

trial. The vitamin-bearing substances were the only noteworthy sources of either fat or carbohydrate, and supplied 4-8 per cent. of the food eaten. Whether rats will attain adult size and normal function on such diets, furnishing protein as the almost exclusive source of energy and tissue substance, remains to be determined further. If future experiments prove as successful as those here described various problems of nutrition and physiological function can be approached from new experimental standpoints.

80 (1662)

The addition of yeast to a milk diet.

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The experiments were made on white rats, one group of rats being fed a diet of pasteurized milk and a second group being fed a milk and yeast diet. The rats receiving the yeast made more satisfactory growth gains than did the rats receiving no yeast. Inasmuch as milk has been shown to be low in the water-soluble "B" vitamine, which is present in high concentration in yeast, it would seem that yeast may be found to be an important dietary adjunct for use in baby feeding.

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The rate of fixation of complement at various temperatures.

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This investigation embraces three types of complement-fixing substances: (1) those elicited in rabbits due to injection of purified proteins; (2) those produced in the same animals due to injection of bacteria, and (3) those found in the serum of syphilitic patients. The antigens employed in the first two cases were specific, while

in the case of the syphilitic sera, non-specific; four different Wassermann antigens being employed with each serum. Three fixation temperatures—water-bath, room and ice-box—were resorted to. Some phases of this investigation are still in progress and in this preliminary report, the work with the purified proteins only will be reported, although our findings indicate that the rate of fixation of complement is the same, no matter what type of fixing antibody is used.

Two purified proteins were employed: Edestin obtained from hempseed and phaseolin obtained from the kidney bean. These were kindly furnished by Dr. Thomas B. Osborne. Two rabbits were immunized with edestin and two with phaseolin. In order to elicit quantitative differences in the antibody production in the rabbits, four modes of immunization were resorted to. The edestin rabbits were injected intravenously according to "Immunization Methods No. 1 and No. 2," respectively, described by Kahn and McNeil in another paper.¹ The phaseolin rabbits were injected intraperitoneally. One rabbit received 100, 150, 200, 250 and 300 mgm. of phaseolin at 48-hour intervals, and the other 100, 150 and 200 mgm. of this protein at 24-hour intervals.

The complement fixation experiments were carried out in one tenth quantities of regular Wassermanns, otherwise in the usual manner, with 2 units of complement, 2 units of amboceptor and 0.1 c.c. of a standard 5 per cent. suspension of sheep-cells. The respective antigens were prepared by weighing out 10 mgm. of the protein and dissolving these in 10 c.c. of $N/1000$ NaOH to which was added 0.05 c.c. of $N/10$ NaOH. The alkali was necessary in order to get the proteins in solution. One c.c. of this protein solution was added to 9 c.c. of saline and 0.1 c.c. of this final solution (0.01 mgm. of the protein) was used in the tests. The serum dilutions employed in the tests were the following: 0.01 c.c., 0.007 c.c., 0.004 c.c., 0.003 c.c., 0.002 c.c., 0.001 c.c., 0.0005 c.c., 0.0003 c.c., and 0.0001 c.c.

After establishing the presence of specific complement fixing antibodies in the rabbit's sera by preliminary tests, fixation experiments were carried out with the serum dilutions indicated above, varying both the lengths of time and the temperatures of fixation.

¹ *J. Immunol.*, 1918, iii, 281.

Thus the first series were run both at water-bath and ice-box temperatures with the following fixation periods: 15 minutes, 30, 45, 60, 90 and 120 minutes. In view of the fact that complement has a tendency to be destroyed when exposed for too lengthy periods in the water-bath, the fixation tests at this temperature were not extended beyond 2 hours. Neither were the fixation periods extended beyond this time when the fixations were at room temperature. In the case of the ice-box, however, the fixation periods were continued for 3, 4, 5 and 6 hours, and occasionally longer. At the end of each fixation period, standard amounts of sheep cells and amboceptor were added to each set and placed in the water-bath to determine whether or not the complement had been "fixed."

The results indicate:

(1) That the phenomenon of fixation of complement goes on equally well at water-bath, room or ice-box temperature.

(2) That from 50 to 75 per cent. of fixation takes place during the first hour and that fixation is completed in the neighborhood of 4 hours at ice-box temperature.

82 (1664)

**The quantitative relation between complement and
complement fixing antibody.**

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In the course of investigation on precipitin and complement fixing antibodies produced by injections of edestin, it was observed that, while the serum of a rabbit immunized with this protein showed the presence of precipitin antibodies, it did not show any complement fixing antibodies when employing the usual 2 units of complement in the fixation tests. It appeared reasonable at first to accept this finding as evidence of the lack of relation between these two types of antibodies. It seemed, however, that possibly the employment of 2 units of complement in the tests might give a sufficient excess of this ingredient to render a serum