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A neonatal echovirus 11 outbreak in an obstetric clinic

Ju-Hsin Chen[1], Nan-Chang Chiu[1], Jui-Hsing Chang[1], Fu-Yuan Huang[1], Kun-Bin Wu[2], Tsuey-Lih Lin[2]

[1]Department of Pediatrics, Mackay Memorial Hospital, Taipei; and [2]Center for Disease Control, Department of Health, Taipei, Taiwan

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An echovirus 11 outbreak occurred among neonates in an obstetric clinic in November 2003. Thirteen neonates were transferred to our medical center, and all were found to have echovirus 11 infection. Viral studies were performed for 32 other infants born in the clinic during the same period, including 30 asymptomatic neonates and 2 febrile infants transferred to another hospital. Two of the asymptomatic infants had echovirus 11 isolated from rectal swabs. The first patient transferred to our medical center developed extensive hemorrhage and died 6 days later. Three family members of this infant were also proved to have echovirus 11 infections. One other infant had a fulminant course and had residual hepatic impairment. The other infants had no complications. Viral studies in the 24 nursery staff were all negative. This outbreak shows how a neonatal enterovirus outbreak can occur in a nursery, starting from an infected infant in the incubation period. Early recognition and prompt management of an outbreak is important to prevent further spread of the infection.

Key words*:* Disease outbreaks, echovirus infections, hospital nurseries, newborn infant

Enterovirus infection in neonates is difficult to diagnose because of the broad range of clinical syndromes caused by the virus, from nonspecific febrile illness to aseptic meningitis, encephalitis, myocarditis, and necrotizing hepatitis [1]. Enteroviruses are sometimes isolated from the oropharynx or feces of asymptomatic newborns [2]. The virus is transmitted primarily by the fecal-oral route by contaminated hands. Fomites such as tableware, food, or drinking water have also been implicated in transmission. At the peak of infection, it can also be transmitted by droplets. Group B coxsackie virus serotypes 2 to 5 and echovirus 11 are the enteroviruses most frequently associated with overwhelming systemic neonatal infection [3-6]. Vertical transmission usually occurs at birth by cervical or fecal contamination or occasionally by postnatal exposure. Horizontal transmission is usually through direct or indirect contact by the fecal-oral route and respiratory secretions [6]. Most neonatal outbreaks have been reported from the neonatal unit of a hospital. Here we describe a neonatal enterovirus infection outbreak in the nursery of a standalone obstetric clinic.

Corresponding author: Dr. Nan-Chang Chiu, Department of Pediatrics, Mackay Memorial Hospital, 92, 2nd Section, Chung-Shan North Road, Taipei, Taiwan.
E-mail: ncc88@ms2.mmh.org.tw

Materials and Methods

Clinical setting

A standalone obstetric clinic had a nursery with 10 to 20 beds in 1 large room. The nursing staff moved freely between the delivery room and the nursery. The infants were not assigned a specific nurse; whichever staff member was available to change a diaper or feed or bathe a baby would do so. Babies were brought to their mothers for feeding and then taken back to the nursery. Although they stayed in the same bed, the bed was not necessarily returned to the same spot in the nursery as before.

From October 30 to November 30, 2003 45 babies were delivered in the clinic and were housed in the nursery. They had all been delivered uneventfully and were normal on examination within 24 h of birth except for 1 who had a cleft lip. On November 21, 2 days after being discharged from the clinic, a 5-day-old baby was found to have hyperbilirubinemia, poor activity, and decreased appetite (case 1). The infant was taken back to the clinic and was then transferred to Mackay Memorial Hospital (MMH). On November 24, another baby born in the same clinic was transferred to MMH at 10 days of age also because of hyperbilirubinemia (case 2). The first infant deteriorated after admission to MMH and was thought to have a fulminant viral infection. After the second patient was admitted, we

suspected an outbreak, so we urged the clinic to notify the families of discharged babies. Between November 21 and 30, an infant with a cleft lip and another 10 febrile infants were also transferred to MMH and were placed in strict isolation.

Laboratory investigations

Bacterial cultures were done in 12 of the patients transferred to MMH, excluding only the infant with the cleft lip. In case 1, a rectal swab was done for viral culture. Cerebrospinal fluid (CSF) and throat and rectal swabs for virus isolation were taken in case 2, and from the 10 febrile infants. Throat and rectal swabs for viral culture were obtained 2 weeks later in 7 patients. Rectal swabs for viral culture were also obtained from another 2 febrile infants (transferred to another hospital) and 30 asymptomatic infants born in the same clinic from November 16 to 30.

