for the evidence cited above in opposition to this theory. The agent which is effective in neutralizing the acid is not the mucus which is already present at the beginning of secretion. On the contrary throughout the experiment there is a continuous flow of such neutralizing and diluting fluid, due in great part to irritation of the mucosa by the catheter. With reduction in this mucus flow, the late fall in acidity becomes less and less observable.

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Does the Virus of Poliomyelitis Survive in the Monkey Testicle?*

CLAUS W. JUNGEBLUT AND RICHARD THOMPSON.

From the Department of Bacteriology, College of Physicians and Surgeons, Columbia University.

Among the points of major interest in experimental poliomyelitis is the question as to how long the virus is capable of surviving in different organs. Such data not only furnish information which is helpful in studying the factors responsible for selective tissue susceptibility, but may also add to our knowledge concerning the mode of invasion in the disease. From this viewpoint, the testicle has assumed particular prominence in practically all neurotropic virus diseases. It may either serve as portal of entry from which a generalized infection takes its beginning, or, more particularly, offer excellent ground for the development of a circumscript local lesion (herpes, vaccinia). It appears that infection by the testicular route so far has not been attempted in experimental poliomyelitis, save for the recent work of Thompson,[1] who reported his inability to infect rabbits by intratesticular injection with monkey virus. In these experiments it was also found that the virus does not survive for a period of even 24 hours in the rabbit testicle, as determined by subsequent transfer to the monkey.

We considered it important to extend these experiments to a study of the capacity of the testicle of a susceptible animal, i. e., the monkey, to serve either as a portal of entry for the disease or as a suitable living culture medium for the infectious agent. These experiments moreover afforded an opportunity to determine whether

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[1] Thompson, R., *J. Exp. Med.*, 1930, li, 777.

intratesticular injection of virus conveyed any active, generalized immunity to the monkey.

A total of 8 Rhesus monkeys, in several experiments, were injected intratesticularly with poliomyelitis passage virus (Aycock strain), the animals receiving doses of 0.5-1. cc. of a 20% monkey virus cord emulsion into each testicle. The virulence of the virus in each case was confirmed by parallel intracerebral inoculation of a control monkey with the same cord emulsion. It is necessary to select somewhat older animals with well-developed testicles for this work since considerable difficulty may be encountered in forcing so much fluid into the organ which is of unusual hardness in the monkey. In some of the 8 monkeys the injected testicles were left *in situ,* and the animals were observed for the occurrence of any symptoms of the disease. With the majority however, the testicles were removed after various periods of time, ranging from 24 hours to 15 days, and tested for presence of virus. For this purpose, the greater part of the fresh organ was ground and emulsified and the emulsion injected intracerebrally (any excess intraperitoneally) into a new monkey, the remaining portion of the tissue being saved for histological examination.

None of the animals infected intratesticularly ever showed any evidence of generalized poliomyelitis during a period of observation extending from 1 to 2 months. We succeeded in one single instance only in recovering potent virus from an infected testicle, which in this case happened to carry virus for a period of 3 days. On repetition, however, 3-day testicles yielded only negative results. In one case in which the inguinal lymph glands were tested they were found free from virus. Neither the original testicle-infected monkey nor the transfer animals which had been injected intracerebrally with the testicle emulsion, had acquired any active immunity, as evidenced by their full susceptibility to subsequent intracerebral re-infection with potent virus.

Histological examination of the various testicles demonstrated an almost complete absence of any characteristic tissue reaction. A slight leucocytic infiltration found in the earlier specimens was duplicated by similar findings in the testicle of a monkey which had, for reasons of control, received an intratesticular injection of normal monkey cord. We could discover no unmistakable evidence of the presence of inclusion bodies in any of the testicles studied particularly for this purpose.

We conclude from the above experiments that infection by the intratesticular route with poliomyelitis virus, is impossible in the

monkey and that survival of the virus in this organ is exceptional, even within very short limits of time. With this agrees the absence of any demonstrable specific tissue lesion in the infected testicle. Finally, it is noteworthy that neither the testicle-infected monkeys nor the animals receiving testicular emulsion by intracerebral injection had acquired any active immunity.

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The Use of the Stuphenphotometer for Measuring Percentage Hemolysis.

ERIC PONDER.

From Washington Square College, New York University.

The methods for measuring percentage hemolysis may be divided into two classes, (1) subjective methods, such as those in which the observer adjusts the depth of a suspension until some object is just visible, or in which the hemolysing suspension is matched nephelometrically against standards, and (2) methods in which a photosensitive element,such as a potassium or selenium cell, a radiometer, or a thermopile, is used to measure the intensity of light transmitted by the suspension. The methods in the first group are unsatisfactory because too much depends on the judgment of the observer, and because rapid work is usually difficult or impossible; the photometers included in the second group, on the other hand, are usually unsatisfactory because of the difficulty in obtaining sensitivity and stability at the same time when the apparatus is so arranged as to make rapid consecutive readings possible. All methods, moreover, (except the radiometer method) are insensitive to changes in the range of 0% to 10% lysis. Ideally, the method used should be (a) capable of an accuracy of not less than $\pm 2\%$, even in the range from 0% to 10% hemolysis, (b) sufficiently rapid to allow readings at intervals as short as 5 seconds, and (c) perfectly reliable in the sense that its performance does not depend on factors which require continual control. This last condition virtually rules out any method which is not entirely optical.

These conditions are met by the use of the "Stuphenphotometer" ("Stupho"), a photometer recently designed by Pulfrich of Carl Zeiss, in conjunction with a simple device for containing the cell suspensions whose opacity is to be measured. This