. M., *Arch. Path.*, 1937, **23**, 850.