Isolation of Parainfluenza Virus Type 3 from Cerebrospinal Fluid Associated with Aseptic Meningitis

RANDALL D. CRAVER, MD,* ROBERT S. GOHD, PHD,* DANIEL R. SUNDIN, PHD,* AND JOHN C. HIERHOLZER, PHD†

Parainfluenza virus type 3 has been isolated from the cerebral spinal fluid (CSF) from six individuals—four children and two adults—over a 10-year period. All had fever, and four had signs of meningitis. All recovered uneventfully, including one child undergoing chemotherapy for medulloblastoma. The clinical presentation of this child who developed parainfluenza virus type 3 meningitis is described, and the cases of five other individuals with parainfluenza virus type 3 isolated from the CSF are briefly reviewed. The paramyxovirus parainfluenza virus type 3, in addition to mumps virus, may be considered capable of infecting the central nervous system. (Key words: Parainfluenza virus type 3; Mumps; Aseptic meningitis) Am J Clin Pathol 1993;99:705-707.

CASE REPORT

A girl underwent a partial resection of a medulloblastoma at 20 months of age. She was treated with systemic cyclic monthly chemotherapy consisting of two courses of vincristine and cyclophosphamide followed by a third month of cisplatin and etoposide. Before her eleventh course of chemotherapy (vincristine, cyclophosphamide) at the age of 2 years and 6 months, she complained of headache and had a temperature of up to 38.7 °C. Complete blood count at this time included a white blood cell count of 2.7 × 10⁹/L (2700/mm³) with a differential of 68% neutrophils, 27% lymphocytes, and 5% monocytes. The cerebrospinal fluid (CSF) from a spinal tap had a glucose level of 2.4 mmol/L (44 mg/dL) (normal, 2.5-4.4 mmol/L [45-80 mg/dL]), a protein of 1.1 g/L (113 mg/dL) (normal, 0.12-0.6 g/L [12-60 mg/dL]) and cell counts of 420 × 10⁶ white blood cells per liter (420 white blood cells per cubic millimeter) and 0 × 10⁶ red blood cells per liter (0 red blood cells per cubic millimeter). Differential on cytocentrifuge preparation included 42% neutrophils, 42% lymphocytes, and 16% monocytes. Cultures for bacteria and fungi were negative for disease. Cerebrospinal fluid was submitted for viral cultures at this time. The patient was treated with intravenous ceftazidime for 7 days, and her fever and symptoms resolved. After her course of antibiotics, she received her eleventh course of chemotherapy and has continued to do well. No additional neurologic sequelae were identified. One year later, she is off chemotherapy but still has residual medulloblastoma.

MATERIALS AND METHODS

Tube tissue cultures of primary rhesus monkey kidneys (MK) were inoculated with 0.2 mL of cerebrospinal fluid and incubated at 36 °C. At 5 to 7 days the inoculated tissue cultures were tested for the presence of a hemadsorbing virus. The main-
tenance medium (Basal Medium Eagle) was removed from the tube and stored. Then 1 mL of Hank’s balanced salt solution and 0.2 mL of 0.4% guinea pig erythrocyte suspension were added. Tubes were incubated at room temperature for 20 minutes and examined for the presence or absence of hemadsorption. Positive cultures were washed twice with 2-mL aliquots of Hank’s balanced salt solution. To remove cells from the culture tube, cells were scraped from the tube wall and centrifuged at 700g for 10 minutes. The pellet of cells was resuspended in about 0.3 mL of residual media and spotted on several pre-cleaned slides containing two 15-mm circles. Slides were air-dried, fixed with cold acetone, and examined for mumps virus using an indirect fluorescent antibody test with mouse monoclonal antibody (Chemicon, Temecula, CA) and a fluorescein-conjugated rabbit anti-mouse serum. When tests were repeatedly negative, culture cells were tested for the presence of other hemadsorbing viruses using a commercial kit (Bartels, Seattle, WA) that contained monoclonal antibodies to parainfluenza viruses types 1, 2, 3, and influenza A and B viruses. The isolated virus was positive for parainfluenza virus type 3 and negative for the others.

The isolate then was sent to Centers for Disease Control for confirmation using different laboratory procedures. The virus was grown in NCI-H292 cells, a new cell line sensitive to all the human paramyxoviruses, and virus growth was documented both by cytopathologic studies and by hemadsorption with guinea pig erythrocytes. The isolate then was typed as parainfluenza virus type 3 by hemagglutination-inhibition tests using reference horse antisera using a standardized protocol. Both the MK isolate and the NCI-H292 passage then were identified by the highly sensitive time-resolved fluorimunoassay with monoclonal antibodies. The time-resolved fluorimunoassay was set up with monoclonal antibodies to parainfluenza virus types 1, 2, 3, 4, mumps, and respiratory syncytial virus, and only the parainfluenza virus type 3 was positive.

**DISCUSSION**

Parainfluenza virus type 3 is a common respiratory pathogen that is rarely recognized as an agent of aseptic meningitis. In the literature, parainfluenza virus type 3 has been isolated from the CSF of eight children: five with fevers, one with muscular hypotonia, and one with convulsions. The only reported death occurred from disseminated disease in a child with severe combined immune deficiency. Parainfluenza virus type 3 has been isolated from one adult with meningitis, one adult with a demyelination syndrome, and from the CSF (without CSF pleocytosis or meningitic signs) in a 19-year-old man with the clinical diagnosis of Guillain-Barre syndrome. Isolation from pharyngeal swabs has been reported in children with seizures and Reye’s syndrome.

Table 1 summarizes and compares our six individuals, in whom parainfluenza virus type 3 was isolated from the CSF. The cases covered the 10-year period between 1981 and 1991.

Our isolates have come from both children and adults with evidence of meningitis. The meningitis caused by parainfluenza virus type 3 seems to be benign and self-limited in otherwise healthy individuals. Even a child who was immunosuppressed by chemotherapy recovered within 1 week and continued her systemic chemotherapy without untoward sequelae.

Aseptic meningitis due to parainfluenza virus type 3 does not seem to be restricted to one locale. These isolates were collected during the summer in the warm climates of New Orleans, Kansas, and Trinidad, and through late fall in Minnesota. Two cases occurring in Minnesota within 6 weeks of one another were not geographically related. The three cases occurring in 1991 were probably a random event.

Finally, parainfluenza virus type 3, although a common respiratory virus pathogen, joins one other paramyxovirus—mumps—as a cause of central nervous system infection. Both of these viruses also share the propensity for causing parotitis, with mumps more commonly associated with this syndrome.

Other members of this paramyxovirus group apparently do not infect the central nervous system or the parotid glands. None of us have isolated parainfluenza virus types 1, 2, 4A, or 4B from spinal fluids or from cases of parotitis, nor are there reports of such cases in the literature.

**REFERENCES**