A neutralization test for serum immunoglobulin G (IgG) to echovirus 11 was performed for all the hospitalized infants except the first. Six had repeat antibody titers measured 2 weeks later. The CSF and rectal swabs in case 2 and the throat and rectal swabs of the other 11 hospitalized infants (i.e., excluding case 1), were sent for reverse-transcriptase polymerase chain reaction (RT-PCR) analysis to detect enterovirus RNA.

Throat and rectal swabs for viral culture were obtained from the mothers in cases 1 and 2. Serum IgG to echovirus 11 was assessed in all the mothers with infants with a proven enterovirus infection. The mother in case 1 had a second examination 2 weeks later. The father, grandmother, aunt, and cousin in case 1 also had viral studies as they either lived together with or had had contact with the baby.

Blood for serum IgG levels to echovirus 11, and throat and rectal swabs for viral culture were obtained from all staff of the clinic, including 2 doctors and 22 nurses.

Results

Clinical presentation

The time course of the outbreak is shown in Fig. 1. Patients 1 and 2 with complicated courses initially presented with hyperbilirubinemia and progressively poorer activity but without fever. However, because the presentation resembled sepsis and was thought to be due to a viral infection, the 2 infants were isolated and

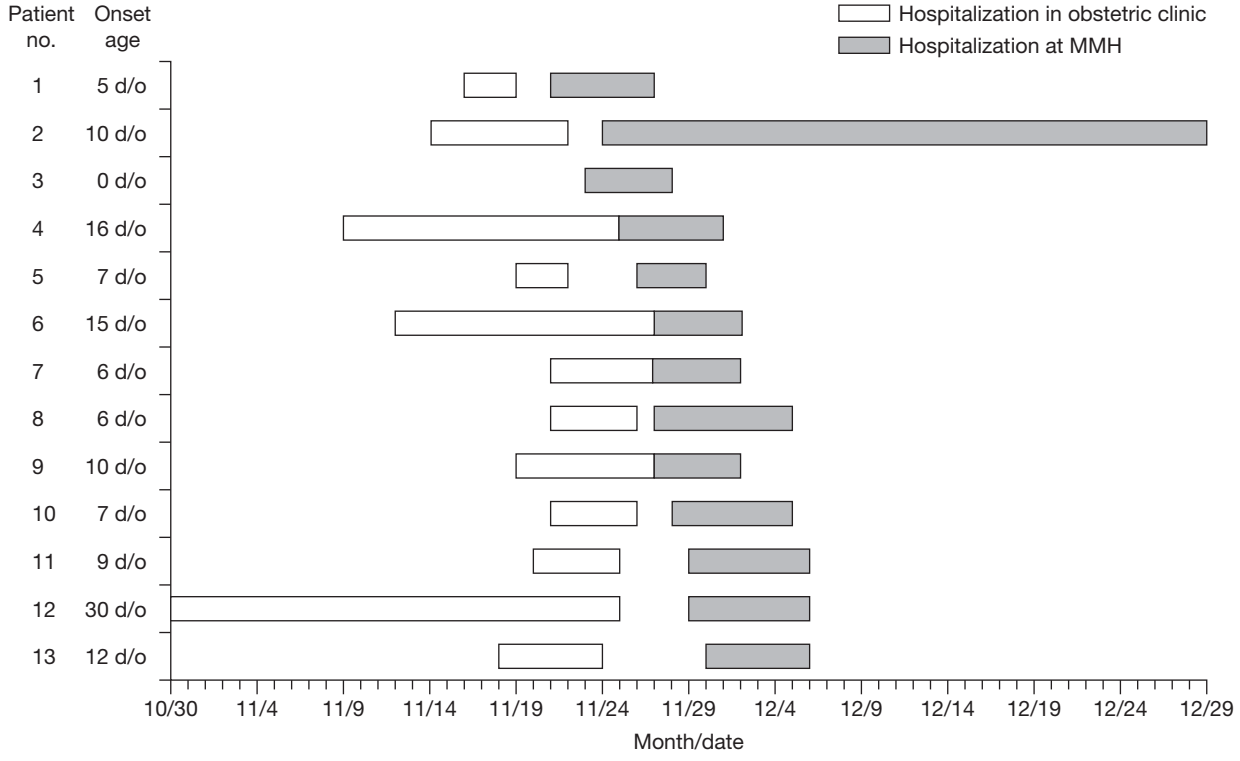

Fig. 1. Time course of a nosocomial echovirus 11 outbreak in neonates. The onset age was defined as the first day of hospitalization in Mackay Memorial Hospital (MMH). d/o = days old.

