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Cluster of necrotizing enterocolitis in a neonatal intensive care unit: New Mexico, 2007

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Background: Although the cause of necrotizing enterocolitis (NEC) is unknown, infection control practices have been shown to play an important role in containing many outbreaks. We investigated the etiology of a cluster of NEC in a level 3 neonatal intensive care unit and monitored for new cases following the implementation of enhanced infection control measures.
Methods: Investigators performed a chart and laboratory review for neonates with a diagnosis of NEC during January 1, 2007, to February 13, 2007, to identify risk factors. Enhanced environmental cleaning, cohorting of infants and nurses, and increased attention to hand hygiene were instituted. Commercial feeding products in the unit were tested for bacterial contamination. Close monitoring for new cases continued for 2 months following the identification of the cluster.
Results: Eleven cases of NEC were identified during the study period. Patients had a median of 5 disease risk factors (range, 3-8). Four distinct pathogens were detected in blood or stool specimens from 4 different patients. One sample of human milk fortifier (HMF) tested contained a colony count of *Bacillus cereus* at the US Food and Drug Administration's upper microbiologic limit for contamination. Seven (65%) patients received HMF before symptom onset, and 9 (82%) patients received 1 or more types of liquid formula. Only 1 new case was identified during the period of close monitoring.
Conclusion: A microbiologic cause was not identified, and, although the cluster might have resolved spontaneously, enhanced infection control and changing batches of HMF might have played a role in controlling this outbreak.
Key Words: Human milk fortifier; outbreak; infant; case series; infection control.

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Necrotizing enterocolitis (NEC) is one of the most common serious gastrointestinal diseases among newborns[1] and is a major cause of morbidity and mortality among preterm infants.[2] Incidence of NEC nationally in neonatal intensive care units (NICUs) has ranged from 1 per 100 infants to 5 per 100 infants,[3,4] and the case fatality rate has been estimated to range from 10% to 30%.[5] Although the pathogenesis is not understood, it is likely multifactorial (eg, prematurity, aggressive feeding regimens, infection, and inflammation).[2,3] Neonates experiencing NEC typically present with intestinal symptoms (including feeding intolerance, bloody stool, and abdominal distention), systemic symptoms (including apnea, bradycardia, temperature instability, and septic shock), and characteristic radiographic findings of pneumatosis intestinalis. Limited progress has been achieved in delineating the etiology of NEC because both older[3] and recent[2] articles have described its enigmatic etiology. The majority of cases is sporadic; however, the occurrence of clusters indicates an infectious component to the disease.[1] Multiple causative organisms have been proposed, including coagulase-negative *Staphylococcus* species, various gram-negative bacilli, and *Clostridium* species. However, in approximately 35% of published outbreaks, no etiologic organism was identified.[1]

This report summarizes an investigation of a NEC cluster among 11 premature infants in a NICU in New Mexico from January 22 to February 13, 2007. Personnel from the New Mexico Department of Health (NMDOH), the hospital in which the cluster occurred, and the Centers for Disease Control and Prevention conducted an investigation to identify the cause of

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A. M. W. had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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this cluster, evaluate risk factors associated with development of NEC, and implement appropriate control measures.

MATERIALS AND METHODS

This cluster of NEC was identified by the physicians on February 5, 2007, after the first 5 cases were diagnosed. Assistance from NMDOH was requested on February 12, 2007. Cases were defined according to modified Bell's staging.[1,6] (In 1978, Bell et al[7] developed a uniform staging system for NEC, which was modified by Walsh et al[6] in 1986 to more explicitly describe the systemic, intestinal, and radiographic signs.) Annual rates of NEC at this hospital were calculated for 2005, 2006, and 2007. Medical charts for each case-patient were reviewed, and the following information was collected based on factors identified from previous reports of NEC clusters and the judgment of the clinical staff and investigative team: date of birth; estimated gestational age; birth weight; sex; admission date; APGAR score; date of NEC onset; NEC staging and accompanying signs and symptoms; previous diagnosis of NEC; detailed feeding history for ≤7 days before symptom onset; antimicrobial therapy and other pharmaceutical treatments; umbilical catheter; surgical treatment; hyaline membrane disease; and maternal risk factors, including prenatal care, gestational diabetes, drug abuse, and preeclampsia. Information regarding bed location and number of nurses and other health care providers caring for case-patients during the 2 days before symptom onset was also collected. Enhanced surveillance for NEC was conducted for 2 months following the last case of the cluster.

