

stimulating blood regeneration, but subsequent experiments made it evident that we were not justified in this assumption. Our most positive findings were with the alcoholic spleen extracts. In every instance this extract prevented blood regeneration and if the injections were continued the animal died.

Our results are particularly interesting in view of those obtained with the feeding of liver by Whipple<sup>1</sup> in the experimental anemias of dogs, and by Minot and Murphy<sup>2</sup> in pernicious anemia. Several explanations of the discrepancies are possible. First, rabbits may not respond to the liver treatment; second, the active substance may have been destroyed by the heat used in drying our alcoholic extracts, though it did not exceed 45° C., or it may have become oxidized; third, our preparations were given by subcutaneous injection, theirs by mouth.

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<sup>1</sup> Robsheit-Robbins, and Whipple, G. H., *Am. J. Physiol.*, 1925, lxxii, 408.

<sup>2</sup> Minot, G. R., and Murphy, W. P., *J. Am. Med. Assn.*, 1926, lxxxvii 470.

### 3542

#### **Effects of Repeated Injections of Toxins of Intestinal Bacteria on Blood Regeneration in Rabbits.\***

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There is doubt as to how much significance should be attached to the belief that certain pathological conditions are due to the absorption of intestinal toxins. Similar doubts are expressed concerning the value of interpretations based on the experimental work done, since most of it has been done with either a pure culture of bacterium isolated from the intestinal tract, or with extracts of the feces. It seems possible that mixtures of bacteria grown in the presence of protein or carbohydrates may produce toxic substances not to be found in pure cultures, and it is also possible that some unknown organism may grow under such symbiotic conditions and produce its specific toxin. The following experiments were conducted to determine the effect of toxins of the intestinal bacteria on blood regeneration.

The culture medium consisted of bouillon containing either finely

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chopped beef, or brain. To half of each series was added 1 per cent soluble starch. One half of the flasks were inoculated with saline emulsions of feces from a patient with pernicious anemia, and the other half with feces from a normal individual. Control uninoculated flasks were incubated for the same periods. The flasks were removed from the incubator at intervals of 1 week, 2 weeks and 4 weeks, and the contents passed through Berkefeld filters. Only those filtrates found to be sterile by aerobic and anaerobic methods were used. The filtrates were kept in the ice-box, but even then the toxicity soon decreased.

Rabbits were used in all experiments. The filtrates were given intravenously in amounts of 0.25 to 0.5 cc., diluted 1 to 3 with salt solution, or subcutaneously in 1 cc. doses. The injections were given at intervals of 48 hours. Blood examinations were made at frequent intervals both before and after the injections were started.

All the animals developed an anemia of the secondary type, beginning after the third or fourth injection. The lowest count observed was 1,530,000. At times remissions occurred in spite of the continued injections, but they were usually of brief duration. Only an occasional nucleated red cell was observed. At times the reticulated cells were slightly increased, but for the most part they remained within the normal limits. The white count remained low. Very little, if any, change was observed in the size and shape of the red cells. Practically no immunity was produced, as doubling the dose of toxin would frequently kill animals which had received many previous injections.

### 3543

#### Effect of Bile, Sodium Salts of Bile Acids, and Unsaturated Fatty Acids on Bacteriophagic Action.

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Previous investigators record different observations on the effect of bile on bacteriophagic action. Hauduroy,<sup>1</sup> Jong and Hauduroy,<sup>2</sup> and d'Herelle<sup>3</sup> found that bile was decidedly inhibitory, whereas Caublot,<sup>4</sup> in observations extending over a period of two years, obtained no evidence of an inhibitory action of bile on bacteriophage.

In the studies, recorded here, of the action of bile on bacterio-