A case of acute measles meningoencephalitis with virus isolation

D. R. PURDHAM AND P. F. BATTY

From the Virology Laboratory, Children's Hospital, Ladywood, Birmingham, and Worcester Royal Infirmary

SYNOPSIS A case of measles meningoencephalitis is described. Measles virus was isolated from the cerebrospinal fluid cells, in which virus antigen was also detected by indirect immunofluorescence.

Measles virus is difficult to isolate from infections of the central nervous system. Only by cocultivation techniques is it possible to isolate the virus from brain material in cases of subacute sclerosing panencephalitis (Horta-Barbosa, Fucillo, Sever, and Zeman, 1969; Purdhham, 1974), and it has been suggested that this is also the case in primary measles encephalitis (ter Meulen, Käckell, Müller, Katz, and Meyermann, 1972). Transmission of infection to monkeys using brain material (Scott, 1967), and a single report of successful virus isolation from cerebrospinal fluid taken from a patient with measles encephalitis (McLean, Best, Smith, Larke, and McNaughton, 1966) are among the scanty reports on the recovery of virus from acute measles encephalitis.

Case Report

A 6-year-old English boy became ill with a characteristic morbilliform rash, fever, and anorexia. Four days later he was admitted to hospital with generalized convulsions which were controlled with intravenous diazepam. On admission his temperature was 40°C and pulse 160/minute. He had a fading generalized rash and appeared emaciated and slightly dehydrated. The total blood leucocyte count was 23 000/cmm, of which 80% were neutrophils; serum electrolytes were normal; blood urea was 9.8 mmol/litre. He was treated with intravenous maintenance fluids throughout, and also initially with ampicillin and sulphadiazine.

He showed some improvement nine days after the onset of the rash, in both consciousness and speech, but still remained confused and disorientated. Lumbar CSF taken on the eleventh day contained 190 nucleated cells/cmm, of which 85% were lymphocytes; CSF sugar was 5.7 mmol/litre, and protein was 0.42 g/litre. When the diagnosis of measles encephalitis was determined from this specimen, ACTH therapy was started. However, 14 days after the onset of the rash, he deteriorated with increasing tone, dilating pupils, unconsciousness, and vomiting; he died the following day. Permission for necropsy was not given.

Virological Studies

IMMUNOFLUORESCENCE

The lumbar CSF was lightly centrifuged and the cellular deposit washed in phosphate-buffered saline (PBS), pH 7.2, three times, from which drop preparations were made on standard microscope slides. These were air-dried and fixed in acetone at −20°C for one minute. A conventional indirect immunofluorescence method was used employing rabbit anti-measles serum, and a conjugated goat antirabbit-globulin serum. In one control preparation the specific antimeseles serum was replaced by pre-immune rabbit serum, on a second preparation by rabbit antiherpesvirus hominis serum, and a third control preparation was treated with no serum, i.e., fluorescent conjugate only; the control smears were identical to those of the test and were treated in parallel with them.

SEROLOGY

The patient’s serum was tested for the presence of antibodies to measles virus by complement-fixation (CF), and his CSF by complement-fixation and haemagglutination-inhibition (HAI) tests.

ISOLATION

The PBS-washed cells from the CSF were inoculated into primary rhesus monkey kidney cell monolayers, which were incubated stationary at 35°C. The tissue cultures were examined for cytopathic effects thrice weekly, and tested at five-day intervals for haemadsorption using 0.5% rhesus monkey erythrocytes.
in PBS at 37°C. Typing was carried out by neutralization, haemadsorption-inhibition, and indirect immunofluorescence, with specific antisera.

**Results**

**IMMUNOFLUORESCENCE**

Bright fluorescence was seen in a number of CSF cells in both the cytoplasm and the nucleus, and was generally granular in appearance. A few of the cells also exhibited fluorescent intranuclear areas (fig), which by phase-contrast microscopy resembled inclusion bodies. Only very faint non-specific staining was visible in the control cells.

![Cerebrospinal fluid cell containing fluorescing measles antigen in both cytoplasm and nucleus.](image)

**SEROLOGY**

The only serum sample available for virus studies was collected 10 days after the appearance of the rash and had a CF antibody titre to measles virus of 1:256. The supernatant fluid of the CSF taken on the eleventh day was negative for measles antibody by both CF and HAI tests.

**ISOLATION**

After 10 days' incubation, foci of infection in the form of multinucleated giant cells and cellular degenerative areas were visible in the tissue cultures, all of which were positive by haemadsorption. On subculture these effects were transmissible to further monkey kidney cultures but not to HeLa or HEp 2 cells. Immunofluorescence tests revealed measles antigen in the inoculated cultures, and the isolate was typed by both virus neutralization and haemadsorption-inhibition tests.