All clinical samples were processed through the hospital laboratory. Blood cultures were ordered for each case-patient, and, after the outbreak was detected, stool was tested for the following pathogens: *Clostridium difficile* via toxin test (n = 7), norovirus (n = 7), rotavirus (n = 6), and other enteric pathogens (n = 6). Information regarding test results and their accompanying dates was collected from hospital medical records. Anaerobic testing of stool or environmental samples was not available through the New Mexico Scientific Laboratory Division or local private laboratories limiting the ability to identify anaerobic pathogens.

When the cluster was identified, the nursing and dietary staff immediately isolated all remaining commercial feeding products for possible testing. Human milk fortifier (HMF) (Enfamil; Mead Johnson, Glenview, IL) and single-use liquid formula with and without iron in 20- and 24-calories/oz formulations were among the isolated products and were sent to New Mexico Scientific Laboratory Division for aerobic bacterial contamination testing. HMF, used to increase levels of nutrients and calories in expressed breast milk, was available in single-use, 0.71-g packets of dry powder. Because of the limited volume in each HMF packet, contents from multiple packets sharing the same prefix lot code were combined to produce sufficient volume for testing. Microsoft Excel, 2003, (Microsoft Corp, Redmond, WA) was used for all data management, descriptive analyses, and cluster analysis.

RESULTS

The rate of NEC in this NICU during the outbreak period of January 22 to February 13, 2007, was 16.9 of 100 infants, compared with 3.3 of 100 infants and 2.4 of 100 infants in 2006 and 2005, respectively. (The annual rate of NEC decreased to 3.4 of 100 infants by the end of 2007 in this NICU.) The median estimated gestational age of case-patients was 33 weeks (range, 27-37 weeks), and the median time from birth to NEC onset was 8 days (range, 5-81 days). Seven case-patients were male, and 4 were female. Distribution of staging was as follows: 3 stage I (suspect), 5 stage II (definite), and 3 stage III (advanced). All patients experiencing advanced NEC required surgical resection, and 1 died. No other infants died.

Health care providers maintained a heightened index of suspicion (increased awareness of Bell's stage I criteria) for NEC for 2 months from the last identified patient. One case of NEC was identified 22 days (March 7) after the last case associated with the cluster. Although data were collected regarding this infant (by using 2 months of surveillance data), we decided to exclude it from the cluster under investigation because of the relatively substantial time between the preceding cluster and this case. The epidemic curve summarizing the time line for this outbreak is depicted in Fig 1, stratified by NEC staging. Each patient had multiple, previously recognized risk factors for NEC (Table 1). Patients had a median of 5 (range, 3-8) risk factors for NEC. The median 1-minute (and 5 minute) APGAR scores for stages I, II, and III were 7 (8), 7 (9), and 6 (8), respectively. No appreciable patterns were detected regarding antibiotic use or other medications prescribed. Three of the infants' mothers (1 from each stage) had <5 prenatal care visits. A total of 48 nurses cared for the patients in the 2 days before diagnosis. No single nurse or physician was identified as a common caregiver to all case-patients. No other health care providers were identified as potential common sources. Case-patients were distributed throughout the NICU.

