

third dose of the initial course of vaccination. There was, however, a rather wide variation in the agglutination titer for the individual cases. The titers were the same regardless of whether vaccines or living suspensions prepared from the homologous or heterogeneous strains of the organisms were used as agglutinin, provided their concentrations were the same.

After an interval of from 4 to 19 weeks after the first course of prophylactic inoculations, when the agglutinin titer of the blood had declined considerably, bilateral injections of 1.5 cc of vaccine each were given in the deltoid regions of all the 5 infants. The results shown in Table I indicate that these injections were invariably followed by a very marked increase of agglutinin within one week far above the immediate response to the initial vaccination. After another interval of from 15 to 28 weeks, the effect of such a stimulating dose was tried for the second time with more or less similar results. Although we are aware of the fact that agglutinins do not necessarily represent immunity, this observation suggests that if a stimulating dose of pertussis vaccine is given to an already vaccinated child shortly after exposure to infection, it may increase the amount of antibody during the incubation period of the disease so as to enhance the chance of protection.

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A Dialyzable Component of Complement.

BACON F. CHOW AND SAMUEL H. ZIA.

From the Department of Biochemistry and the Department of Bacteriology and Immunology, Peiping Union Medical College, Peiping, China.

When serum of a normal guinea pig is dialyzed against a normal saline solution with a cellophane tube* the hemolytic activity of the solution decreases rapidly. A prolonged dialysis will destroy almost all of the original activity. We have now found that this loss of activity is due to the loss of one of the components of complement which passes through the membrane into the saline solution, and that the original activity may be almost completely restored by the addition of the dialyzable to the non-dialyzable fraction.

A typical experiment of the dialysis is as follows: 10 cc of fresh guinea pig serum containing 80 units of complement per cc were

* Obtainable from DuPont Cellophane Co.

poured into a cellophane tube both ends of which were closed with rubber stoppers and tightened with rubber bands. The sealed tube was then suspended in a graduated cylinder containing 100 cc of a cold normal saline solution. The dialysis was allowed to take place at about 5°C. After 24 hours, the bag was opened and a half cubic-centimeter of serum was pipetted out, and it was found to contain 50 units per cc. The rest of the serum was again dialyzed with another 100 cc portion of a normal saline solution. After 24 hours another half cc of the serum was taken out and its hemolytic activity was found to be 20 units per cc. The rest of the serum was again dialyzed with a fresh volume (100 cc) of the saline solution. Two days later the bag was opened and the dialyzed serum now contained less than 10 units per cc. The total volume of serum after dialysis was about 9 cc. Therefore the decrease of activity was not due to the dilution of serum. A control sample of the same guinea pig serum was stored without dialysis in another cellophane tube and kept at about 5°C. Its complement titer dropped from 80 to 60 only in 4 days.

It was of interest to ascertain whether the loss of activity during dialysis is due to the separation of the components of complement or to some chemical reaction such as oxidation. We have found that the original complementary activity may be restored by combining the dialyzed serum and the dialyzate. This was done as follows: To 0.5 cc of the dialyzed serum diluted 30-fold with 0.35% NaCl were added varying amounts of dialyzate, and enough normal saline solution to make a total volume of 2 cc. Five-tenths cc of hemolysin (2 units/cc) and 0.5 cc of 2% sheep cells were then added. After 30 minutes at 37°C readings were made. The results of a representative experiment are presented in Table I.

TABLE I.
Restoration of Alexin Activity by Addition of Varying Amounts of Dialyzate to 0.5 cc of 1:30 of Dialyzed Serum.

Dialyzate obtained after hr	Volume of dialyzate in cc						
	1.5	1.2	1.0	0.8	0.6	0.5	0
24	4	4	4	4	2	—	—
48	4	4	4	4	4	—	—

4 = means complete hemolysis.

2 = " partial "

— = " no "

From the results given in Table I it may be noticed (1) that since a half cc of the dialyzed serum diluted 30 times with a normal saline

solution was reactivated by the addition of a small amount of the dialyzable fraction, the activity of the combined fractions therefore corresponded to at least 60 units of complement per cc, and (2) that the amount of the dialyzable component necessary for the reactivation was considerably less than that present in the original serum. As little as 0.8 cc of the greatly diluted solution was sufficient to restore activity. Therefore, one may conclude that the dialysate is a component of complement.

It is well known that certain components of complement may be altered or inactivated by the treatment of yeast,¹ ammonium hydroxide,² or oxidation with iodine.^{3, 4} It was of interest to ascertain whether the dialyzable fraction corresponds to one of these fractions. We have tried to reactivate the complementary activity of sera treated with each one of the above 3 reagents, by adding to them varying amounts of the dialysate; the results have been consistently negative. Furthermore, if the sera inactivated yeast, ammonium hydroxide or iodine were dialyzed separately, the resultant dialysate of each of the treated sera will reactivate the inactive dialyzed untreated serum, that of the oxidized serum being the least active. These results, therefore, indicate that the dialyzable component of complement is different from the so-called "third" or "fourth" or the oxidizable components of complement.

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Influence of Color Filters on Photodynamic Action of Fluorescent Dyes on *Gonococcus*.

T. L. CH'IN. (Introduced by S. H. Zia.)

From the Department of Bacteriology and Immunology, Peiping Union Medical College, Peiping.

In the study of photodynamic action, Tappeiner¹ found that with eosin the green rays were more lethal to paramecium than the other

¹ Osborn, W. W. B., *Complement or Alexin*, Oxford University Press, 1937, p. 16.

² Gordon, J., Whitehead, H. K., and Wormall, A., *Biochem. J.*, 1926, **20**, 1028, 1036, 1044.

³ Chow, B. F., and Wong, Sam C., *PROC. SOC. EXP. BIOL. AND MED.*, 1938, **35**, 120.

⁴ Chow, B. F., and Zia, Samuel H., *ibid.*, 1938, **38**, 695.

¹ Tappeiner, H. V., *Munch. Med. Wchnschr.*, 1900, **47**, 5.