Synchronous Recurrence of Group B Streptococcal Late-Onset Sepsis in Twins

abstract

Group B *Streptococcus* (GBS) remains the leading cause of neonatal sepsis and meningitis in industrialized countries. Whereas the use of intrapartum antibiotic prophylaxis has led to a significant decline in early-onset sepsis, the incidence of late-onset sepsis has remained unchanged. Whether late-onset sepsis usually originates from established mucocutaneous GBS colonization of the infant or whether it results from an acute exogenous GBS infection remains controversial. Here we report on twins who both twice developed GBS sepsis in a strikingly parallel fashion, with both instances originating from a single hypervirulent GBS clone. Factored together, the presentation as cervical soft tissue infection in both cases, the synchronicity of the episodes, and the detection of GBS DNA in breast milk all strongly suggest an enteral mode of transmission with a short incubation period. *Pediatrics* 2014;133:e1388–e1391

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KEY WORDS

sepsis, late-onset sepsis, *S agalactiae*, group B streptococcus, breast milk

ABBREVIATIONS

CSF—cerebrospinal fluid
EOS—early-onset sepsis
GBS—group B *Streptococcus*
LOS—late-onset sepsis

Dr Elling was involved in patient care including immunologic workup of the patients, analyzed patient charts and microbiologic data, and wrote the manuscript together with Dr Henneke; Dr Hufnagel initiated specimen collection, contributed to the concept of the study, supervised the scientific workup, and critically reviewed and revised the manuscript; Dr de Zoysa conducted molecular analysis of the breast milk specimens and reviewed the initial draft of the manuscript; Mr Lander was involved in patient care and analysis of the microbiologic data and reviewed the initial draft of the manuscript; Ms Zumstein performed bacterial characterization and reviewed the manuscript; Dr Krueger was responsible for the intensive care treatment of the patients and provided critical input on the manuscript; Dr Henneke initiated, conceptualized, and supervised the study; he was responsible for the immunologic workup of the patient and wrote the manuscript together with Dr Elling; and all authors approved the final manuscript as submitted.

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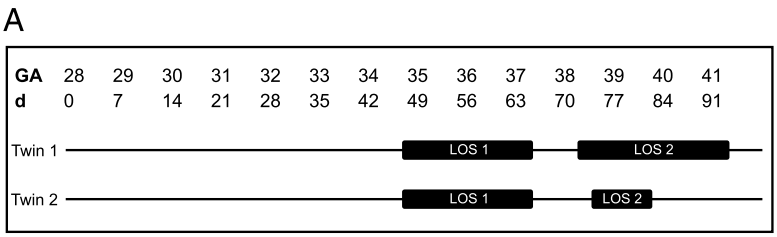
Group B *Streptococcus* (GBS; *Streptococcus agalactiae*) is the most important cause of sepsis in newborn infants who have no underlying disease.[1] GBS causes early-onset sepsis (EOS) in the first week of life (age 0–6 days) and late-onset sepsis (LOS) after the first week of life (age 7–89 days). For EOS, transmission is usually vertical via GBS-contaminated amniotic fluid or vaginal secretions. By contrast, both the mode and time point of GBS transmission in LOS remain unclear. Mother-to-child transmission seems plausible, but nosocomial transmission also has been described.[2] It is particularly ambiguous whether mucocutaneous colonization of the infant precedes invasive disease by days or even weeks, or whether LOS occurs after acute transmission of bacteria. However, understanding GBS transmission in LOS is important because it would enable the implementation of effective LOS prevention strategies. In recent years, it has been suggested that breast milk may be a potential source for GBS late-onset disease. This hypothesis has been based on the isolation of the same GBS serotype or clone from breast milk as well as from the affected infant.[3–5] Yet, these observations provide only modest evidence for a causal relationship between breast milk contamination and LOS. Women are usually colonized by a single GBS clone.[6] Moreover, GBS can be isolated from breast milk samples in up to 10% of healthy breastfeeding mothers.[7] Furthermore, rectovaginal colonization densities vary substantially over time.[6] Accordingly, GBS isolates from mother and infant are usually clonally related, independently of whether neonatal GBS colonization occurs around the time of birth and then persists, or whether GBS is acquired later.

