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The first case of fatal pneumonia caused by Panton–Valentine leukocidin-producing *Staphylococcus aureus* in an infant in the Czech Republic

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Abstract Panton–Valentine leukocidin-producing strains of *Staphylococcus aureus* can cause severe skin and soft tissue infections and necrotizing pneumonia with a high mortality rate. This is a report on the first case of fatal pneumonia with mediastinitis in an infant in the Czech Republic. The causative agent was a methicillin-susceptible *S. aureus* strain with pronounced production of the PVL toxin and hyperproduction of enterotoxin A.

Abbreviations

PVL	Panton–Valentine leukocidin
MRSA	methicillin-resistant *Staphylococcus aureus*
MSSA	methicillin-susceptible *Staphylococcus aureus*
ENT	ear, nose, and throat investigations
NRL	National Reference Laboratory
RPLA	reversed passive latex agglutination
ICU	intensive care unit
CT	computed tomography

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Introduction

Panton–Valentine leukocidin (PVL) is a necrotizing exotoxin produced by both methicillin-susceptible and methicillin-resistant *Staphylococcus aureus* (MRSA) strains. PVL is a pore-forming toxin that causes damage to the membrane of leukocytes, leading to the destruction of white blood cells and tissue necrosis. Consequently, the immune response is altered, and the patient develops a highly virulent infection with a high mortality rate of up to 80 % (Kefala-Agoropoulou et al. 2010). The infection affects mostly younger patients. It is typically manifested by skin and soft tissue lesions (Daskalaki et al. 2010; Masiuk et al. 2010; Fontanilla et al. 2010) and often by fatal necrotizing pneumonia (Hidron et al. 2009; Kreienbuehl et al. 2011). In addition, PVL can be implicated in a number of other conditions such as bacteremia, sepsis, osteomyelitis, arthritis (Hall et al. 2010), myositis (Lehman et al. 2010), intravascular thrombosis (Kramkimel et al. 2009), brain abscess (Ramos et al. 2009), infectious endocarditis (Tsai et al. 2008), or Lemierre syndrome-imitating disease (Shivashankar et al. 2008). PVL-producing strains of *S. aureus* are found globally, and necrotizing skin lesions have repeatedly been reported in travellers returning from the tropics (Schleucher et al. 2008).

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According to literature data, PVL is produced by 2 % of *S. aureus* strains. In the Czech Republic, the PVL toxin-producing strains are monitored by the National Reference Laboratory (NRL) for Staphylococci. The first detection of PVL in *S. aureus* in the Czech Republic was reported in 2004. In the past 8 years (2004–2011), a total of 5,584 *S. aureus* strains were investigated, and 318 (5.7 %) of them were PVL producers. Of the PVL producers, 244 were methicillin-susceptible *S. aureus* (MSSA), and 74 were community MRSA strains. The higher rate of PVL producers detected by the NRL for Staphylococci in comparison with literature data can be explained by the fact that the staphylococcal strains referred to the NRL by field laboratories were collected selectively from patients with uncommon conditions.

Most PVL-producing strains were from patients with skin diseases; only eight of them were from those with necrotizing pneumonia (seven adults and one child). Three of these patients survived, and five died (a case fatality rate of 62.5 %). The first case of fatal abscessing pneumonia in the Czech Republic was reported in 2008 (Petras et al. 2008). The infection had a rapid fatal outcome: a 22-year-old male died within 6 days of the onset of symptoms, i.e. 2 days after admission to the hospital. Septic shock in fatal pneumonia caused by PVL-positive *S. aureus* in a 29-year-old man was reported by Benes et al. (2010). Despite intensive care, the patient died 8 h after admission for severe septic shock and incipient pneumonia.

This paper is a case report of a 10-month-old boy with infection caused by a PVL-producing strain of *S. aureus*. It is the first known fatal case in a child in the Czech Republic. The child was treated successively in three clinical departments (Infectious Diseases, Paediatrics, and Anaesthesiology and Resuscitation) of three Prague teaching hospitals.

Methods and the patient

Origin of strains

A total of 5,584 *S. aureus* strains were screened for carriage of the *lukS*-PV and *lukF*-PV genes. The strains were referred to the NRL for Staphylococci, NIPH, Prague, by hospital microbiology departments of 14 regions of the Czech Republic.

