Reports and Letters

Blood Volume of the Rhesus Monkey

In the course of an investigation of the hemopoietic response of the rhesus monkey to total-body x-irradiation, it became necessary to express data in terms of the average blood volume of these animals. A review of the literature failed to reveal this information and the study reported here was undertaken to establish this value.

Twenty prepuberty rhesus monkeys, 10 males and 10 females, were used in the study. For a period of 6 wk prior to the determinations, the animals were maintained on a standard laboratory diet. During this period the animals did not exhibit any sign of the diseases common to the species.

The blood-volume determinations were made according to the method of Aust *et al.* (*1*), using radioactive iodinated human serum albumin (IHSA) as the tracer agent. In brief, 2.0 μc of IHSA in a volume of 0.5 ml was injected into the right femoral vein of each animal. Ten minutes later, a sample of blood was withdrawn from the left femoral vein into a heparinized syringe. A 1.0-ml aliquot of this sample was then assayed in a well-type scintillation counter. Previously, 0.5 ml of the IHSA had been diluted to a total volume of 1000 ml. A 1.0-ml aliquot of this known dilution was used as a standard and was assayed in the same manner. The blood volume was computed by the following formula:

$$\frac{\text{Net counts/min of standard} \times 10^3}{\text{Net counts/min of sample}} = \text{B.V.}$$

The 20 animals included in this study weighed from 2.2 kg to 5.3 kg. The blood volumes ranged from 49 ml/kg to 71 ml/kg. The average blood volume was found to be 60.9 ml/kg, and the probable error of this mean is 0.96 ml/kg. This distribution about the mean closely corresponds to values given by Freinkel *et al.* (*2*) and Storaasli *et al.* (*3*) in similar studies performed on human beings.

Although human serum albumin is a foreign protein to these animals, the half-time of the plasma clearance of the IHSA was found to be 22 hr, and this approximates values reported by Storaasli (*3*) in human beings. In addition, repeated injections of this material into individual monkeys have not induced demonstrable foreign protein reactions.

M. A. Bender
Roswell Park Memorial Institute, Buffalo, New York

References

1. J. B. Aust *et al.*, *Proc. Soc. Exptl. Biol. Med.* 77, 514 (1951).
2. N. Freinkel *et al.*, *J. Clin. Invest.* 32, 138 (1953).
3. J. P. Storaasli *et al.*, *Surg. Gynecol. Obstet.*, 458 (1950).

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Identification of Vaccine Strains of Newcastle Disease Virus

Before random purity and potency tests can be applied to living virus vaccines for Newcastle disease, a practical method is needed for typing and identifying the strain of virus in the vaccine (*1*). Such a method based on several years of study of strains of virus from the Newcastle disease repository at Wisconsin (*2*) has been tested successfully, using samples of vaccines submitted by the Biological Products Section of the U.S. Department of Agriculture.

The vaccine sample is reconstituted so that 100 chicken vaccine doses are contained in 1 ml, and five tests are made on selected dilutions of the sample: LD_{50} for 10-day chicken embryos; mean death time of the minimum lethal dose; intracerebral pathogenicity for day-old chicks; susceptibility to neutralization by Newcastle disease virus antiserum of known titer; and bacterial contamination (*3*). Allanto-amniotic fluids harvested from embryos inoculated for the LD_{50} test are examined for equine erythrocyte agglutinins and for resistance of the fowl erythrocyte agglutinins to inactivation at 56°C.

The strains are typed according to their growth rate (*4*), which is measured by the mean death time of the minimum lethal dose. The lentogenic, or slowly growing, strains, which take 90 to 150 hr to kill embryos, include only a few strains that are all of low pathogenicity for chickens. Two strains are used in intranasal or aerosol vaccines. The mesogenic, or intermediate, strains, which take 60 to 90 hr to kill, include a larger group of strains possessing low to high pathogenicity for chickens. Two strains are used in wing-web vaccines. The velogenic, or rapidly growing, strains, which kill in 40 to 60 hr, include a large group of strains of moderate to high pathogenicity for chickens. All are unsatisfactory as vaccines.

The strain is defined by the use of additional test characteristics. A comparison (Table 1) of the four widely used American living virus vaccine strains—Bl, LaSota, Roakin, and MK107 (L), illustrates the use of definitive tests (tests 6 and 7) in conjunction with typing tests (tests 2 and 3).

The LD_{50} in test 1 is dependent on vaccine production procedure, such as the quantity of diluent and buffers used in packaging, and is not of diagnostic importance. The intracerebral pathogenicity index of lentogenic strains should be less than 0.25, so the index (test 3) helps to confirm the test for the mean death time (test 2). The capacity to agglutinate equine erythrocytes (test 6) differentiates the two strains in each type. Stability of the fowl erythrocyte agglutinins at 56°C (test 7) serves as an additional check on the strain definition, since all four strains should be inactivated in 15 min.

Some of the afore-described tests have been made on all of the 80 strains of Newcastle disease virus available in the repository, and other tests have been made on fewer of these strains. Twelve additional characteristics are being con-

Table 1. Characteristics of four vaccine strains

Sample	Test 1, LD_{50} (log)	Test 2, mean death time (hr)	Test 3, intracerebral pathogenicity (index)	Test 6, equine hemagglutinin (index)	Test 7, resistance 56°C (min)	Diagnosis	
						Type	Strain
A	7	120	0.1	0	> 15	Lentogenic	Bl
B	7	100	0.1	0.4	> 15	Lentogenic	LaSota
C	5	70	1.1	0	> 15	Mesogenic	Roakin
D	5	60	1.1	0.5	> 15	Mesogenic	MK107 (L)