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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(Introduced by M. B. Visscher)

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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.

cutaneous or intramuscular injection. All doses mentioned are in terms of histamine base. Two tests were employed to determine the efficacy of the methods used to slow the rate of absorption of histamine from the sites of injection. The first was a comparison in normal guinea pigs of the effects of large doses of histamine in saline solution with the effects of the same dose of histamine in other substances. While this method was satisfactory for the preliminary experiments it was inadequate for the quantitative determination of the extent and degree of prolonged histamine action. The second test was the measurement of the amount and quality of the secretion from gastric pouches of dogs made according to the method of Heidenhain. This test provided a quantitative basis for studying the extended action of histamine. The pouches were prepared under ether or nembutal anaesthesia using the usual surgical technic and sufficient time for recovery was allowed before tests were commenced.

Histamine was first ground with glycol stearate and then suspended in mineral oil. In guinea pigs, injection of this material was quickly followed by typical symptoms and fatalities. Similar results were obtained when histamine particles were covered with paraffin and suspended in oil. Definite protection was obtained with a mixture of finely powdered histamine, pure beeswax or beeswax containing resin and mineral oil. Quantities of histamine which caused fatal reactions when given in saline solution produced only mild symptoms when administered with beeswax. Hot saline extraction of this dose of the beeswax mixture gave watery solutions which when injected produced profound reactions or death. Mixture of the histamine with the beeswax had not destroyed the histamine.

The histamine beeswax mixture has been quantitatively tested by the gastric secretion method in 4 dogs. Doses of histamine ranging from 15 to 60 mg were given. As a routine the volume of material injected was about one cubic centimeter and this was divided among approximately 20 intramuscular sites. Reactions were noticed in only 2 out of 10 experiments. As a rule gastric secretion commenced 10-15 minutes after the injection and continued for from 24 to over 40 hours. The total volume of juice secreted in 24 hours expressed as equivalent volume N/10 HCl ranged from 957 to 1919 cc. In the majority of instances the maximum rate of secretion was reached during the first 4 hours following which it declined gradually. The water, chloride and hydrogen ion loss was combated by dilute saline drinking water, saline solution by vein and under the skin and by the return of gastric juice with a stomach tube. In

these experiments prolonged action of a single injection of histamine was obtained by suspending the histamine particles in a beeswax mixture.

11500

Salt After Adrenalectomy. I. Growth and Survival of Adrenalectomized Rats Given Various Levels of NaCl.*

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It is well known that the administration of sodium chloride to animals deprived of their adrenals considerably delays the onset of adrenal insufficiency, but the importance of the amount of sodium chloride given such animals has not received adequate attention. There is an "optimal" amount of sodium chloride from which such an animal derives benefit; an excess of sodium chloride is injurious. Moreover, inadequate study has been given the matter of the degree to which functional restoration occurs in adrenalectomized animals given salt. These are the problems which concern us here. This study deals with the growth and survival of adrenalectomized animals in response to varying amounts of sodium chloride. Two succeeding reports deal with the urinary excretion of Na and K and with the storage of fed carbohydrate by such rats.

Adrenalectomized rats fed a standard diet and allowed to drink 1% NaCl take in an amount of salt which appears to be optimal for maintaining growth and health. For this study male rats were used; they were 10 weeks of age at the time of adrenalectomy, and weighed approximately 200 g. Controls were subjected to a sham operation in which the adrenals were dissected free of the surrounding tissue but not removed. From a group of 25 adrenalectomized rats given 1% NaCl solution, 15 had an average survival of 45 days (range 12-80 days) and 10 lived beyond the 110th day post-operative. During the course of the experiment all of the animals were tested for completeness of adrenalectomy by withdrawing salt and allow-

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