velops separated from the infundibulum its growth is excessive, it shows cellular hypertrophy and produces an excess of the pigmentary hormone which induces intense hyperpigmentation in the host. The infundibulum normally controls p. intermedia function by inhibition.

11498 P

Influence of Age on Rate of Immune Response of Mice to Formolized Equine Encephalomyelitis Virus.

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It has been reported[1] that the ability of mice to be immunized with formolized virus of Eastern equine encephalomyelitis increases with age. The question then arose as to whether this was an expression of different rates of development of immune response, or rather of maximum responses of which various age-groups were capable. Rate of antibody response to a trypanosomal infection in rats was found to increase with age.[2]

The rate of development of neutralizing antibodies in serum of mice of 3 age-groups was studied. Mice 3 months, 14-15 days and 4-5 days of age were injected intraperitoneally on the 1st, 3rd and 5th days with 0.2-0.25 cc of formalin-inactivated virus of Eastern equine encephalomyelitis. This consisted of a 10% suspension of infected mouse brain in 0.5% formalin, which proved to be non-infectious on intracerebral injection of normal mice. Mice in each group were bled from the heart and the sera pooled, on the days indicated in Fig. 1, *i.e.*, 4th, 5th (4 hours after the last injection of formolized virus), 6th day, etc. Serum-neutralizing antibodies were measured by the intraperitoneal protection test[3] in normal mice from 13 to 15 days of age, using 4 mice for each virus dilution-serum mixture. Sera of different age-groups taken on the same day were compared simultaneously.

The antibody titer is recorded in Fig. 1 as doses of virus neutralized. This was calculated from the difference between the infective

1 Morgan, I. M., PROC. SOC. EXP. BIOL. AND MED., 1939, **42**, 501.

2 Kolodny, M. H., *Am. J. Hyg.*, 1940, **31**, 1, Sec. C.

3 Olitsky, P. K., and Harford, C. G., *J. Exp. Med.*, 1938, **68**, 173.

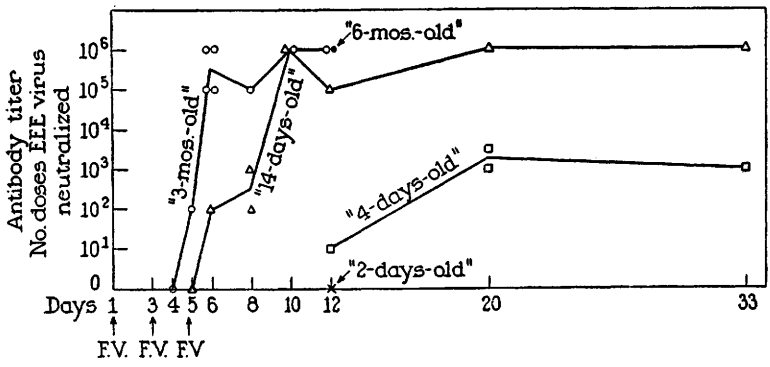

FIG. 1.

Rate of development of neutralizing antibodies in serum of mice of various ages in response to 3 doses of formolized Eastern equine encephalomyelitic virus (F.V.).

titer in control and test groups of mice.* It was found that differences in titer must be more than tenfold to be significant. In serum of mice immunized at 3 months of age, neutralizing antibodies began to appear on the 5th day reaching a maximum by the 6th day. In mice 14-15 days old at the beginning of immunization, antibodies were not demonstrable until the 6th day and reached a maximum between the 8th and 10th days, which was maintained for at least 23 days longer. Considerably slower in response were mice 4-5 days old at the beginning. A minimum of antibody was demonstrable on the 12th day. Sera taken on the 20th and 33rd days protected ¼ to ¾ of mice in groups receiving a wide range of virus dilutions, indicating moderate antibody content. This is in contrast with sharp endpoints obtained with weak or strong antiserum. Sera of "2-days-" and "6-months-old" mice are also shown. Thus the rate of antibody production was found to increase with age. Below a certain age, the final titer reached depended on the age at which immunization was begun.

In order to determine whether the low grade of active immunity reported previously[1] in young mice would increase with time, 2 large groups of mice, 14 days and 3 months old, were immunized as described. Mice from each age group, as well as 3-months-old normal controls, were tested by intracerebral injection of tenfold dilutions of active virus after 2, 3 and 4 weeks. The "3-months-old" immunized mice resisted 10^7, 10^5-10^6 and 10^6-10^7 doses respectively, as measured by difference in titer between control and test groups.

* For example, if the titer of virus in the presence of normal serum were 10^{-7} and with test serum, 10^{-5}, the difference in titer would be 10^2 or 100 doses neutralized.

The "14-days-old" mice were not uniformly resistant; 50% of mice receiving from 1 to 1,000 doses survived. The proportion surviving, of those tested 2, 3 and 4 weeks after immunization, did not increase. It was shown therefore that the high degree of active immunity of "3-months-old" mice was maintained during the 2-4 weeks' period following immunization; and the low degree of resistance of mice immunized when 14 days old did not change significantly during that period, in spite of the observation (Fig. 1) that mice of this age group possessed maximum titer of circulating antibodies.[4]

Summary. The rate of development of neutralizing antibodies in serum of mice immunized with formalin-inactivated virus of Eastern equine encephalomyelitis has been shown to increase progressively with age. The antibodies in serum of mice immunized at a very early age did not reach the maximum titer found in mice immunized when older.

The low degree of active immunity to intracerebral injection of active virus induced in mice 14 days old at the beginning of immunization did not increase from 2 to 4 weeks after immunization. During that interval, mice immunized at 3 months of age maintained a high degree of active immunity.

11499 P

Chronic Histamine Action.*

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Histamine placed in the body in watery solution rapidly produces acute effects of relatively short duration (Dale and Laidlaw[1]). During the past 2 years studies have been undertaken to develop a procedure by which injected histamine would act over prolonged periods and produce chronic effects. The ultimate aim of the investigation was the study of chronic histamine poisoning.

Experimental Procedure. Histamine was administered by sub-

[4] Olitsky, P. K., and Harford, C. G., *J. Exp. Med.*, 1938, **68**, 779.

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[1] Dale, H. H., and Laidlaw, P. P., *J. Physiol.*, 1910, **41**, 318.