Three different organisms (*Klebsiella pneumonia*, *Staphylococcus aureus*, and *Enterobacter hormaechei*)

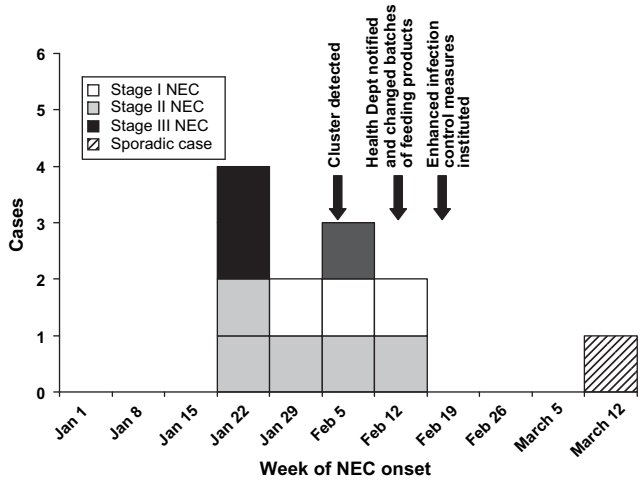

Fig 1. Number of necrotizing enterocolitis (NEC) cases (n = 11) by stage in a neonatal intensive care unit in New Mexico, January-March 2007.

Table 1. Risk factors for necrotizing enterocolitis patients by stage in a neonatal intensive care unit in New Mexico, January 22 to February 13, 2007

	Stage I (n = 3)		Stage II (n = 5)		Stage III (n = 3)	
Potential risk factors for NEC	**n**	**%**	**n**	**%**	**n**	**%**
Infant risk factors						
Human milk fortifier	0	0	4	80	3	100
Liquid formula	2	67	5	100	2	100
Very low birth weight (<1500 g)	2	67	2	40	3	100
Very low gestational age (≤32 wk)	1	33	1	20	3	100
Small for gestational age	1	33	1	20	0	0
Umbilical catheter	2	67	2	40	3	100
Hyaline membrane disease	1	33	1	20	3	100
Positive blood culture	0	0	1	20	2	66
Positive *C difficile* toxin test	0	0	1	20	0	0
Previous NEC episode	1	33	0	0	0	0
Maternal risk factors						
Gestational diabetes	1	33	2	40	0	0
Preeclampsia	1	33	2	40	0	0
Drug abuse	0	0	1	20	1	33

NEC, necrotizing enterocolitis.
NOTE. Number of identified NEC patients: n = 11.

were isolated from blood cultures from 3 different patients, and *C difficile* toxin was detected in a fourth patient. No stool cultures were positive for enteric pathogens.

Infant formula and HMF lot codes were not routinely recorded or tracked by the NICU staff. Seven (64%) of the case-patients received HMF before symptom onset, and 9 (82%) affected infants received 1 or more types of liquid formula. One affected infant was fed an amino acid-based formula and was not exposed to HMF or standard formula.

Of the 4 lot codes of HMF tested, 1 contained 100 colony-forming units (cfu)/g of *Bacillus cereus*, 1 contained 10 cfu/g of *B cereus*, and 2 contained <10 cfu/g of *B cereus*. No other bacteria were recovered from HMF, and all single-use liquid formula sampled tested negative for bacterial contamination. None of the infant blood specimens tested positive for *B cereus*, and stool samples were not specifically cultured for *B cereus* because it is not part of the standard microbiologic panel and there was no stool available for additional testing once the environmental tests were performed. The finding of 100 cfu/g of *B cereus* in 1 sample is at the US Food and Drug Administration's (FDA) upper microbiologic limit for healthy term infants. No limit is available for preterm infants.

We recommended that patients be placed under contact isolation and reemphasized the need for strict adherence to hand hygiene and appropriate use of personal protective equipment for both health care personnel and visitors. A 1-time thorough environmental cleaning of the entire NICU was performed with a 1:10 dilution of household bleach (ie, 5000 parts/million of hypochlorite), and infants and nurses cohorted when possible. The hospital also elected to change to new lot codes of HMF and formula. Only 1 isolated case of NEC (previously mentioned) was detected 3 weeks following the implementation of these infection control measures; however, the NEC cluster possibly would have spontaneously resolved regardless.

DISCUSSION

A cluster of NEC in a New Mexico NICU was detected and investigated. No single pathogen was identified, a fact that might not be surprising given the likely multifactorial etiology of NEC. NICU staff initiated the following additional infection control measures: enhanced cleaning of all surfaces and instruments by using a sporicidal disinfectant, increased attention to hand hygiene, isolating patients until outbreak termination, staff cohorting, and increased index of suspicion for new cases.

