ment on chimpanzees (No. 3), the viremia was transient and limited to the 4th and 5th days. In the human disease, as in orally infected monkeys, antibodies are already present at the time of onset of clinical symptoms(11,12), which suggests that the time to look for viremia in man is early in the incubation period.

Summary. Seven of 10 cynomolgus monkeys and 3 of 4 chimpanzees have been found to have viremia early in the incubation period following oral infection with poliomyelitis virus. Viremia was present in animals infected with a Brunhilde type (Egypt) as well as a Lansing type (Y-SK), and persisted in some animals over the 4th, 5th, and 6th days after virus ingestion. Virus was not isolated from blood specimens collected during the first 3 days after feeding, nor on the 7th day, in the two instances in which it was tested. The interval between viremia and the appearance of paralysis varied between 3 and 7 days in the cynomolgus monkeys.

1. Flexner, S., and Lewis, P. A., *J. Exp. Med.*, 1910, v12, 227.

2. Sabin, A. B. and Ward, R., *Science*, 1942, v95, 300.

3. Melnick, J. L., PROC. SOC. EXP. BIOL. AND MED., 1945, v58, 14.

4. Ward, R., Horstmann, D. M., and Melnick, J. L., *J. Clin. Invest.*, 1946, v25, 284.

5. Kling, C., Levaditi, C., and Lépine, P., *Bull. Acad. Med.*, Paris, 1929, v102, 158.

6. Burnet, F. M., Jackson, A. V., and Robertson, E. G., *Austral. J. Exp. Biol. and Med. Sci.*, 1939, v17, 375.

7. Howe, H. A., and Bodian, D., *J. Exp. Med.*, 1945, v81, 255.

8. Melnick, J. L., and Horstmann, D. M., *J. Exp. Med.*, 1947, v85, 287.

9. Von Magnus, H., *Separatum. Acta Pathologica*, 1950, v27, 222.

10. Horstmann, D. M., and Melnick, J. L., *J. Exp. Med.*, 1950, v91, 573.

11. Hammon, W. McD., and Roberts, E. C., PROC. SOC. EXP. BIOL. AND MED., 1948, v69, 258.

12. Steigman, Alex. J., and Sabin, A. B., *J. Exp. Med.*, 1949, v90, 349.

Received February 6, 1952. P.S.E.B.M., 1952, v79.

Measurement of Total Body Water in the Dog with Antipyrine.* (19399)

MARCIA HERROLD AND LEO A. SAPIRSTEIN.

From the Department of Physiology, Ohio State University, Columbus, O.

Body water has been measured in man(1) and in other animals(2,3) by determining the volume of distribution of antipyrine, which appears to equilibrate with all compartments of body water. The originators of the method have, however, questioned the usefulness of this technic in the determination of body water in the dog, for although antipyrine is present in all tissues and organs in this species in amounts proportional to the water content, it is so rapidly destroyed in the dog that it was thought questionable that equilibration could be attained before nearly complete destruction of administered antipyrine.

In preparation for some experiments involving the determination of body water in human subjects, we attempted to study the technic in dogs. In agreement with previous workers(2) we found that blood antipyrine concentrations in the dog decreased at a rate some 5 times greater than the corresponding decrease in man. Since equilibrium in man is not attained before one to 2 hours, it appeared probable that destruction in the dog might be complete before adequate mixing in the body fluids could be accomplished. However, in investigating this question further, we found that the rate of equilibration of antipyrine with body fluids in the dog was so rapid that it compensated for its rapid destruction and made feasible the determination of body water in this species by the antipyrine method.

Methods. The dogs used were mongrel

* Supported by a grant for Cancer Research by the Ohio Department of Health.

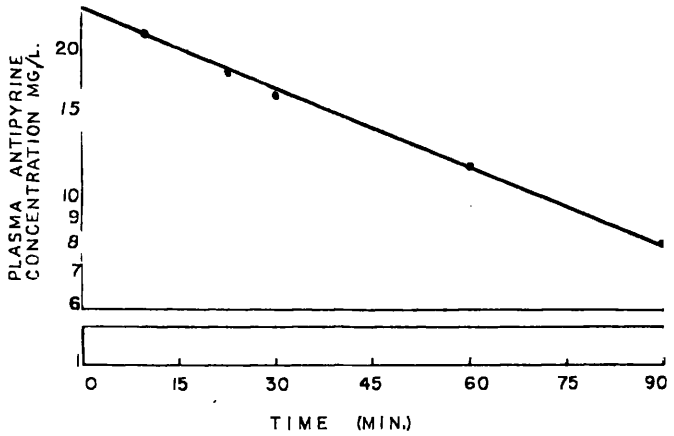

FIG. 1. Typical antipyrine concentration-time curve in the dog (plot on semilog paper).

dogs of both sexes which were chosen to represent a large range of body size and nutritional status. Some animals appeared to be suffering from severe inanition. All animals were fasted overnight and antipyrine was administered intravenously on the following morning at doses of 250 mg in 25 ml of saline. A control blood sample was taken from the external jugular vein just before the injection, and further blood samples were taken at 10, 20, 40, 60, and 90 minutes after the administration of the antipyrine. The concentrations of antipyrine were then determined on the protein free plasma by the method of Brodie *et al.*(2), using the Beckman DU spectrophotometer with the UV light source but with glass rather than quartz cuvettes. Optical densities of 0.020 to 0.035 were typical for the blanks, and the dose of antipyrine administered was sufficient to give optical densities of 0.100 to 0.150 in dogs of moderate size (10 kilos) at 60 minutes when plasma was diluted 1:4. The values of plasma antipyrine obtained were plotted on semilog paper against time, and the concentration at zero time was extrapolated from the decay curve. Anesthesia (intravenous pentobarbital sodium) was used in some dogs. The results did not appear to be significantly affected by this anesthetic.