Here, we report on premature twins, both of whom suffered from severe recurrent GBS LOS with similar clinical features and striking synchronicity of these episodes. All 4 sepsis episodes were characterized by cervical soft tissue infections accompanied by systemic disease. Taken together, the sequence of events, the almost uniform clinical manifestation, and the detection of GBS in the breast milk of the nursing mother strongly suggest that GBS LOS can occur as an enteral infection with a short incubation period.

PATIENT PRESENTATIONS

Dizygotic male twins (gestational age: 28 0/7 weeks; birth weights: 1125 and 1190 g) were born to a 31-year-old primigravida by cesarean delivery 24 hours after premature rupture of amniotic membranes. The mother received cefuroxime antenatally. Antenatal vaginal swabs and urine samples were negative for GBS. Empirical antibiotic therapy with piperacillin and tobramycin was initiated in both infants for suspected perinatally acquired sepsis. Therapy was stopped when blood cultures remained sterile. Enteral feeding with breast milk was started on the fifth day of life. On day 48 of life, twin 2 suddenly deteriorated and developed irritability, paleness, respiratory failure requiring mechanical ventilation, and a rapidly enlarging left-sided cervical swelling. Six hours later, twin 1 developed similar clinical signs; yet in this second case, the cervical swelling was right-sided. Laboratory workup in both infants revealed leucopenia, thrombocytopenia, and an increase in immature granulocytes. Blood cultures grew GBS in both patients. Cerebrospinal fluid (CSF) culture was sterile in twin 2 but was positive for GBS after enrichment in twin 1. Because CSF cell count and chemistry were unremarkable, growth of GBS probably resulted from minimal contamination of the CSF with blood during the lumbar puncture. In both twins, ultrasound examination of the cervical swelling revealed diffuse tissue infiltration consistent with cellulitis. Both infants received initial empirical treatment with meropenem and vancomycin before antibiotics were switched to penicillin G (twin 1: 347 000 U/kg per day; twin 2: 340 000 U/kg per day) plus tobramycin on the basis of blood culture results. The total duration of antibiotic therapy was 19 days. Both twins recovered completely and were discharged 4 days after cessation of antibiotic therapy. However, only 3 days after discharge, twin 1 presented with clinical sepsis and with a new cervical swelling and reddening on the left side (Fig 1B), in contrast to the initially right-sided episode. Laboratory parameters (leucopenia, thrombocytopenia, and bandemia) and clinical course (mechanical ventilation and circulatory support) were reminiscent of the first GBS sepsis episode. CSF analysis was unremarkable; however, a blood culture again grew GBS. Less than 48 hours later, twin 2 presented with decreased oral intake and a right-sided (as opposed to the initially left-sided) cervical skin erythema without substantial swelling. A blood culture for twin 2 remained sterile. Antibiotics were administered for 19 days to twin 1 (piperacillin plus tobramycin followed by penicillin G) and for 10 days to twin 2 (piperacillin plus tobramycin). Again, both twins recovered completely.

Due to the recurrent severe infections, further microbiologic and immunologic workup was performed. Repeated culturing of breast milk samples expressed by different electronic pumps never grew GBS or any other potentially pathogenic bacteria. However, the GBS-specific *CYLB* gene was detected by real-time polymerase chain reaction in independent milk samples from both breasts (for method, see ref 8). Rectovaginal sampling from the mother was repeated during the first LOS episode and was positive for GBS. This



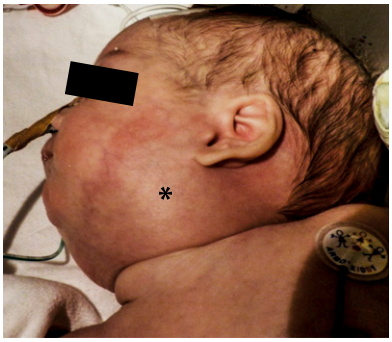


FIGURE 1

A, Time line of recurrent GBS sepsis. The infants' age is depicted at the top as gestational age (GA; in weeks) and postnatal days (d). The black bars indicate the duration of antibiotic treatment during both episodes of LOS (1 and 2). B, Clinical picture of twin 1 during the second LOS episode with left-sided cervical cellulitis (*).

