A rare case of infant sepsis due to the \textit{emm}-89 genotype of Group A \textit{Streptococcus} within a community-acquired cluster

Salvatore Pignanelli$^1$, Sandra Brusa$^2$, Giovanna Pulcrano$^1$, Maria Rosaria Catania$^3$, Enrico Cocchi$^2$, Marcello Lanari$^2$

$^1$Department of Diagnostic Services, O.U. Laboratory Analysis of Clinical Chemistry and Microbiology, S. Maria della Scaletta Hospital, (BO), Italy; $^2$Department of Maternal and Child Health, Pediatrics and Neonatology Unit, S. Maria della Scaletta Hospital, Imola (BO), Italy; $^3$Department of Molecular Medicine and Medical Biotechnology, Medicine School, University of Naples Federico II, Naples, Italy

**Corresponding author**
Salvatore Pignanelli MD
Via Guelfa, 30 - 40138 Bologna, Italy
E-mail dott.pignanelli@libero.it

**SUMMARY**

Invasive Group A \textit{Streptococcus} disease is a severe and sometimes life-threatening infection with only few cases reported in literature. We describe the case of a 49-day-old male infant with invasive Group A \textit{Streptococcus} infection characterized by acute otitis media and development of septicemia within a probably community-acquired cluster. The causative agent resulted to be a rare \textit{emm}-89 genotype of \textit{Streptococcus pyogenes}. Group A \textit{Streptococcus} must be considered responsible for sepsis in newborns and young infants.

**KEY WORDS:** Group A \textit{Streptococcus}, \textit{Streptococcus pyogenes}, \textit{emm}-89 genotype, Acute otitis media, Sepsis.

\textit{Streptococcus pyogenes}, known as Group A \textit{Streptococcus} (GAS), is a major bacterial pathogen affecting children globally (Martic \textit{et al.}, 2010). GAS causes a wide array of infections ranging in severity from mild pharyngitis and cellulitis (the most common caused by GAS in children) to severe invasive infections (Paul \textit{et al.}, 2012). GAS was also an important cause of puerperal, obstetric, neonatal morbidity and mortality from the 16th century until the beginning of the antibiotic era. Since then, the frequency of neonatal sepsis caused by GAS has decreased drastically (Paul \textit{et al.}, 2012). Unfortunately, an increasing incidence of invasive GAS (iGAS) infections has been observed in the last three decades (Olafsdottir \textit{et al.}, 2014). In Europe, the overall iGAS disease incidence is currently estimated between 2.5 and 3.1 per 100,000 inhabitants per year, increasing directly proportional to age (Olafsdottir \textit{et al.}, 2014). Interestingly, an inverse relationship between iGAS incidence and age has been proved in children (Filleron \textit{et al.}, 2012). iGAS disease has a high mortality with an overall case fatality rate of 19\% in Europe (Luca-Harari \textit{et al.}, 2009). When focusing on pediatric patients the mortality rate is lower than that observed in adults (4.4\%), but a higher death rate is observed in children under 1 year of age (8\%) than in older children (4\% between 1 and 14 years) (Filleron \textit{et al.}, 2012). Risk factors such as extremes of age, immunosuppression, skin lesions and diabetes mellitus are associated with the development of invasive infections (Lepoutre \textit{et al.}, 2011). Complications like necrotizing fasciitis, toxic shock syndrome, pneumonia and bacteremia in the absence of an identified focus are associated with a high risk of fatal outcome (Lepoutre \textit{et al.}, 2011). Currently, GAS is an infrequent cause of severe infection in the neonatal period and early infancy and the evidence of these cases is sporadic (No-
Nevertheless, GAS has been reported to cause neonatal sepsis, meningitis, skin infections and toxic shock syndrome (Lepoutre et al., 2011). We report the clinical history of a previously healthy full term infant with acute otitis media and documented GAS sepsis. A 49-day-old male infant was admitted to our Pediatric Unit because of fever (38.9°C axillary) and poor feeding in the past few days.

Ten days before, he had presented a similar episode of fever lasting one day, without other symptoms and with a reported negative objectivity. Urinalysis done at another hospital proved negative and the baby was treated only with antipyretics. History-taking failed to disclose perinatal and neonatal infection risk factors and the mother’s vagino-rectal swab for GBS screening at pregnancy week 37 was negative.

At physical examination, the baby showed a tachycardia of 200 beats per minute, no murmurs, present and regular peripheral arterial pulses, 99% O₂ saturation, stable breathing and circulation.

A red tympanic membrane on the right was revealed by otoscopic examination. A diffuse bright-red erythema with pronounced edema and induration on the ipsilateral angle of the jaw was recorded. On palpation of this area, the skin was warm and tender, but not fluctuant, and the baby complained of pain.

Other physical signs were normal, there were no signs of meningeal irritation, no dermographism, the anterior fontanelle was normoten-sive and non-pulsating and crying was valid.

A red tympanic membrane appeared matt and retracted 24 hours after admission, but returned to normal in the next 48 hours. After 48 hours of antibiotic treatment, the infant became afebrile and the jaw erythema, edema and pain were markedly reduced. The blood and ear swabs culture revealed GAS, sensitive to ceftriaxone.

Monotherapy was considered adequate and ceftriaxone was continued alone, until the infant’s discharge. The inflammatory markers became normal 5 days after admission and the infant was discharged from hospital two days later. Oral amoxicillin treatment for 7 days was prescribed on the basis of antimicrobial susceptibility test results. No signs of residual disease were present at the clinical follow-up 15 days later. An audiology follow-up was normal two months after recovery.