Results. The antipyrine concentration-time curve obtained was similar in all dogs, though the rate of decay showed some variability, ranging from 20% to 60% per hour with an average of 31% per hour. A typical curve is shown in Fig. 1.

Examination of our curves shows that in all cases even the earliest plasma samples taken (10 minutes) fall on the line extrapolated from the later periods. If mixing were inadequate at this time, one would expect to find the earlier concentrations higher than the extrapolated linear curve. The fact that they are not so indicates that antipyrine equilibrates with the body fluids in the dog in the surprisingly short time of less than 10 minutes.

The rapid equilibration of antipyrine with the body fluids in the dog makes the determination of antipyrine space feasible in this animal despite the high destruction rate of this substance. We have found that samples taken at 20, 40, 60, and 90 minutes will ordinarily give a sufficient number of usable points to enable construction of a linear decay curve with assurance.

By use of this method, the antipyrine space of 8 apparently normal dogs in good nutritional status was found to range from 53.9 to 67.9% of the body weight. The average value was 59.9%. This value correlates excellently with the reported values for the water content of dogs determined as deuterium oxide space of 63% (4) and adds further support to the view that antipyrine is distributed throughout the body water. Emaciated dogs showed somewhat higher values for antipyrine space, values of 75.0, 75.6, and 79.5% being obtained in 3 such animals. This finding is consistent with Behnke's observation(5) that body water is a constant proportion of lean body mass, and that it is reduced in relation

TABLE I. Volume of Distribution and Rate of Destruction of Antipyrine in the Dog.

Sex	Wt, kilo	Antipyrine space, %	Decay rate, %/hr
♀	10.4	55.2	31
♂	8.2	60.8	28
♀	17.3	60.1	23
♀*	13.3	75	29
♀*	18.4	79.5	46
♀	14.8	53.9	31
♀*	4.2	76.5	30
♀	16.9	56.8	21
♂	24	67.9	60
♂	10	59.1	24
♂	18.5	66, 66.4†	20

* Very lean or emaciated dogs.
† Repeat determination after a 3 week period.

to total body mass in obese animals and men. The results in 11 dogs are given in Table I.

Summary and conclusions. 1. Intravenously injected antipyrine is destroyed in the dog at the rate of 20 to 60% per hour. 2. Nevertheless, antipyrine space may be used for the determination of total body water in the dog, because of the fact that the distribution of antipyrine appears to be essentially complete after 10 minutes. 3. Dogs in adequate nutritional status are found to have antipyrine spaces of 53.9% to 67.9%. The average value obtained from 8 normal dogs was 59.9%. 4. Inadequate nutrition and emaciation result in an increase in the antipyrine space, a result consistent with the idea that body water is related more closely to lean body mass than to body weight.

1. Soberman, R., Brodie, B. B., Levy, B. B., Axelrod, J., Hollander, V., and Steele, J. M., *J. Biol. Chem.*, 1949, v179, 31.
2. Brodie, B. B., Axelrod, J., Soberman, R., and Levy, B. B., *J. Biol. Chem.*, 1949, v179, 25.
3. Kraybill, H. F., Hankins, O. G., and Bitter, H. L., *J. Applied Physiol.*, 1951, v3, 681.
4. Smith, H. W., *The Kidney*, Oxford University Press, New York, 1951.
5. Behnke, A. R., Feen, B. G., and Welham, W. C., *J.A.M.A.*, 1942, v118, 495.

Received February 6, 1952. P.S.E.B.M., 1952, v79.

Passive Immunity in Poliomyelitis. IV. Protection of Rhesus Monkeys Against Cerebral Challenge.* (19400)

A. J. RHODES, F. T. SHIMADA, EINA M. CLARK, W. WOOD, AND R. C. RITCHIE.
(Introduced by R. C. Parker.)

From the Connaught Medical Research Laboratories, University of Toronto, and The Hospital for Sick Children, Toronto.

During the last 20 years, several workers have investigated the protective properties of immune serum from human and animal sources in experimental poliomyelitis. The significance of much of this earlier work is difficult to assess, as technics for the accurate titration of virus pools and immune sera had not been developed, the existence of distinct immunologic types of virus was unsuspected, and often the number of animals used was too small for the results to have statistical value. The experiments of Kramer(1,2), however, indicated that human convalescent serum exerted a significant degree of protection in mice challenged cerebrally with the Lansing strain. Interest in this subject was revived when it was shown that gamma globulin prepared from human plasma protected mice against the Lansing strain(3). Bodian(4) demonstrated the presence in this material of antibody to all three immunologic types of poliomyelitis virus. Later he showed that some protection against an intramuscular challenge was achieved in rhesus monkeys inoculated intramuscularly with 2 ml of gamma globulin per kilogram body weight(5). We have shown that mice can be protected against a cerebral challenge of Lansing mouse virus by the inoculation of monkey immune serum, as well as plasma and placental gamma globulins(6). The present paper describes 3 passive pro-

* Aided by a Grant from The National Foundation for Infantile Paralysis.