

Attempt to Demonstrate Hemadsorption on Cells Infected with Dengue Virus

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PURPOSE AND DESCRIPTION: A hemagglutinating antigen is found on the coat of dengue viruses. The question arises whether this antigen, or a functionally similar one, appears and can be detected on the surface of cells following infection by dengue virus. Thus the purpose of this experiment was to demonstrate the appearance of hemadsorption on MK2 cells following dengue infection. The ability of cells infected with dengue virus to hemadsorb would provide a rapid diagnostic test for the virus. In addition the existence of a phenomenon indicating that cell surfaces are altered as a consequence of dengue infection would provide a more solid conceptual framework for in vitro experiments designed to elucidate the interaction of dengue infected cells and host immune mechanisms.

PROGRESS: Four-day old monolayer cultures of MK2 cells in 1 oz. prescription bottles were infected with Dengue 4 (IM 75) and Dengue 2 (SP-BKM-551). The virus: cell multiplicity of infection was 1:10 for Dengue 4 and 1:2 for Dengue 2. On day 2, 4, and 6 following infection, separate cell monolayers were washed to remove free virus. Erythrocytes in a 0.4% suspension buffered at pH 6.4 and 6.8 were added to cultures infected with Dengue 2 and Dengue 4 respectively. The types of erythrocytes employed were goose, human type O, guinea pig, and 2-day old chick. Non-infected cultures served as virus control cultures. After adding 1.6 ml of the erythrocyte suspensions, the cultures were incubated at 4°C for 20 minutes and then at 27° for 1-2 hours before they were examined for hemadsorption. In addition to using erythrocytes in buffers of pH 6.4 for Dengue 2 and pH 6.8 for Dengue 4 (representing the HA pH optimum for each virus) buffers of pH 8.0 and pH 9.0 were also employed. Virus replication was monitored by CPE beginning on day 6 for the Dengue 2 strain; no CPE was noted in Dengue 4 infected cultures by day 6. No hemadsorption was noted in Dengue 2 and Dengue 4-infected MK2 cell monolayers at 2, 4, and 6 days after infection under the conditions tested.