**Discussion**

The isolation of measles virus from the central nervous system in cases of acute encephalitis may be as difficult as in cases of subacute sclerosing panencephalitis (ter Meulen et al, 1972). However, virus in a 'defective' form was not a problem in this case, as the agent was freely recoverable by standard tissue culture methods, although it might be argued that in fact cocultivation of washed CSF cells and rhesus monkey kidney cells was done. The use by others of immunofluorescence for demonstrating virus antigen in CSF cells (Somerville, 1966; Dayan and Stokes, 1973) has encouraged us to apply this rapid technique at every opportunity, but like others (Tomlinson and MacCallum, 1973), unsuccessfully (until now). The antisera specificity, control methods employed, morphology of reacting cells, and the fact that measles virus was isolated validate the immunofluorescence result. Contributory factors in this case to successful isolation and immunofluorescence may include: (1) plenty of nucleated cells in the CSF and (2) absence of measles antibody in the CSF, which, even if present at undetectable levels, could have been removed mechanically when the cells were washed. Detection of measles virus replication in tissue culture is greatly facilitated by regular haemadsorption tests with monkey erythrocytes; this will often detect virus even in the absence of cytopathic effects, which may not become evident until further passage. The fact that the results of Somerville and Dayan have not been confirmed, apart from our present single success, suggests that the presence of viral antigen detectable by immunofluorescence in the cells of lumbar CSF is not a common occurrence.

We thank Dr T. H. Flewett for his valuable advice in preparing this paper.

**References**


The November 1974 Issue

THE NOVEMBER 1974 ISSUE CONTAINS THE FOLLOWING PAPERS

Immunological studies in a case of T-cell leukaemia
G. STATHOPOULOS, M. PAPAMICHAEL, P. SHELDON, D. CATOVSKY, A. J. S. DAVIES, E. J. HOLBOROW, AND EVE WILTHAW

High counting rates from $^{125}$I in radioimmunoassays
T. K. BELL

The role of circulating hepatitis B antigen/antibody immune complexes in the pathogenesis of vascular and hepatic manifestations in polyarteritis nodosa
CHRISTIAN G. TREPO, ARIE J. ZUCKERMAN, RICHARD C. BIRD, AND ALFRED M. PRINCE

Capillary blood collection in haematology
J. STUART, B. A. BARRETT, AND D. R. PRANGNELL

Folates in human serum
K. RATAVASTHIN, J. A. BLAIR, R. J. LEEMING, W. T. COOKE, AND V. MELIKIAN

A simple method for the clinical determination of the mitotic activity of the human prostate in vitro
K. E. O. SENIUS, J. PIETILÄ, I. ARVOLA, AND P. TUOHIMAA

Culture of specimens other than sputum for Mycobacteria
D. A. MITCHISON AND V. R. ABER

Unusual skin sepsis due to a strain of multiresistant Staphylococcus aureus
JOY HARRISON AND R. W. LACEY

Comparison of the titres of ASO, anti-DNase B, and antibodies against the group polysaccharide of group A streptococci in children with streptococcal infections

Sensitivity of Streptococcus pyogenes to sulphamethoxazole, trimethoprim, and cotrimoxazole
E. YOURASSOWSKY, M. P. VANDERLINDEN, AND E. SCHOUTENS

The rapid recognition of Lancefield group B haemolytic streptococci
R. J. FALCON

Bactericidal capacity of peripheral blood leucocytes in relation to bacterial infections in acute lymphoblastic leukaemia in childhood
EILEEN N. THOMPSON AND RALPH WILLIAMS

Indirect immunofluorescence test for the detection of Aspergillus fumigatus antibodies
D. W. WARNOCK

Separation of three isoenzymes of N-acetyl-$\beta$-D-hexosaminidase from human tissues by cellulose acetate membrane electrophoresis
A. WESTWOOD AND D. N. RAIN

Plasma alkaline phosphatase isoenzymes in hepatobiliary disease
A. O. AFONJA AND D. N. BARON

Sterilization of the biological safety cabinet
S. W. B. NEWSOM AND B. M. WALSINGHAM

Technical method
A simple and rapid method for the differentiation of the members of the genus Klebsiella
R. FREEMAN

Book review
Notice

The Association of Clinical Pathologists: 93rd general meeting

Copies are still available and may be obtained from the PUBLISHING MANAGER,
BRITISH MEDICAL ASSOCIATION, TAVISTOCK SQUARE, LONDON, WC1H 9JR, price £1.05.