

Circulating Antibodies in Vitamin Deficiency States. Pantothenic Acid Deficiency.* (17338)

PETER P. LUDOVICI, A. E. AXELROD, AND BETTINA B. CARTER (Introduced by Ralph R. Mellon.)

From the Institute of Pathology, The Western Pennsylvania Hospital, and the Department of Chemistry, University of Pittsburgh, Pittsburgh, Pa.

In a previous paper, we have reported that pantothenic acid-deficient rats show a severe impairment of antibody response.¹ This finding was in contrast to the observation of Stoerk, Eisen, and John that pantothenic acid deficiency did not affect the level of circulating antibodies.² We have attempted to explain this difference in results on the basis that the sheep erythrocytes employed by Stoerk *et al.* were a poorer antigenic stimulus for hemagglutinin production in the rat than the human red blood cells employed by us. Differences in the immunization procedures were also noted. Stoerk, on the other hand, believes that this difference may be attributed to the fact that we were dealing with a relatively more severe pantothenic acid deficiency.³ Our failure to utilize inanition controls for the pantothenic acid deficient rats, as well as the supposed inadequacy of "paired-fed" controls as contrasted with "paired-weighted" controls were also stressed by this author.^{3,4} Some of these points have already been discussed.⁵

The present experiments were planned to determine the effects of varying degrees of pantothenic acid deficiency upon antibody production in the rat utilizing "paired-weighted" animals as inanition controls.

Experimental. Fifty-eight male weanling albino rats of the Sprague-Dawley strain were

distributed into 3 groups as indicated in Table I. The animals were housed individually in wide-meshed, screen-bottom cages and weighed daily. Daily food consumption records were taken. All rats were fed a basal diet of the following percentage composition: sucrose, 56.76; Labco "vitamin-free" casein, 25.00; salts,⁶ 4.00; cod-liver oil, 2.00; hydrogenated vegetable oil, 10.00; corn oil, 2.00; choline chloride, 0.20; 1-inositol, 0.03; p-aminobenzoic acid, 0.01; and 2-methyl-1, 4-naphthoquinone, 0.001. All rats received additional vitamins in the form of a daily pill. Each of the pills given to the control animals supplied the following vitamins: thiamin, 40 γ ; riboflavin, 60 γ ; pyridoxine, 50 γ ; nicotinic acid, 100 γ ; biotin, 1 γ ; folic acid, 1 γ ; and calcium pantothenate, 300 γ . For the pantothenic acid deficient rats, calcium pantothenate was omitted from the pill. The deficient rats were fed the basal diet *ad libitum*. However, each control rat was fed a restricted amount of the basal diet sufficient to maintain its weight equal to that of its pantothenic acid-deficient partner (paired-weighting). After 3, 5, and 7 weeks on experiment, the rats of each group were immunized. A 10% suspension of washed Group O, Rh positive human erythrocytes in physiological saline was injected intraperitoneally as antigen. An initial dosage of 0.5 ml of the red blood cell suspension was followed by 2 one ml injections on alternate days. Five days after the final injection the rats were bled from the heart under ether anesthesia and the sera tested for agglutinin titers as described previously.¹ Immediately after bleeding, the thymus of each rat was removed and weighed.

Results. The individual hemagglutinin titers are recorded in Table II. It is evident that

* This investigation was supported in part by a grant from the Division of Research Grants and Fellowships of the National Institute of Health, U. S. Public Health Service.

¹ Axelrod, A. E., Carter, B. B., McCoy, R. H., and Geisinger, R., *Proc. Soc. Exp. Biol. and Med.*, 1947, **66**, 137.

² Stoerk, H. C., Eisen, H. N., and John, H. M., *J. Exp. Med.*, 1947, **85**, 365.

³ Stoerk, H. C., *Nutrition Rev.*, 1948, **6**, 191.

⁴ Stoerk, H. C., private communication.

⁵ Axelrod, A. E., Carter, B. B., and McCoy, R. H., *Nutrition Rev.*, 1948, **6**, 351.

⁶ Jones, J. H., and Foster, C., *J. Nutrition*, 1942, **24**, 245.

CIRCULATING ANTIBODIES IN PANTOTHENIC ACID DEFICIENCY

TABLE I.
Summary of Growth, Food Consumption, and Thymus Weight Data.*

Type	No. of rats	Body wt†		Thymus‡ wt	Daily food consumption†
		Initial	Final§		
Group I*					
Control	10	42	83	195	4.4
Pantothenic acid-deficient	10	42	84	116	4.8
Group II*					
Control	10	43	83	140	4.0
Pantothenic acid-deficient	10	43	84	78	4.4
Group III*					
Control	9	42	94	90	3.9
Pantothenic acid-deficient	9	43	96	69	4.7

