Recovery of adenovirus type 7 from human brain cell cultures

ANN LORD, R. N. P. SUTTON, AND J. A. N. CORSELLIS

From the Departments of Medical Microbiology, King’s College Hospital Medical School, Denmark Hill, London, and of Neuropathology, Runwell Hospital, Essex

SYNOPSIS A strain of adenovirus type 7 was recovered from cultured brain cells, taken at necropsy from a patient aged 71 years with chronic schizophrenia. This recovery may indicate the reactivation of a latent infection with one of the few adenoviruses that has regularly—if rarely—been associated with clinical encephalitis.

Infection with adenoviruses in man is usually accompanied by respiratory symptoms and involvement of the central nervous system is unusual. Chou et al. (1973) culled from the literature 35 cases of encephalitis in which infection with adenoviruses could reasonably be assumed to have played a part. Adenovirus type 7 or 7a was recovered from 13 of these.

Recovery of adenoviruses from brain tissue is even rarer, although these viruses grow well in standard tissue cultures. Of three reports, of such recoveries, two were adenovirus type 7 or 7a (Lelong et al., 1956; Jen et al., 1962) and the third was adenovirus type 32 (Roos et al., 1972).1

During the course of an investigation into the role of viruses in psychiatric disorders in the elderly, we recovered a strain of adenovirus from brain tissue: this is the fourth such recovery to be reported.

METHODS

PATIENTS Elderly patients in a psychiatric hospital were investigated. Samples of brain tissue were taken at necropsy; this was carried out as soon as possible and invariably within 18 hours of death. These samples were approximately 3.5 cm³ in size and were taken from temporal, frontal, and, occasionally, occipital lobes. They were placed in Eagle’s Minimal Essential Medium (MEM) with 5% bovine albumin and added antibiotics and were transported at 4°C by rail to King’s College Hospital.

Tissue cultures were initiated by inoculating 30 ml plastic flasks (Falcon) with a suspension of minced tissue (Katz et al., 1969) in a medium based upon Eagle’s MEM with 20% fetal calf serum (FCS) and 600 mg/l glucose. Tissue specimens were also trypsinized and a suspension of 5 x 10⁶ cells inoculated into plastic flasks. When the cell sheets became confluent, the medium was changed to one containing 5% FCS. Aliquots of the supernatant tissue culture fluid were taken at weekly intervals and stored at −70°C. These aliquots were then inoculated into HEp-2, BSC-1, and WI-38 cells. Two blind passages were carried out before specimens were discarded as negative. Cells were observed for up to 15 days and—in the case of BSC-1 and WI-38 cells—haemadsorption tests with human group O erythrocytes were carried out on days 5, 10, and 15.

A portion of each specimen of brain tissue was homogenized and stored at −70°C: this was later inoculated into HEp-2, BSC-1, and WI-38 cells in the same way.

RESULTS

Thirty-eight specimens from 15 patients were received. Five of these were successfully propagated as tissue cultures; of the remaining 33, some were bacterially contaminated or, for other reasons, did not grow. The time taken for cells to become confluent and the length of time that the cultures were under observation are given in the Table.

Adenovirus type 7 was recovered in WI-38,
BSC-1, and in HEp-2 cells from samples of tissue culture fluids overlaying cultured temporal lobe cells taken from patient R3. This patient was a woman aged 71 years with long-standing chronic schizophrenia who died in February 1974; the immediate cause of death was lobar pneumonia and no macroscopic or histological evidence of meningitis or encephalitis was found. The virus was identified by neutralization tests and it was present in samples taken during the 10th, 11th, and 12th weeks of culture.

No viruses were recovered from any of the other cultures or from cells inoculated with the homogenized brain cell suspension.

**DISCUSSION**

The recovery of an adenovirus from brain tissue is, in itself, sufficiently unusual to merit record. Whether the presence of this virus indicates an active infection or represents a possible reactivation of a latent agent deserves discussion.

Previous recoveries of adenoviruses from brain tissues have been made by inoculation of tissue homogenates into susceptible tissue cultures and the patients who yielded these strains had all suffered from severe illness with clinical encephalitis. The recovery of adenovirus type 7 in these circumstances was not, perhaps, surprising, for this virus is probably more frequently associated with encephalitis than any other type (Chou et al., 1973). However, the patient from whose brain we recovered adenovirus type 7 was not suffering from encephalitis and died from respiratory illness. This respiratory illness may have been associated with adenovirus type 7 infection (which was epidemic in the United Kingdom at the time) and our recovery may merely represent one aspect of a generalized viraemia. If this were so, then recovery of virus by direct inoculation of brain tissue into susceptible tissue cultures would have been expected. Our strain was not recovered by direct inoculation of tissue cultures but could be demonstrated only after prolonged culture of the brain cells; this suggests that the virus was released from a masked or latent state. The situation closely resembled that described by Precious and her colleagues (1974) who induced a chronic infection in brain cells of mice which had previously been inoculated intraperitoneally with Chikungunya virus.

On a number of occasions, latent viruses, including adenoviruses, have been unmasked in primate brains by culture of explants (Rogers et al., 1967; Basnight et al., 1971; Hooks et al., 1973). They have been recovered with far greater ease from primates than from man but 'human tissues certainly harbour their own complement of latent viruses' (Gajdusek, 1972). These latent viruses may become activated by immuno-suppressive treatment, as was suggested in the case of the patient from whom adenovirus type 32 was recovered (Roos et al., 1972). Natural factors, including those associated with ageing, may also play a part and the possibility of such reactivation of potentially neurotropic viruses in the elderly brain clearly deserves further investigation.

We are grateful to the McAlpine Foundation for financial support and to Mr R. J. Barnes for valuable help.

**REFERENCES**


