days old they are similarly put on the milk diet. The Hb per cent and the number of red cells decrease more rapidly than they did in the first generation. When the low levels are reached some of the animals die, but the majority show a tendency to a slow spontaneous recovery. Hb values of less than 4 gm. per cent are fatal in the majority of cases.

The red cells in this type of anemia vary considerably in size and shape and many of them show polychromatophilia. The average corpuscular volume is 35 cu. microns as compared with 51 cu. microns for the normal red cell, and the saturation is low, 30% as compared to 40% in the normal cell. During the spontaneous recovery the stroma increases more rapidly than the Hb so that in the later stages the saturation is further diminished. An increased resistance of the red cells to hypotonic solutions is a striking and constant feature of this anemia.

Measured doses of radiant energy from quartz mercury vapor and flaming C arc lamps in various quantities and with varying time intervals between them were administered. In some experiments the irradiation was begun as soon as the animals were placed on the milk diet; in others not until a severe anemia had developed. The following groups served as controls: irradiated rats on the stock diet, non-irradiated rats on the stock diet, and non-irradiated rats on the milk diet. The differences in Hb and in red cells in the irradiated and non-irradiated groups were small, being slightly higher in the irradiated groups. The irradiated animals often present a more healthy appearance, and any slight increase in blood levels is referred to an improvement in the general nutritional state of the animal, rather than to direct action on the hemopoietic system. Excessive irradiation in some cases retarded growth but was not observed to have any detrimental effect on the blood.

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Action of Bacteriophage upon Production of in vivo Prepared Toxic Substance of Bacillus Typhosus.*

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In a previous communication[1] we reported the results of the inoculation of guinea pigs with the toxic material obtained from the

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[1] Harris, W. H., and Larimore, O. M., *J. Exp. Med.*, 1928, xlviii, 885.

exudate of experimental typhoid peritonitis. The filtered toxic material produced lesions closely simulating those of human typhoid fever and the animals died after 3 or 4 inoculations. The toxic substance produced in this manner was found to be of rather low potency. Single injections of from 3 to 5 cc. failed to produce death although the animals became sick and showed marked reactions to the inoculations.

It was thought that the employment of typhoid bacteriophage in conjunction with this *in vivo* method may serve the purpose of yielding a more toxic substance. It has already been shown that the action of bacteriophage *in vitro* has but little if any effect upon the toxicity of certain microorganisms. It was, however, thought that since the toxin employed previously by us was prepared *in vivo*, the presence of this lytic agent in the field of conflict between the host and the invading microorganism might provoke the liberation of a poison of more highly toxic nature.

Forty white mice and 24 guinea pigs were employed in the experiments. Typhoid culture 7D, a stock strain which had produced an accidental typhoid infection in one of us (Larimore) approximately a year ago and had been recovered from the blood stream, was employed for the inoculation. The typhoid bacteriophage used consisted of 2 separate "strains" kindly sent to us by Dr. D'Herelle and Dr. Bronfenbrenner.

Three sets of experiments were conducted: Series I consisted of the production of typhoid peritonitis in the mouse with the presence of varying amounts of phage introduced simultaneously with the infecting microorganism. The exudate was procured and filtered and this filtrate inoculated intraperitoneally into the other mice. Control animals in which no phage was introduced were run parallel and the filtered exudate injected intraperitoneally into other mice in similar doses to the previous animals. While the animals became sick they did not die from the single injection.

In Series II, guinea pigs were inoculated intraperitoneally with doses of 2, 3, 4 and 5 cc. of typhoid culture 7D and varying amounts of the bacteriophage were introduced one hour before, simultaneously and one hour after the infecting typhoid injection. Control animals without phage were employed. From these animals, dead of typhoid peritonitis, the peritoneal exudate was procured and filtered. These filtrates were inoculated intraperitoneally in amounts varying from 1 to 5 cc. In 4 of the animals in which phage had been used in the production of toxin, marked toxic effect as shown by twitchings and transient partial paralysis of the posterior extremities were noted. These effects were transient and the animals recovered over

night. The remaining animals including the controls showed loss of activity and were manifestly sick but were lively after 24 hours.

In Series III, guinea pigs were inoculated in a similar manner as in Series II but the recovered filtrate material was inoculated into white mice. In this manner comparatively large doses could be used for a very small animal but the results obtained did not indicate any difference in degree of potency of toxin in the exudate procured from animals with and without phage.

A group of animals, both white mice and guinea pigs, were inoculated with fatal doses of typhoid suspension and another group with phaged doses of typhoid bacilli. In these animals no differences in the period of time of death production was noted. It should be stated that the phaged suspension in these latter experiments was not filtered, whereas, in the other 3 series of experiments the bacteriophage employed was always filtered and tested for sterility before it was employed.

These experiments indicate that the employment of typhoid bacteriophage in conjunction with our previously reported process, has no significant effect upon the potency of toxic substance procured from the typhoid bacillus through this means.

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Action of Bacteriophage in Experimental Typhoid Peritonitis.*

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While engaged in certain other experimental work, we had the opportunity to observe the effect of *Bacillus typhosus* bacteriophage upon peritonitis produced in guinea pigs and white mice, by means of the intraperitoneal injection of *Bacillus typhosus*. In view of the apparent possibilities of bacteriophage therapy, it was thought that the observations herein noted in the experimental animals, should be recorded.

A race of bacteriophage virulent for *Bacillus typhosus* was used, which was isolated and kindly furnished us by Professor D'Herelle and which in his hands had been found potent or virulent for several heterologous strains of typhoid bacilli.

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