isolate of the mother and the 3 blood culture isolates of the twins belonged to the same hypervirulent GBS serotype III and sequence type ST-17 (Table 1). Additionally, real-time polymerase chain reaction revealed expression of the 3 virulence genes *CYL*, *SRR1*, and *HVGA* in all strains (data not shown). Recently discovered as a critical virulence factor of the ST-17 clone, the *Hvga* protein mediates adherence to epithelial cells and is required for bacterial translocation across the intestinal barrier.[9]

GBS sepsis was shown to be a putative initial manifestation of IRAK4 deficiency,[10] a primary immunodeficiency syndrome which affects Toll-like receptor signaling. Accordingly, Toll-like receptor signaling was tested in peripheral monocytes and revealed a normal cytokine response in both twins. Furthermore, granulocyte function (formation of reactive oxygen species and adhesion) was normal (data not shown).

After counseling on the potential risk of breast milk transmission of GBS, the mother chose to discontinue breastfeeding. At 20 months of follow-up after the second set of sepsis episodes, both twins remain well and no further infectious complications have occurred.

DISCUSSION

GBS LOS often presents with meningitis and remains a devastating disease, with a mortality rate of up to 10% and permanent neurologic sequelae in 25% to 35% of meningitis survivors.[9] Unfortunately, however, the design of effective prevention strategies for LOS remains hampered by incomplete understanding of the disease pathogenesis.

The cases presented here are instructive for several reasons. First, they strongly suggest that the cause of LOS may be an exogenous infection with a short incubation period. Early intestinal GBS colonization with later translocation and development of sepsis would not be consistent with the observed synchronicity of the infectious events described here (Fig 1A). Second, our report implicates breast milk as a source of GBS infection, thereby supporting previous reports on this issue.[3–5] The transmission of GBS via contaminated hands appears to be unlikely in our cases, because the mother strictly adhered to hygiene procedures, including alcoholic disinfection of hands and breasts before breastfeeding or using the breast pump.

In the cases presented here, GBS DNA was found in the breast milk, even though cultures remained sterile. This finding may be due to large differences in GBS density in breast milk over time (similar to what has been observed with rectovaginal swabs[6]), due to generally low GBS concentrations in breast milk or due to the growth-inhibiting effect of unpasteurized breast milk on GBS, a discovery made 35 years ago.[11]

Prematurity is an important risk factor for LOS.[12] Most likely contributive to this risk are alterations of the adaptive immune system, such as decreased immunoglobulin G levels and a slower maturation of the V_H gene repertoire in premature infants.[13] The idea that young infants in general may be exquisitely sensitive to small GBS inocula, for example, in breast milk, is supported by a GBS sepsis model in neonatal mice, in which subcutaneous injection of 15 bacteria was lethal for 20% of the challenged mice.[14] Furthermore, 60% to 70% of preweaned mice (15–21 days old) succumbed to an

TABLE 1 Source, Serotype, and Clonal Subtype of Isolated GBS Strains

Patient	Source	GBS Serotype	GBS Sequence Type
Mother	Rectovaginal swab	Serotype III	ST-17
Twin 1	Blood culture/CSF, LOS 1	Serotype III	ST-17
Twin 2	Blood culture, LOS 1	Serotype III	ST-17
Twin 1	Blood culture, LOS 2	Serotype III	ST-17

enteral infection with *HvgA*-expressing GBS, whereas mice ≥4 weeks are protected.[9]

The cases described here support a model in which some GBS-exposed infants develop sepsis quickly, likely via translocation of GBS across the bowel wall and subsequent spread to lymph nodes and blood, whereas others become healthy carriers without development of disease. The observation that intrapartum antibiotic prophylaxis, which transiently reduces GBS colonization of the mother, does not reduce the incidence of LOS indicates that in most LOS cases GBS is likely to have been acquired after birth.[15] Furthermore, the kinetics of GBS EOS, wherein 80% of cases show symptoms within 24 hours after birth, supports the idea that GBS is a virulent neonatal pathogen that rapidly elicits symptoms.[16]

Systematic studies involving analysis of GBS in breast milk are necessary to elucidate the prevalence and exact pathophysiologic role of GBS colonization of the mammary duct system along with its transmission via breast milk feeding.

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