Abortive Infection of Hamster Cells by Polyoma Virus

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Mouse embryo cells are permissive for polyoma-virus replication. They support viral DNA replication and virus production and are finally killed.

In contrast, cells of the hamster cell line BHK 21/13* are non-permissive and undergo abortive transformation (Stoker, 1968). A small proportion of such cells are transformed and show permanently altered growth properties. Transformed cells contain at least part of the viral genome integrated into the cellular DNA (Westphal & Dulbecco, 1968; Sambrook, Westphal, Srinivasan & Dulbecco, 1968). Cellular DNA synthesis is probably necessary for the integration process (Todaro & Green, 1967).

Infection of stationary permissive cells, initially having a low rate of DNA synthesis, with polyoma virus results in a marked stimulation of cellular DNA synthesis (Dulbecco, Hartwell & Vogt, 1965). Studies on the interaction between polyoma virus and stationary BHK 21/13 cells are reported.

BHK 21/13 cells were maintained in a stationary state by using a method based on that of Fried & Pitts (1968) for mouse embryo cultures. Cells maintained in Eagle's medium containing 1% of calf serum for 5 days had low rates of DNA synthesis, low DNA polymerase activity and low thymidine kinase activity. DNA polymerase activity and thymidine activity increased 3–40-fold and DNA synthesis increased 200-fold when the cells were overlaid with fresh medium containing 10% of calf serum.

No significant increase in DNA synthesis, DNA polymerase activity or thymidine kinase activity occurred after infection of stationary BHK 21/13 cells with polyoma virus at a multiplicity of 50 plaque-forming units/cell. Infection of stationary mouse embryo cells under identical conditions resulted in 4–10-fold increases in DNA polymerase activity and thymidine kinase activity and 10–20-fold increases in DNA synthesis.

Viral-specific mRNA sequences were detected in polyoma-virus-infected BHK 21/13 cells by using a DNA-RNA competitive hybridization technique developed by Martin & Axelrod (1969).

Polyoma viral DNA replication could not be detected in BHK 21/13 cells (stationary or growing) or L929 cells, a mouse fibroblast cell line, after viral infection. However, limited viral DNA

replication did take place in polyoma-virus-infected hybrid BHK 21/13–L929 cells.

These results suggest that BHK 21/13 cells may lack a cellular function that is necessary for polyoma-virus replication.

G. A. N. is the holder of a Research Studentship from the Ministry of Education for Northern Ireland.

Dulbecco, R., Hartwell, L. H. & Vogt, M. (1965). *Proc. natn. Acad. Sci. U.S.A.* **53**, 403.

Fried, M. & Pitts, J. D. (1968). *Virology*, **34**, 761.

Martin, M. A. & Axelrod, D. (1969). *Proc. natn. Acad. Sci. U.S.A.* **64**, 1203.

Sambrook, J., Westphal, H., Srinivasan, P. R. & Dulbecco, R. (1968). *Proc. natn. Acad. Sci. U.S.A.* **60**, 1288.

Stoker, M. (1968). *Nature, Lond.*, **218**, 234.

Todaro, G. J. & Green, H. (1967). *J. Virol.* **1**, 115.

Westphal, H. & Dulbecco, R. (1968). *Proc. natn. Acad. Sci. U.S.A.* **59**, 1158.

Cause of the Heterogeneity of 'Silent' Plasma Cholinesterase Variants

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Rubinstein, Dietz, Hodges, Lubrano & Czebotar (1970) have reported that the homozygotes for the 'silent' cholinesterase (Ch_1^s) gene can be divided into two groups. One group (type 1 or S_1) has no cholinesterase except that originating from erythrocytes, and type 2 or S_2 has 2% of normal plasma cholinesterase activity. Thus type 1 and type 2 represent total absence and suppressed synthesis of plasma cholinesterase respectively.

The plasmas of two subjects homozygous for the apparent Ch_1^s gene were studied by a thiocholine assay procedure (Das & Liddell, 1970) with acetylthiocholine and butyrylthiocholine as substrates in the presence and in the absence of several inhibitors, by polyacrylamide-gel electrophoresis and by an immunoabsorption test. The results were compared with those obtained with normal plasma and a soluble preparation of normal erythrocyte cholinesterase (Das, Watts & Watts, 1971).

No activity could be demonstrated in either of the 'silent' samples with acetylcholine, butyrylcholine and benzocholine as substrates.

Both the samples had only about 1.5 and 0.8% of the activities in normal plasma with 5mM-acetylthiocholine and 7.5mM-butyrylthiocholine as substrate respectively. They hydrolyse acetylthiocholine slightly faster than butyrylthiocholine whereas the reverse is true for the normal plasma. But erythrocyte cholinesterase hydrolyses acetylthiocholine almost specifically.

Comparison of the Michaelis curves obtained with acetylthiocholine as substrate shows optimum

* Abbreviation: BHK cells, baby-hamster kidney cells.