Since his mother and sister had presented a sore throat episode with normal temperature two weeks before the baby’s admission, pharyngeal cultures were taken and GAS was isolated from both. Streptozyme test (ELITech MICRO-BIO, Signes, France) performed on blood samples from the baby and from his mother and sister revealed titres of 1:400 and 1:800 respectively (normal values <1:200). In order to understand whether the mother and/or sister were the source of the child infection, the GAS isolates were investigated by Pulsed-Field Gel Electrophoresis (PFGE). Small colonies, exhibiting a beta-hemolysis, were isolated from Sheep Blood agar plates (Becton Dickinson Erembodegem-Aalst, Belgium) after incubation in ambient air at 35°C±1°C for 24 hours. The isolates were identified as S. pyogenes strains using bacitracin-test (Bio-Rad, Milan, Italy), catalase-test, Gram-staining, latex-agglutination (BioMérieux, Marcy l’Etoile, Craponne, France) and Vitek-2 automated biochemical system (BioMérieux) as described previously (Pignanelli et al., 2013).

Minimum inhibitory concentration (MIC) values were obtained by epsilon-test (BioMérieux), according to the 2014 European Committee on Antimicrobial Susceptibility Testing standards. Streptococcal isolates displayed identical antimicrobial profile, with sensitivity against all antimicrobial agents tested, except trimethoprim/sulphamethoxazole (SXT). MIC values (μg/mL) of antimicrobials were as follows: 0.06 for amox-
A rare case of infant sepsis due to the emm-89 genotype of Group A Streptococcus within a community-acquired cluster

Amplification of emm genes from streptococcal genomic DNA samples was performed using the primers 1 and 2 recommended by the Centers for Disease Control and Prevention (Facklam et al., 1999). Amplification products were analyzed on 1% agarose gel electrophoresis, sequenced at the Cinge-Biotecnologie Avanzate s.c.a.r.l. (Naples, Italy) and the sequences obtained were analyzed at the National Centre for Biotechnology Information via the blast program (http://www.ncbi.nlm.nih.gov/blast). PFGE analysis and emm-typing revealed that the strains were identical and belonged to emm-89 type (Figure 1).

A change in the epidemiology of severe GAS infection, with an increased incidence and severity of the disease has been reported since the mid 1980s (Paul et al., 2012). The reasons for such change have yet to be fully elucidated. This case represents an invasive disease and development of sepsis in an infant with acute otitis media (AOM) within a community-acquired cluster most likely from a benign throat infection of the mother (or sister). The causative agent proved to be a GAS belonging to the emm-89 type. M protein, the major GAS virulence factor, is a surface constituent sustaining the bacterial resistance to phagocytosis and evoking a type-specific host immune response (Olafsdottir et al., 2014). M protein is widely used as marker for GAS type isolates (Olafsdottir et al., 2014). Many works have reported correlations between clinical manifestations, age distribution and emm GAS genotype. In Europe the most prevalent emm types identified in children are the emm-1, emm-3, emm-4, emm-12 and emm-28 (Olafsdottir et al., 2014). emm-1, emm-3 and emm-28 resulted responsible for invasive infections (Luca-Harari et al., 2009; Meisal et al., 2010) and the highest case fatality rate appeared related to emm-1 and emm-3 (Olafsdottir et al., 2014).

Although the distribution of the GAS strains is not stable over time and varies according to the geographical area, it is noteworthy that our GAS strains belonged to emm-89 type. This genotype is known to be more frequently associated with arthritis, skin and soft tissue infections (Lepoutre et al., 2011; Luca-Harari et al., 2009; Olafsdottir et al., 2014), puerperal sepsis (Filleron et al., 2012; Meisal et al., 2010) and rarely rheumatic fever (Martin et al., 2006).

It is also more common in adults than children (Luca-Harari et al., 2009; Olafsdottir et al., 2014). Moreover, as in our case, emm-89 is predominant in the pharynx compared with other emm types (Shulman et al., 2009) and has been reported described as cause of clusters or outbreaks (Asteberg et al., 2006; Lepoutre et al., 2011; Liu et al., 2014; Luca-Harari et al., 2009). The focus of infection reported in our case is un-
usual (Olafsdottir et al., 2014) and, in comparison with other microorganisms causing AOM such as *Haemophilus influenzae*, *Streptococcus pneumoniae* and *Moraxella catharralis*, GAS is less common and occurs most frequently in children ≥24 months of age (Nir-Paz et al., 2010). Our case demonstrates that a trivial infection of the upper airways in household contacts (especially mothers) could become a potential source of invasive infection for a newborn or an infant. Environmental hygienic measures and timely treatment of similar infections can prevent the transmission to the baby.

After transmission, early diagnosis and subsequent prompt appropriate therapy (Olafsdottir et al., 2013; Paul et al., 2012), as in our case, can avoid life-threatening complications and possible sequelae. A quick swab for GAS on the external ear secretion, if present, would have allowed to avoid life-threatening complications and possible sequelae. A quick swab for GAS on the external ear secretion, if present, would have allowed to avoid life-threatening complications and possible sequelae. A quick swab for GAS on the external ear secretion, if present, would have allowed to avoid life-threatening complications and possible sequelae.

In conclusion, there are few reports of iGAS disease in the first months of life (Nohara et al., 2013), nevertheless our case and recent reports from the literature (Busetti et al., 2013) show that GAS must be considered responsible for sepsis in newborns and young infants.

REFERENCES


