A Pink Milk Bottle Mystery

The mother of a 5-week-old term infant brings the infant to the pediatrician's office, along with a plastic milk bottle used to feed the baby. The bottle has splotches of red-pink material on the inside (Fig. 1). She wants to know what is happening to the milk in the bottle. The mother has been breastfeeding and has recently been transitioning to pumped breast milk in preparation for returning to work. For the past few days, she has noticed that the breast milk remaining in bottles that were left in a sink overnight after evening feedings turns bright pink by morning. The breast milk has been normal in color during pumping, in the pump tubing, and in bottles stored in the refrigerator before feedings.

The mother denies taking any medications, herbal supplements, or illicit drugs while breastfeeding. The pregnancy was complicated by multiple urinary tract infections, which were treated with nitrofurantoin and trimethoprim-sulfamethoxazole. She denies any signs or symptoms of mastitis.

The infant has been asymptomatic, feeding well, and is gaining weight. Birth was uncomplicated. The infant is afebrile and is well-nourished, well-hydrated, and in no distress. There are no lesions or abnormal findings on the oral mucosa. There is no lymphadenopathy. The remainder of the physical examination appears normal.

The pediatrician sent samples of pink exudate in the bottle as well as stored, expressed breast milk (which had not yet been fed to the infant) for culture. Both cultures yielded the same microbe.

What microbe is the most likely cause of this scenario? When the pediatrician calls to tell you this story and the culture results, what advice do you give? (See next page for discussion.)

Figure 1. Infant bottle with pink material that developed after sitting at room temperature overnight after use for feeding of expressed breast milk.

Journal of the Pediatric Infectious Diseases Society, Vol. 1, No. 4, pp. 347–50, 2012. DOI:10.1093/jpids/pis102

Pediatric ID Consultant

Geoffrey Weinberg and Charles Woods, Section Editors

A Pink Milk Bottle Mystery

Victoria A. Statler,[1] Miranda L. Smith,[2] Brandy B. Fouch,[3] and Charles R. Woods[1]
[1]Pediatric Infectious Diseases, University of Louisville, [2]University of Kentucky College of Medicine, Lexington, and [3]Mt Sterling Pediatrics, Kentucky

Corresponding Author: Victoria A. Statler, Pediatric Infectious Diseases, 571 S Floyd St, Ste 321, Louisville, KY 40202. E-mail: vastat01@exchange.louisville.edu.

Received October 1, 2012; accepted October 5, 2012; electronically published Month 00, 0000.

Microbial answer to "A Pink Milk Bottle Mystery"

Specimens from the pink milk bottle and stored breast milk grew *Serratia marcescens*. The strain was susceptible to amikacin, cefepime, cefoxitin, ceftazidime, ceftriaxone, ciprofloxacin, gentamicin, imipenem, levofloxacin, tobramycin, and trimethoprim-sulfamethoxazole.

The pediatrician was hopeful the mother could continue to breastfeed. Is this reasonable? Should the mother, baby, or both be given antimicrobial therapy?

Serratia marcescens, the microbial culprit in this case, is widespread in the environment, especially in areas that remain moist. *S marcescens* is often the etiology behind pinkish slime or discoloration that is sometimes seen in showers, tubs, toilets, and even soap dishes [1–4]. The microbial metabolite responsible for red or pink color production is prodigiosin, which is a member of the prodiginine family of linear and cyclic tripyrrolic compounds [5].

Serratia marcescens commonly colonizes the respiratory and gastrointestinal tracts in infants [1, 6, 7] and the respiratory tracts in adults. In the early 1990s, *S marcescens* was thought to be nonpathogenic, but the microbe is now well recognized as a cause of infection, almost exclusively as a nosocomial pathogen. It is seen most commonly in intensive care settings, and at least 34 outbreaks have occurred in neonatal or pediatric intensive care units (ICUs) [8].

Contaminated expressed breast milk and various water supplies have been implicated as primary or secondary reservoirs of infection [6–10]. Contamination of an inhalation solution was the source of 1 neonatal ICU (NICU) outbreak of infection [2]. Recurrent outbreaks in the same NICU due to different clones of *S marcescens,* consistent with the ubiquitous environmental presence of this microbe, have been described [11].