given intravenous immunoglobulin (1 g/kg). Patient 1 developed respiratory failure, hypotonia, disseminated intravascular coagulopathy (DIC), and bilateral grade IV intraventricular hemorrhage. He died from extensive pulmonary hemorrhage 6 days after admission. Echovirus 11 was isolated from his rectal swab. Patient 2 developed fulminant hepatitis, anemia, aseptic meningitis, and DIC. She was discharged 1 month later with hepatic impairment, which finally resolved after 3 more months.

The third patient was asymptomatic but was transferred to the hospital soon after delivery because of the cleft lip. Echovirus 11 was also isolated from his rectal swab. The other 10 infants, referred to MMH for temperatures higher than 38°C, all recovered completely without sequelae. Their fevers all subsided within 4 days after admission (mean, 1.4 days). None of the 13 patients had skin rash or oral ulcers.

Laboratory data

Data from the neonates

Patients 1 and 2 both had anemia, thrombocytopenia, direct hyperbilirubinemia, coagulopathy, high levels of fibrinogen degradation products and D-dimer, and elevated hepatic and muscle enzymes. All 13 infants had normal C-reactive protein levels.

Bacterial cultures from blood, urine and CSF were uniformly negative in all 13 infants, but echovirus 11 was isolated from 1 or more sources in each case (Table 1). Initial rectal swabs were more likely to yield positive viral cultures than were throat swabs (12/13, 92% vs 7/12, 58%). Seven sets of cultures were repeated 2 weeks later, but virus was grown from only 2 of the rectal swabs and none of the throat swabs. IgG to echovirus 11 was not tested in case 1 but was high (1:1024) in case 2. The antibody titers were negative in the acute phase in the other 11 infants. Five of them had repeat serologic tests, and all had a more than 64-fold increase in the convalescent phase titer. RT-PCR for enterovirus was done in 12 infants and was positive in 11 (92%). CSF pleocytosis was found in 3 patients. CSF enterovirus RT-PCR was performed only in case 2, and it was positive.

Among the other 32 infants born at the clinic during the same period, including the 2 febrile infants managed at another hospital and the 30 asymptomatic babies, only 2 asymptomatic infants had positive rectal swabs for echovirus 11. Therefore, the attack rate for this outbreak was 33.3% (15/45).

Data from family members and obstetric clinic staff

The mother in case 1 had mild diarrhea just before delivery. She had an echovirus 11 infection confirmed by positive throat and rectal swabs (Table 2). Her IgG titer was 1:128 on both initial and repeat testing 2 weeks later. The first serum specimen was collected on November 27, 6 days after the onset of disease in her baby. Viral cultures from the mother in case 2 were negative, but she had a high titer of echovirus 11 IgG (1:1024), measured 9 days after the onset of disease in her baby. The mothers in the other 11 cases had low IgG titers.

Table 1. Viral studies in 13 neonates with echovirus 11 (E11) infection

Case no.	First virus culture[a]		Second virus culture[a]		IgG to E11[a]	
	Rectal	Throat	Rectal	Throat	First	Second
1	E11	NM	NM	NM	NM	NM
2	NG	E11	NG	NG	1:1024	1:1024
3	E11	NG	NG	NG	<1:8	NM
4	E11	NG	NM	NM	1:16	NM
5	E11	NG	NM	NM	1:16	NM
6	E11	NG	NM	NM	<1:8	NM
7	E11	E11	NM	NM	<1:8	NM
8	E11	E11	E11	NG	<1:8	1:512
9	E11	E11	NG	NG	<1:8	NM
10	E11	E11	NM	NM	<1:8	1:512
11	E11	E11	NG	NG	<1:8	1:512
12	E11	E11	NG	NG	<1:8	1:512
13	E11	NG	E11	NG	<1:8	1:512

Abbreviations: IgG = immunoglobulin G; NG = no growth; NM = not measured

[a]Viral cultures and testing for IgG to E11 were done on the first day of hospitalization at Mackay Memorial Hospital and repeated 2 weeks later. The first blood sample was obtained on the first day of disease onset.