This report highlights the need for systematic diagnostic testing to cover the spectrum of potential causal organisms when investigating nosocomial outbreaks of NEC, especially for those organisms not routinely isolated by standard blood or stool culture (eg, anaerobic bacteria and *B cereus*). Three blood cultures were positive with 3 different organisms (*K pneumonia*, *S aureus*, and *E hormaechei*). This finding would not be unusual for sporadic cases of NEC, but, given the clustering of these cases, a common exposure increasing the risk of sepsis with the identified organisms is possible. Although comprehensive testing might not always be necessary in sporadic cases of NEC, it is critical in establishing the etiology when a substantial cluster or

outbreak is identified. Specifically, testing for anaerobic pathogens (eg, *Clostridium* species), coronavirus, rotavirus, and echovirus might be added to the initial set of diagnostic tests because they have been implicated in past outbreaks.[1] Unfortunately, systematic testing was not performed on all infants in this cluster. Echovirus and coronavirus testing were not performed, stool cultures were not ordered for the first 5 patients, and tissue samples were not available. The only testing for *Clostridium* species consisted of *C difficile* toxin testing. The clinical significance of a positive *C difficile* toxin test in this age group is uncertain.

The potential role of HMF in this outbreak cannot be excluded. Specifically, prematurity is a widely reported risk factor for NEC,[3] which might be associated with prematurity of the gastrointestinal tract.[2,3,8] In addition, formula-fed infants are reported to have 6 to 20 times the risk of experiencing NEC compared with breast milk-fed infants.[9-11] An increased risk of NEC among preterm infants who were fed HMF has also been reported,[12,13] and contamination[14] (including with *B cereus*[15,16]) has been postulated as a potential mechanism. In this investigation, 1 lot code of sequestered HMF tested positive for *B cereus* at the FDA's upper microbiologic limit (100 cfu/g).[17] Risk assessment models have concluded that powdered infant formula containing ≤100 cfu/g of *B cereus* and reconstituted with cooled boiled water (25 °C) and stored for ≤24 hours at ≤10 °C would not expose term infants to an infectious dose of *B cereus*; however, the risk to premature infants is unknown. Thus, because of similarities in ingredients and manufacturing processes between powdered formula and HMF, potential to improperly reconstitute and store fortified breast milk, and the unknown risk that *B cereus* poses to premature infants, a common mechanism might be shared by formula and HMF and warrants additional examination. Because anaerobic cultures of HMF were not performed, isolation of *B cereus* might be a marker of similar or even greater levels of contamination with anaerobic spore-forming organisms (eg, *Clostridium* species), which have been previously indicated in the etiology of NEC.[18] In this instance, the volume of HMF was insufficient to conduct both aerobic and anaerobic cultures.

Findings in this report are subject to some limitations. First, pathogens previously implicated in certain NEC outbreaks were not uniformly tested for in this investigation, in part because an outbreak was not apparent until 5 cases had been identified. This includes the lack of cultures for anaerobic bacteria in both clinical and food product specimens. Second, no tissue samples from patients' gastrointestinal tracts were available for confirmatory testing. Third, a case-control study would have allowed us to better characterize risk factors for NEC (including HMF) in this outbreak. (A case-control study was not conducted because of the insufficient power this small number of cases would have yielded.) Despite these limitations, this investigation demonstrates that considering other risk factors is critical when investigating NEC clusters and might serve as the basis for conducting more rigorous studies to evaluate the role of HMF consumption in the pathogenesis of NEC.

No new cases in this NEC cluster occurred after institution of enhanced infection control interventions and switching lot codes of HMF and formula fed to the neonates; however, no pathogens discovered in HMF were identified in any of the infants, and no causal associations were made to explain this temporal association. Because powdered formula products, including HMF, are not sterile, careful attention should be given to contraindications for their use. NICU staff should consider documenting lot codes of nutritional products administered to their patients. Strict adherence to proper infection control practices is a key recommendation in controlling NEC clusters.[1] Systematic testing of blood, stool, and tissue specimens (when available) is necessary for better understanding of the etiology of NEC clusters.

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