Infections tend to occur mostly in compromised patients, but even in outbreak situations, the colonization rate is high compared with the infection rate [6, 12]. Common sites of invasive disease, when it occurs, include meningitis, pneumonia, bacteremia, urinary tract infection, soft tissue infection, and gastrointestinal disease. *Serratia marcescens* also causes central venous and urinary catheter-related infections, and it is a common contaminant of contact lens solutions and cause of contact lens-associated ocular infections [13]. *Serratia* is catalase-positive, and infection with this organism may be a sentinel event in patients with chronic granulomatous disease (CGD). In a study of 368 patients on a national registry of patients with CGD, *Serratia* was the most common cause of osteomyelitis [14].

"Red" pigment production by *S marcescens* during contamination of various foods in antiquity led to beliefs in miraculous appearances of blood and other superstitions. The microbial nature of such events was first recognized by Bartomoleo Bizio in 1819 when he correctly attributed a "strange reddening of polenta (cornmeal mush)" to a living organism [15]. Bizio named the substance *Serratia* after Italian physicist Serafino Serrati, who had developed a type of steamboat.

Before recognition of *S marcescens* as a potential pathogen, this pigment-producing property was considered to be a useful biomarker. In 1950, the US government, in an attempt to study wind currents in the event of biological warfare, aerosolized *S marcescens* over the Pacific Ocean and evaluated inland areas for red pigment to see where wind carried the organism [16]. Inoculation of the microbe also has to evaluate hand-to-hand transmission of bacteria and development of bacteremia after dental extraction.

When grown on solid agar, *S marcescens* colonies may appear to be white, gray, red, or pink [17]. Prodigiosin production is less common among clinical strains of *S marcescens* than among environmental strains (9% vs 61% in 1 study), and it varies among serotypes as well [18]. Prodigiosin production by *S marcescens* occurs

primarily at room temperature, it is inhibited at 37°C and below 20°C, and it is maximal when the microbe is cultured in darkness [19]. (These factors may be relevant to the case described, in which growth was observed when bottles sat overnight at room temperature.)

Prodigiosin and related compounds have various antimalarial, antifungal, and/or antibacterial activities [5]. Prodigiosins also have cytotoxic and apoptotic effects against a wide range of human cancer cell types that are selective, sparing normal cells. T lymphocyte proliferation also can be inhibited by these compounds at concentrations below cytotoxic levels [20]. *Serratia marcescens* is the primary species used for bioproduction of prodigiosins [19]. Other *Serratia* species, actinomycetes (eg, *Streptomyces coelicolor*), and various marine bacteria such as *Pseudoalteromonas* species, *Vibrio psychroerythreus*, and *Hahella chejuensis* (a distant relative of pseudomonads) sometimes produce prodigiosin. Other bacterial species can produce closely related compounds [5, 19].

Serratia isolation from expressed breast milk has been described in several reports, and most have involved NICUs. In an outbreak in 2 NICUs that involved 17 infants with 2 deaths, the outbreak strain was isolated from expressed breast milk from a mother whose infant was known to be colonized. Breast pumps were shared on the units and disinfection practices were not adequate [10]. A similar outbreak involved 30 infants with no deaths. Rectal carriage among these infants was common and often prolonged [21].

Serratia has also been isolated from breast milk pumping units. In 1 such report, a mother presented to her obstetrician stating that her breast milk-pump tubing had turned bright pink. The expressed breast milk was being fed to her twin infants. The mother was well and had no signs of mastitis, but she was treated with antibiotics and subsequently stopped breastfeeding; the infants remained well [22]. One full-term neonate who developed *S marcescens* sepsis on day of life 17, after being fed expressed breast milk contaminated with the microbe, has been reported. The onset of this infection occurred 4 days after hospitalization for another illness [23].