Table 2. Viral studies in the mothers of 13 neonates with echovirus 11 (E11) infections

Case no.	Virus culture		IgG to E11	
	Rectal	Throat	First	Second
1	E11	E11	1:128[a]	1:128[a]
2	NG	NG	1:1024[b]	NM
3	NM	NM	<1:8	NM
4	NM	NM	<1:8	NM
5	NM	NM	<1:8	NM
6	NM	NM	<1:8	NM
7	NM	NM	<1:8	NM
8	NM	NM	<1:8	NM
9	NM	NM	<1:8	NM
10	NM	NM	<1:8	NM
11	NM	NM	<1:8	NM
12	NM	NM	<1:8	NM
13	NM	NM	<1:8	NM

Abbreviations: IgG = immunoglobulin G; NG = no growth; NM = not measured

[a]E11 IgG titer was tested in the mother in case 1 six days after her son's disease onset and again 2 weeks later.

[b]E11 IgG titer was tested in the mother in case 2 nine days after her daughter's disease onset.

The results of viral studies in family members in case 1 are shown in Table 3. Echovirus 11 was isolated from rectal swabs of the baby's aunt and cousin. His aunt had measured the baby's rectal temperature the day he became ill. The cousin also had contact with patient 1 at home.

Serum IgG antibodies to echovirus 11 and viral cultures from rectal and throat swabs from the 24 nursery staff members were all negative.

Control measures

After the first patient was admitted to MMH, viral infection was tentatively diagnosed and contact isolation was immediately instituted. Three days later, the second patient was transferred to MMH, at which point the obstetric clinic was notified of an apparent nosocomial outbreak. Enhanced hygiene measures and close observation of infants were recommended. The clinic disinfected the nursery, isolated subsequent newborns to a separate zone, and discontinued rooming in to prevent further cross infection. They also informed the families of infants already discharged who had been born after November 16, i.e., 5 days before disease onset in case 1. Some of those babies had a fever and were thus admitted to MMH. When echovirus 11 was first isolated on November 27 in case 1, the clinic stopped all deliveries. They closed on November 30 after sending all remaining babies home or to hospitals. They then repeated disinfection of the environment and built a new isolation nursery.

In MMH, control measures included strict contact isolation, enhanced hand washing, and the use of masks, gowns and gloves. The first 2 patients with complicated disease were cared for in 2 isolation rooms in the neonatal intensive care unit. The other 11 infants, clustered in 2 isolation rooms, were separated from other patients in the newborn center and cared for by a different group of nurses. No new, uninfected infants were admitted to these rooms until all echovirus-infected babies had been discharged. The Taiwan Center for Disease Control (CDC) conducted the investigation of the outbreak.

Table 3. Viral studies in the family members in case 1

Relationship	Virus culture		IgG to E11[a]	
	Rectal	Throat	First	Second
Mother	E11	E11	1:128	1:128
Father	NM	NM	<1:8	1:8
Grandmother	NM	NM	1:16	<1:8
Aunt	E11	NM	1:1024	1:512
Cousin	E11	NM	NM	NM

Abbreviations: IgG = immunoglobulin G; E11 = echovirus 11; NM = not measured

[a]The paired sera were collected 2 weeks apart.

Discussion

In Taiwan, it is common for standalone obstetric clinics to have a nursery for the babies born there. While outbreaks of neonatal enterovirus infection have been reported in hospitals, one has never before been reported in such a clinic. This episode demonstrates the potential for enterovirus to cause serious illness among newborns — including fever, aseptic meningitis, hepatitis, and coagulopathy, as well as asymptomatic infection [6]. Disseminated enterovirus infection in neonates has a clinical picture indistinguishable from bacterial sepsis [7]. Multiple organ involvement is a hallmark of enterovirus infection, and its presence should prompt early isolation. Coagulopathy is a well-described complication of enterovirus sepsis, reflecting a combination of DIC, hepatocellular damage with consequent insufficient production of clotting factors, and viral-induced thrombocytopenia [8]. Hepatic necrosis with coagulopathy is a predictor of poor outcome in neonatal enterovirus infections. Lin et al suggested that prematurity, maternal history of

illness, earlier age of onset, higher leukocyte count, and a lower hemoglobin concentration were factors significantly associated with hepatic necrosis, while coagulopathy, higher total bilirubin, and concurrent myocarditis were significantly associated with fatality [9]. The majority (77%) of our patients with unspecified febrile illness recovered fully without any sequelae. The first 2 babies who presented with jaundice but without fever, however, developed severe complications, and 1 died.

