mined whether toxin so treated retains its antigenic potency. Lowenstein's[2] tetanus toxin, treated with 0.2% formalin and subjected to ultra-violet light was atoxic and antigenic, but since it is now well known that formalin alone renders certain toxins atoxic while preserving their antigenic powers, the effect of the irradiation in his experiments is uncertain. An experiment was therefore undertaken using as an immunizing agent tetanus toxin rendered atoxic by ultra-violet light. A mixture of 2 tetanus toxins after titration was diluted to contain 1 M.L.D. in each 1 cc. This was irradiated with "C" carbons (National Carbon Co.) at a distance of 25 cm. in the apparatus described in a previous publication,[3] keeping the toxin below room temperature by a current of cool air.

A 2-minute irradiation period failed to destroy the toxin completely, as about half of the inoculated animals developed late tetanus. A further irradiation of 2 minutes rendered the material atoxic. Using not more than a quantity originally containing 1 M.L.D, 9 guinea pigs were given 5 subcutaneous injections of this irradiated toxin at 6 or 7 day intervals and were inoculated 3 weeks later with freshly titrated tetanus toxin in doses of from 1 to 10 M.L.D.'s. No signs of tetanus developed. The period of observation was 45 days.

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Hematopoietic Function in Avitaminosis. IV. Further Studies of Vitamin A Deficiency.*

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In a previous publication[1] we have suggested the occurrence of a secondary anemia in vitamin A deficiency. Our results, however, were not conclusive. We, therefore, continued this investigation with 30 more animals, 18 of which received our vitamin A deficient ration 1749.† Twelve of these rats were allowed a modification of

[2] Lowenstein, E., *Z. Exp. Path. u. Ther.*, 1914, **15**, 279.

[3] Perkins, R. G., and Welch, H., *J. Prev. Med.*, 1929, **3**, 363.

* Research Paper No. 198, Journal Series, University of Arkansas.

[1] Sure, B., Kik, M. C., and Walker, D. J., *J. Biol. Chem.*, 1929, **83**, 375.

† Composition of ration 1749 is as follows: Casein (hot-alcohol extracted), 20; N. W. yeast, 10; McCollum's salts No. 185 (2), 4; lard, 2; dextrin, 64. This ration was irradiated for 30 minutes with a mercury quartz vapor lamp to insure an adequacy of vitamin D.

this diet, so that one to 2% of lard was replaced by equivalent amounts of butterfat, in order to prevent sudden deaths from pneumonia frequently associated with this avitaminosis. In addition, we have studied the effect of vitamin A deficiency on the differential leucocyte count in 9 females and 9 males. In the females the disease was followed with daily examinations of the vaginal smears according to the technique of Evans and Bishop.[3] Daily records were kept of food consumption, and in the majority of animals records were also kept of the daily water intake. Bleedings were performed on each animal twice weekly and determinations were made of hemoglobin and erythrocyte counts. Specific gravity determinations were also made, in order to secure information on blood concentration. The experimental period ranged from 68 to 173 days.

To summarize our results, we found no noteworthy disturbance in hematopoietic function during various stages of this avitaminosis, as characterized by the severity of ophthalmia and loss of body weight, nor any changes in the differential leucocyte count during the onset, and remission produced by vitamin therapy. Our results, therefore, do not substantiate the findings of Koessler and coworkers.[4] Although secondary anemias have been encountered in keratomalacia in man,[5] the investigators admit that other complications or infections associated with this disease may be the influencing factors rather than this avitaminosis.

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Hematopoietic Function in Avitaminosis. V. Vitamin D Deficiency.*

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In a study of the secondary anemia of childhood, Happ and Evans[1] pointed out that 9 of their 10 patients had rickets, although

[2] McCollum, E. V., and Simmonds, N., *J. Biol. Chem.*, 1918, **33**, 63.

[3] Evans, H. M., and Bishop, K. S., *J. Metabol. Res.*, 1922, **1**, 10.

[4] Koessler, K. K., Maurer, S., and Loughlin, R., *J. Am. Med. Assn.*, 1926, **87**, 476. Koessler, K. K., and Maurer, S., *J. Am. Med. Assn.*, 1927, **89**, 768.

[5] Keefer, C. S., and Yang, C. S., *Nat. Med. J., China*, 1929, **15**, 419.

* Research Paper No. 199, Journal Series, University of Arkansas.

[1] Happ, W. M., and Evans, F. A., *Johns Hopkins Hosp. Bull.*, 1922, **33**, 7.