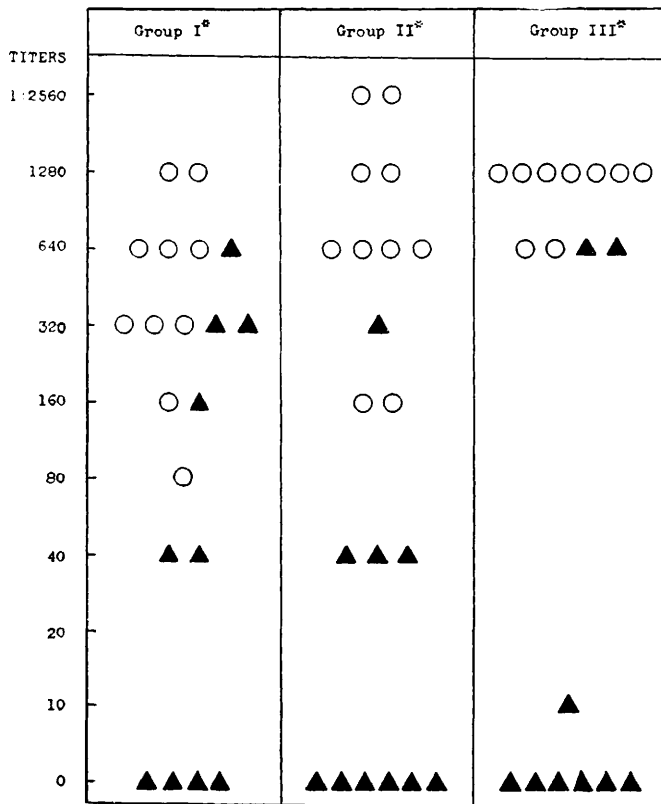
* Groups I, II, III were fed basal diet 3, 5, and 7 weeks respectively prior to immunization.

† Group average in g.

‡ Group average in mg.

§ At the time of bleeding.

Table II.
Individual Hemagglutinin Titers.



* Groups I, II, III were fed the basal diet 3, 5, and 7 weeks, respectively, prior to immunization.

○—Control animals.

▲—Pantothenic acid-deficient animals.

the antibody response was decreased in the pantothenic acid-deficient rats of all groups. In no instance, did the titer of a deficient rat equal that of its "paired-weighed" control. The deleterious effect of a mild pantothenic acid deficiency is evidenced by the decreased titers of the animals in Group I which were on experiment for only 3 weeks prior to immunization. This observation lends no support to the explanation offered by Stoerk for the discrepancy in the findings of the two laboratories. The comparatively high antibody content of the "paired-weighed" controls is further evidence against the role of inanition in the decreased antibody response of pantothenic acid-deficient rats.

In the course of further studies on the mechanism of action of pantothenic acid we have determined the antibody response in 44 pantothenic acid-deficient rats and 29 suitable controls. The results obtained are in excellent

agreement with those recorded in this as well as in a previous paper.¹ All of the evidence to date firmly implicates pantothenic acid as a vital factor in the antibody response to the antigenic stimulus of human erythrocytes.

The thymus weights of the pantothenic acid-deficient rats were lower than those of the control group (Table I). These findings are in agreement with those made by Stoerk *et al.*²

Summary. (1) Hemagglutinin production in response to inoculation with human erythrocytes has been investigated in rats fed a pantothenic acid-deficient diet for 3, 5, and 7 weeks. "Paired-weighed" animals served as inanition controls.

(2) Marked impairment of antibody response was observed in all pantothenic acid-deficient groups.

Received July 27, 1949. P.S.E.B.M., 1949, **72**.

Selective XYZ Factor in C57 Black Mammary Carcinoma Eo771. (17339)

ALBERT E. CASEY, GORDON L. ROSS, AND R. REVAE LANGSTON

*From the Departments of Pathology of the Baptist Hospital of Alabama, and the Medical College of Alabama, Birmingham, Ala.**

The XYZ phenomenon may be defined as the increased incidence and more rapid increment of local and metastatic transplanted tumors following the prior injection in the same or distant sites of an extract from the homologous tumor.¹⁻⁴ The factor in the

Brown-Pearce rabbit tumor responsible for the phenomenon is selective, filtrable (Berkefeld "V") and thermolabile (56°C);⁵⁻⁷ that in Bashford mouse mammary carcinoma^{6,8,9} is likewise highly selective or specific.¹⁰⁻¹²

* The work was aided by grants from the American Cancer Society and the Damon Runyon Fund through the Committee on Growth of the National Research Council. The assistance of Joe Norman, Bruce A. Elrod, Robert C. Rea, Erma L. Salter, Gertrude Hampton, Irene Hartsfield, Betty Baker, Russell Laster, and Ward Talley of the laboratory staff of the Baptist Hospital in the care of the animals, recording of data and preparation of histologic material is acknowledged with thanks.

¹ Casey, Albert E., *Proc. Soc. Exp. Biol. and Med.*, 1932, **29**, 816.

² Casey, Albert E., *Am. J. Cancer*, 1934, **21**, 760.

³ Casey, Albert E., *Am. J. Cancer*, 1934, **21**, 776.

⁴ Casey, Albert E., *Cancer Research*, 1941, **1**, 134.

⁵ Casey, Albert E., *Am. J. Cancer*, 1936, **26**, 276.

⁶ Casey, Albert E., and Moragues-Gonzales, Vincent, *Am. J. Cancer*, 1940, **38**, 59.

⁷ Casey, Albert E., Meyers, Lucille, and Drysdale, George R., 1948, **69**, 570.

⁸ Haaland, M., *Proc. Roy. Soc. London*, 1910, **82**, 293; *Lancet*, 1910, **1**, 787.

⁹ Leitch, A., *Lancet*, 1910, **1**, 991.

¹⁰ Casey, Albert E., *Proc. Soc. Exp. Biol. and Med.*, 1933, **30**, 674.

¹¹ Casey, Albert E., *Proc. Soc. Exp. Biol. and Med.*, 1933, **30**, 1025.

¹² Casey, Albert E., *Proc. Soc. Exp. Biol. and Med.*, 1934, **31**, 663.