A survey of pregnant women for enterovirus suggested that transplacental passage of enterovirus does not occur readily [10]. However, enterovirus has been isolated from placenta and from the tissue of aborted fetuses [11,12]. Intrauterine enterovirus infections during pregnancy are possible but rare. Most vertical transmission occurs during delivery.

The incubation period of enterovirus infection is usually 3 to 5 days, so that infants with disease onset between 3 and 8 days of age are likely to have acquired the infection from their mothers at the time of birth [13]. The outcome is strongly influenced by the presence or absence of passively acquired maternal antibody specific to the infecting enterovirus serotype. Thus, the length of time between the development of maternal antibody and delivery of the infant may be the most critical factor in determining the outcome of vertical transmission [3]. Like the fulminant course in case 1, most severe infections occur in neonates 3 to 8 days after birth who have no protective maternal antibody.

Attack rates of clinical disease in nursery enterovirus outbreaks have ranged from 22% to 52%, and the illness has generally been mild [4]. In one reported outbreak of neonatal echovirus 11 infection, no intervention was undertaken except to emphasize the importance of hand washing; all infected neonates recovered well [14]. However, in another report, 1 of 10 infected infants died [15]. While supportive care may be enough for most patients, in infants at risk for severe or fatal infection, intravenous immune globulin may be life-saving.

In the present episode, all infants had a good outcome except for the first 2. Patient 2 became ill at the age of 10 days, and when tested 9 days later, her mother was found to have a high IgG titer, although no virus was isolated from the mother's rectal or throat swabs. Based on the age at onset, the time sequence, and the mother's negative culture results, it is likely that this infant was infected by horizontal transmission in the nursery, although postnatal vertical transmission cannot be ruled out completely.

Early diagnosis is crucial to identify and end an outbreak. However, while virus culture is the gold standard for diagnosis of viral infection, the results take some time to appear. Serologic testing may confirm an infection, but again it is not particularly helpful in the acute stage. The best test for early diagnosis is RT-PCR [16,17], and phylogenetic sequencing is useful for tracing sources of transmission [18,19].

According to the age at onset, the time sequence, and positive culture results in the mother, we suspect that patient 1 was the index patient and was infected by his mother at birth. The mother's echovirus 11 IgG titers were the same when first drawn and 2 weeks later. Because the first serum specimen was collected 6 days after her baby became ill, it is possible that she was already in the convalescent phase, explaining the absence of an increase in the IgG titer in the second serum sample. A more definitive answer might have been possible if the cord blood or a vaginal swab from the mother could have been tested for virus or if maternal antibodies had been assayed earlier. However, these are not routine studies, and no problem was suspected until the infant became ill 5 days after birth. Phylogenetic analysis of the VP1 RNA sequences from some of the viral isolates was done by the Taiwan CDC laboratory. The RNA sequence was compared with that of previous isolates from the Taiwan CDC and the Gene Bank using Molecular Evolutionary Genetics Analysis. This demonstrated that all the echovirus 11 isolates fell within the same branch of the tree with more than 97% similarity [20]. Therefore, the outbreak was most likely due to horizontal transmission to other infants from the same infectious focus.

None of the staff of the obstetric clinic had evidence of echovirus 11 infection, but horizontal transmission by way of their hands is still possible. Enterovirus can be isolated for a longer period from rectal swabs than from throat specimens. Efficient hand washing after changing diapers is vital to prevent viral spread. This is mandatory both to prevent direct infant-to-infant spread as well as to minimize environmental contamination. Enteric viruses can persist for extended periods on several types of materials commonly used in both institutional and domestic environments [21]. This particular outbreak was rapidly controlled with the cooperation of the obstetric clinic, MMH and CDC. In Taiwan, some obstetric clinic staff may not be well trained in infection control procedures, posing a danger both to mothers and their babies in the nursery. Since an outbreak of a neonatal enterovirus infection may

begin, as this one apparently did, from an asymptomatic infant incubating an infection, clinic staff must be trained to recognize and intervene promptly to control an outbreak before it spreads further.

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