

1. The injection of antiprolan protects a rat for 8 days against the effect of prolan. This process especially points to a passive immunization.

2. Antiprolan is not to be found in the albumin but in the globulin of the serum, namely in certain fractions of the globulin (pseudoglobulins)² thus resembling some of the antibodies.

3. The reversibility of the antiprolan effect also speaks in favor of an immune-biological reaction.†

While our experiments suggest that antiprolan is neither a hormone-like nor a ferment-like body, yet on the other hand we find some, if not all, properties corresponding to those of an immune-biological body. It is possible that here we have to deal with a new kind of body approaching very closely the immune-bodies without producing the *vitro*-reactions typical of those bodies.

Summary. The inactivation of prolan by antiprolan is a reversible process, since prolan and antiprolan may be released from a neutral prolan-antiprolan mixture and thereby reactivated.

The assumption is made that antiprolan is neither an (anti)hormone *strictu sensu* nor a ferment, but is possibly a new kind of factor approaching very closely the immune-bodies to which it is in some respects quite similar.

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Effect of Vitamin D on Growth of Tubercle Bacilli.*

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It was shown^{1, 2} that feeding irradiated cream or oil concentrates moderately high in vitamin D to tuberculous guinea pigs produced no change in the course of the disease.

To control this observation further it was decided to observe the

† In a previous report³ we expressed the supposition that antiprolan destroys prolan. The experiments reported here, however, show that this supposition was wrong for now we can prove the reversibility of the reaction.

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¹ Loewen, David F., and Oatway, Wm. H., *Am. Rev. Tuberc.*, 1936, **33**, 733.

² Steenken, Wm., Jr., and Baldwin, E. R., *Am. Rev. Tuberc.*, 1937, **35**, 656.

effect of the addition of crystalline vitamin D in propylene glycol† upon the growth and virulence of variants from the same parent strain H₃₇.

To small flasks containing 20 cc. of Proskauer and Beck's synthetic medium‡ crystalline vitamin D solution was added in 4 amounts ranging from 0.05 to 0.2 cc. (100 to 400 U.S.P. units). As a control propylene glycol was added in the same concentrations to the basic medium.

Control and experimental media were seeded with Ra and Rv³ variants of the H₃₇ strain. The fully virulent Rv variant was the organism used for infecting all animals in the previous experiments. Both variants were carried through 3 generations on the 2 media, being transferred at 21-day intervals. Growths characteristic of each variant were observed in each generation. The Rv growth was veil-like and spreading whereas the Ra was heaped up and dense with a clear-cut periphery.

At the end of the third 21-day interval of transfer, suspensions were made of the organisms from each flask so that one cc. contained 0.5 mg. dry weight. Each of these suspensions was divided into 2 parts. One part was plated on gentian-violet-egg media for colonial study. At the end of 6 weeks, the colonies were compared and no alteration of their original form was discovered. The other part was divided and inoculated intratesticularly into 2 guinea pigs.

Forty days after inoculation a control pig (Rv) died; its lungs, liver, spleen and glands were riddled with tuberculosis.

On the same day one of each pair was killed. All of the animals receiving the Rv variant had generalized tuberculosis, whereas all of the pigs receiving the Ra variant had only local caseous lesions in the testicle.

Fifty days from the date of inoculation an Rv pig (200 vitamin D units) died of generalized tuberculosis. All remaining Ra and Rv pigs were killed and pathological changes similar to those in previously reported series were observed.

Summary. Crystalline vitamin D (100-400 units) when added to Proskauer and Beck's medium did not change the virulence or

† Supplied to us through the courtesy of Winthrop Chemical Co. (10,000 U.S.P. units per gm.).

‡ Monopotassium phosphate	5.0 gm.
Asparagin	5.0 "
Magnesium sulphate	0.6 "
Magnesium citrate	2.5 "
Glycerol	20.0 cc.
pH	7.4

³ Steenken, Wm., Jr., *PROC. SOC. EXP. BIOL. AND MED.*, 1935, **33**, 253.

growth characteristics of either the Ra or Rv variants of strain H₃₇ *M. tuberculosis*.

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Nitrogen, Sulfur, Sodium, Potassium and Chloride Metabolism in Vitamin B₁ Deficient Rats.

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Metabolic studies in vitamin B₁ deficient animals have been largely limited to studies on carbohydrate metabolism and respiration of the central nervous system and other tissues.

To our knowledge no studies on the metabolism of the usual food elements and electrolytes have been made in vitamin B₁ deficient animals.

Twenty-four male and female albino rats (Wistar strain) were used in the experiments. They were kept in groups of 4 in metabolism cages, as previously described.¹ The diet consisted of purified casein 18%, Osborne-Mendel salt mixture 4%, butterfat 8%, cod liver oil 2%, corn starch 53%, and autoclaved baker's yeast 15%. In order to be able to differentiate clearly between the effects of vitamin B₁ deficiency and of inanition, paired feeding was resorted to. The food consumed by the controls consisted of the same ingredients as that given to the experimental animals, except that unautoclaved yeast was used. They received an amount of food corresponding to that eaten by the experimental animals during the preceding week. Distilled water *ad lib.* was provided from automatic glass fountains.

Urine and feces were collected twice a week. The urine was analyzed for total nitrogen (Kjeldahl), urea,² ammonia (Folin), uric acid,³ creatine and creatinine,⁴ total sulfur,⁵ total and inorganic sulfates (Folin), sodium and potassium,⁶ and chloride (modified Volhard-Harvey method). Feces were analyzed for total nitrogen, total sulfur,⁷ and chloride (open Carius method). Food was analyzed

¹ Sandberg, M., and Perla, D., *J. Exp. Med.*, 1934, **60**, 395.

² Van Slyke, D. D., and Cullen, G. E., *J. Biol. Chem.*, 1914, **19**, 211.

³ Benedict, S. R., and Franke, E., *J. Biol. Chem.*, 1922, **52**, 387.

⁴ Folin, O., *J. Biol. Chem.*, 1914, **17**, 469.

⁵ Benedict, S. R., *J. Biol. Chem.*, 1909, **6**, 363.

⁶ Smith, G. F., and Ross, G. F., *J. Am. Chem. Soc.*, 1925, **47**, 1020.

⁷ Neumann, A., and Meinertz, J., *Z. physiol. Chem.*, 1904-5, **43**, 37.