TECHNICAL PAPERS

Hemagglutinating Behavior of Mouse and Egg-adapted Type A (PR8) Influenza Virus[1]

Seymour S. Kalter

College of Medicine, Syracuse University and The Virus Laboratory, Bureau of Laboratories, Syracuse, New York.

Influenza virus undergoes a change when introduced into the mouse following either primary isolation or adaptation in the chick embryo (*1*, p. 357, and *3*). Both Hirst (*1*, p. 357) and Wang (*3*) showed that the egg-adapted virus multiplies readily in the lungs of mice without producing either the clinical disease or histological changes. Death or pulmonary lesions will occur after the egg-adapted virus has had several passages through mice.

The present study indicates that mouse-adapted influenza virus undergoes a change when the virus is returned to the egg; although this change appears to be easier than the egg-to-mouse adaptation.

The PR8 strain of influenza virus A which was employed in these experiments has been maintained in the laboratory by allantoic passages through fertile eggs. Its virulence for mice had decreased so that the mean lethal dose which kills 50% of the animals (LD_{50}) per 0.05 ml intranasally was 10^{-2}. After 25 mouse passages, this mouse-adapted strain possessed an LD_{50} of 10^{-6}. These passages were performed by harvesting lungs 3–4 days following inoculation, making a 10% suspension in buffered saline by homogenizing in a Waring blender, and reinoculating into 3–4-week-old mice.

Ten-to-eleven-day-old embryos were inoculated into the allantoic sac with 0.2 ml of a 10^{-3} dilution of mouse-adapted or original egg virus. At 2-hr intervals, 5–10 eggs of each group were removed from the incubator and placed in the refrigerator. This was continued for every 2 hr up to 24 hr.

Allantoic fluid was harvested in the usual manner at each 2-hr period and titrated in the hemagglutination test by the pattern method of Salk (*2*) with human type O erythrocytes.

The data recorded in Fig. 1 are the results of a typical experiment with the 27th passage mouse strain. The egg virus was from a pool of allantoic fluid.

It was demonstrated that the egg strain produced hemagglutination at least 4 hr earlier than the mouse strain. Similar results were obtained with material from other mouse passages and other allantoic fluid pools. The earliest that hemagglutination was produced by the egg strain was 12 hr, while for the mouse strain it was 18 hr after inoculation. These were obtained in simultaneous titrations, using material from the 31st mouse passage; the egg material was from an allantoic pool not previously titrated. There was some variation in end points but when tested after 24 hr incubation, they were essentially the same for both strains. One egg passage sufficed to make the mouse strain indistinguishable from the original egg strain in hemagglutinating capacity.

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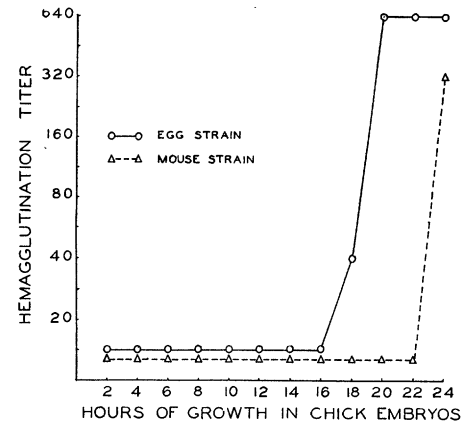

FIG. 1. Hemagglutinating ability (type O red blood cells) of mouse-adapted and original egg strain in the chick embryo. Type A (PR8) influenza virus employed.

When the mouse-adapted PR8 strain was cultivated in the mouse lung and hemagglutinating ability determined at 2-hr intervals, the results were essentially similar to those obtained by Wang (*3*). Perceptible hemagglutination with mouse lung virus was obtained within 6–8 hr after intranasal inoculation. As also observed by Wang, infection of chick embryos occurred more rapidly than hemagglutination.

The data reported here substantiate the observations of others (*1*, p. 357, and *3*) that changes occur in the virus particle which is adapted to a different host. Whereas the results obtained by these workers indicate that there is a marked change in the virus when adapted to the mouse from the egg, little information is available concerning the reversal of this adaptative procedure. It appears, as Hirst suggests (*1*, p. 367), that adaptation to the egg is "a less drastic procedure than mouse adaptation."

Our data appear to agree with Hirst's findings that egg-adapted strains from a given epidemic do not differ significantly (*1*, p. 367). One may postulate, then, that the virus particle in adapting itself to a host alters itself to take advantage of the new host's metabolic systems. Thus, when the virus is introduced into a new host with a slightly different metabolic pattern, a probable rearrangement of the virus molecule occurs.

References

1. HIRST, G. K. *J. exp. Med.*, 1947, **86**, 357; 367.
2. SALK, J. E. *J. Immunol*, 1944, **49**, 87.
3. WANG, C. I. *J. exp. Med.*, 1948, **88**, 515.