Expressed breast milk seldom will be sterile. A recent study in Thailand demonstrated contamination of 44 of 51 (86%) samples of breast milk expressed using breast pumps [24]. In premature infants, contamination of expressed breast milk with >1000 gram-negative bacilli/mL is associated with feeding intolerance. Higher levels of contamination (>1 000 000/mL) were associated with sepsis among premature infants [25]. However, such high density of contamination is unlikely with refrigeration soon after pumping and use for feeding soon after warming.

Pigment production during infection is rarely evident. This likely is due to inhibition of production at temperatures normal for human hosts. In 1 case report, a term infant with birth asphyxia who developed nosocomial sepsis due to *S marcescens* was linked to a pink hypopyon. Culture of fluid from the anterior chamber of the eye grew *Serratia* [26].

Nosocomially acquired strains of *S marcescens* may be multiply drug-resistant. This species also commonly carries chromosomally encoded, inducible AmpC β-lactamases that may lead to development of resistance during therapy with broad-spectrum cephalosporins. This potential should be considered when selecting antimicrobial regimens for treatment of *S marcescens* infections [3, 27, 28]. Options include (1) concurrent use of aminoglycosides when penicillin or cephalosporin agents that exhibit in vitro susceptibility are administered, or (2) use of carbapenems, quinolones, or other agents that may be appropriate for the site of infection.

Advice Given: Based on the above knowledge of *S. marcescens* epidemiology and virulence, it was agreed that the mother could continue to breastfeed and to pump. No antibiotics were recommended for the mother or the baby, as the risk of developing disease from *Serratia* was believed to be extremely low, as well as less than the risks of adverse effects from any antimicrobial agent that might be prescribed. If the mother had had mastitis, it would have been appropriate to prescribe antimicrobial therapy for her and to continue breastfeeding.

The most likely explanation for this scenario is contamination of the pump from a water source or sink in the home, not the mother's skin. The mother's receipt of trimethoprim-sulfamethoxazole prenatally combined with the in vitro susceptibility of the *Serratia* strain in this case is consistent with, though does not prove, acquisition of the strain from an environmental source rather than the mother. No specific recommendations for decontamination measures of the breast pump or bottles beyond routine household washing was made, as it was unclear if any additional measures would be feasible, more effective or preventive of re-introduction of *Serratia* from the environment.

Outcome: The mother continued to pump and breast feed for two more weeks. The infant remained well throughout this time and continued to thrive months later.

Acknowledgments

Potential conflict of interest. All authors: No reported conflicts.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References

1. Newport MT, John JF, Michel YM, Levkoff AH. Endemic *Serratia marcescens* infection in a neonatal intensive care nursery associated with gastrointestinal colonization. Pediatr Infect Dis **1985**;4:160–67.
2. Maragakis LL, Winkler A, Tucker MG, et al. Outbreak of multidrug-resistant *Serratia marcescens* infection in a neonatal intensive care unit. Infect Control Hosp Epidemiol **2008**;29: 418–23.
3. David MD, Weller TM, Lambert , Fraise AP. An outbreak of *Serratia marcescens* on the neonatal unit: a tale of two clones. J Hosp Infect **2006**;63:27–33.
4. Archibald L, Corl A, Aguero S, et al. Amplification of *Serratia marcescens* infection by soap in neonatal intensive care patients. Am J Infect Control **1996**;34:87.
5. Williamson NR, Fineran PC, Leeper FJ, Salmond GPC. The biosynthesis and regulation of bacterial prodiginines. Nat Rev Microbiol **2006**;4:887–99.
6. Christensen GD, Korones SB, Reed L, et al. Epidemic *Serratia marcescens* in a neonatal intensive care unit: importance of the gastrointestinal tract as a reservoir. Infect Control **1982**;3: 127–33.
7. Albers MJ, Mouton JW, Tibboel D. Colonization and infection by *Serratia* species in a paediatric surgical intensive care unit. J Hosp Infect **2001**;48:7–12.
8. Voelz A, Müller A, Gillen J, et al. Outbreaks of *Serratia marcescens* in neonatal and pediatric intensive care units: clinical aspects, risk factors, and management. Int J Hyg Environ Health **2010**;213:79–87.
9. Berthelot P, Grattard F, Amerger C, et al. Investigation of a nosocomial outbreak due to *Serratia marcescens* in a maternity hospital. Infect Control Hosp Epidemiol **1999**;20:233–6.
10. Jones BL, Gorman LJ, Simpson J, et al. An outbreak of *Serratia marcescens* in two neonatal intensive care units. J Hosp Infect **2000**;46:314–19.
11. Maltezou HC, Tryfinopoulou K, Katerelos P, et al. Consecutive *Serratia marcescens* multiclone outbreaks in a neonatal intensive care unit. Am J Infect Control **2012**;40:637–42.
12. Friedman ND, Kotsanas D, Brett J, Billah B, Korman TM. Investigation of an outbreak of *Serratia marcescens* in a neonatal unit via a case-control study and molecular typing. Am J Infect Contro. **2008**;36:22–8.
13. Parment PA. The role of *Serratia marcescens* in soft contact lens associated ocular infections. A review. Acta Ophthalmol Scand **1997**;75:67–71.
14. Winkelstein JA, Marion MC, Johnston RB Jr, et al. Chronic granulomatous disease. Report on a national registry of 368 patients. Medicine (Baltimore) **2000**;79:155–69.
15. Merlino CP. Bartolomeo Bizio's letter to the most eminent priest, Angelo Bellani, concerning the phenomenon of the red colored polenta. J Bacteriol **1924**;9:527–43.
16. Yu VL. *Serratia marcescens*: historial perspective and clinical review. N Engl J Med **1979**;300:887–93.
17. Goluszko P, Nowicki B, Goluszko E, et al. Association of colony variation in *Serratia marcescens* with the differential expression of protease and type 1 fimbriae. FEMS Microbiol Lett **1995**;133:41–5.
18. Aucken HM, Pitt TL. Antibiotic resistance and putative virulence factors of *Serratia marcescens* with respect to O and K serotypes. J Med Microbiol **1998**;47:1105–13.
19. Chang CC, Chen WC, Ho TF, et al. Development of natural anti-tumor drugs by microorganisms. J Biosci Bioeng **2011**;11: 501–11.
20. Han SB, Kim HM, Lee CW, et al. T-cell specific immunosuppression by prodigiosin isolated from *Serratia marcescens*. Int J Immunopharmacol **1998**;20:1–13.
21. Gransden WR, Webster M, French GL, Phillips I. An outbreak of *Serratia marcescens* transmitted by contaminated breast pumps in a special care baby unit. J Hosp Infect **1986**;7:149–54.
22. Faro J, Katz A, Berens P, Ross PJ. Premature termination of nursing secondary to *Serratia marcescens* breast pump contamination. Obstet Gynecol **2011**;117:485–6.
23. Youssef RF, Darcy E, Barone A, et al. Expressed breast milk as a source of neonatal sepsis. Pediatr Infect Dis J **2002**;21:888–9.
24. Boo NY, Nordiah AJ, Alfizah H, et al. Contamination of breast milk obtained by manual expression and breast pumps in mothers of very low birthweight infants. J Hosp Inf **2001**;49: 274–81.
25. Botsford KB, Weinstein RA, Boyer KM, et al. Gram-negative bacilli in human milk feedings: quantitation and clinical consequences for premature infants. J Pediatr **1986**;109:707–10.
26. al Hazzaa SA, Tabbara KF, Gammon JA. Pink hypopyon: a sign of *Serratia marcescens* endophthalmitis. Br J Ophthalmol **1992**;76:764–5.
27. Baggatini M, Crispino M, Gentile F, et al. A nosocomial outbreak of *Serratia marcescens* producing inducible Amp C-type beta lactamase enzyme and carrying antimicrobial resistance genes within a class 1 integron. J Hosp Infect **2004**;56:29–36.
28. Choi SH, Lee JE, Park SJ, et al. Emergence of antibiotic resistance during therapy for infections caused by *Enterobacteriaceae* producing AmpC β-lactamase: implications for antibiotic use. Antimicrob Agents Chemother **2008**;52:995–1000.