Phenotyping methods

Production of *S. aureus* enterotoxins (A, B, C, D), exfoliative toxins (A, B), and toxic-shock syndrome toxin 1 (TSST-1) was assayed by the RPLA method (Denka Seiken kits: SET-RPLA, EXT-RPLA, TST-RPLA). Susceptibility to 16 antibiotics (ciprofloxacin, clindamycin, trimethoprim/sulfamethoxazole, erythromycin, fusidic acid, gentamicin, linezolid, oxacillin, rifampicin, tigecyclin, vancomycin, cefoxitin, mupirocin, tetracycline, tobramycin, and chloramphenicol) was tested by the broth microdilution method or disc diffusion method (Oxoid discs, Muller-Hinton agar) and interpreted according to the EUCAST criteria (EUCAST 2012).

Genotyping methods

Carriage of the *lukS*-PV and *lukF*-PV genes for PVL was tested with the PCR primers described previously (Lina et al. 1999). Spa typing was performed according to Shopsin et al. (1999) using the Ridom SpaServer database. Multilocus sequence typing was performed according to Enright et al. (2000).

Patient

The patient was a 10-month-old so-far healthy boy with an insignificant epidemiological history.

Case report and discussion

Case report

At Christmas 2008, a 10-month-old boy presented to the Department of Infectious Diseases with fever and diarrhoea. His mother suffered from nausea; otherwise, his epidemiological history was insignificant. Two days prior to admission to the hospital, he developed fever around 38 °C. On the day of admission, the boy was agitated, vomited repeatedly, and had one watery stool. After the ENT investigation to rule out otitis media, he was referred to the hospital. On admission, he was conscious, pale, eupneic, had no sign of respiratory infection, hydration within normal limits, clean breathing, adequate cardiopulmonary compensation, no abdominal pain on palpation, and no meningeal signs. Laboratory tests revealed increased inflammatory parameters, i.e. a white blood cell count of 11,700–14,800 $\times 10^9$/L with a shift to the left and C-reactive protein (CRP) of 211–245 mg/L. The patient had haemoglobin values of 96–93 g/L, haematocrit of 0.28, slightly increased urea at 8.4 mmol/L, creatinine and minerals repeatedly within the normal range, and protein and erythrocytes detected in the urine. The stool culture revealed *Citrobacter youngae*, while latex agglutination test for rotaviruses and adenoviruses remained negative. Tonsillar swab culture was negative, and results of ultrasonography of the abdomen and abdominal examination by a paediatric surgeon were normal. Samples were collected for blood and urine culture.

During the first 2 days of hospital stay at the Department of Infectious Diseases, the boy had fever with a peak of up to 38.2 °C on both days, but there were no vomiting, no diarrhoea, and no signs of respiratory infection. Based on urinalysis results, high inflammatory parameters, and suspected urine infection, the patient was treated with cotrimoxazole (no urinary tract infection was detected by urine culture). On the third post-admission day, the patient got worse, became apathetic and developed dyspnoea, tachypnoea, and tachycardia; oxygen saturation was 89 %, and on auscultation, breathing sounds were reduced on the left side. X-ray investigation of the lungs showed a homogenous opacity in the left hemithorax due to infiltrate in combination with exudate in the pleural cavity, and hypoventilation changes were also observed in the right upper pulmonary lobe (Fig. 1a). Intravenous cefotaxime was started, and the patient was transferred to the ICU of the Department of Paediatrics.

On admission to the ICU, the boy was pale, negativistic, with tachypnoea (68 breaths per min), tachycardia (155 beats per min), superficial breathing with intermittent grunting, a significant auscultatory finding above the left lung wing, and oxygen saturation without oxygen supplementation below 90 %. Laboratory tests revealed persistently elevated inflammatory parameters (white blood cells 10,000–49,800 × 10^9/L, CRP 256.7–177.5 mg/L, procalcitonin 63.3–15.2 ng/mL, and D-dimers more than 4,000). At the beginning of his 6-day hospital stay at the Department of Paediatrics, the X-ray investigation revealed alar left-sided pleuropneumonia and minor basal lung inflammatory infiltration on the right side. A control X-ray showed partial aeration of the left hemithorax, left-sided empyema, and also a progression of the inflammatory infiltration in the right upper and middle areas (Fig. 1b).

The CT scan of the thorax showed an extensive exudation in the left pleural cavity with increased density of the left lung lobes, right basal pneumonia, and exudates in the right paramediastinal area and upper anterior mediastinum—v.s. mediastinitis. Puncture of the thorax was performed, and similar to the blood culture done previously at the Department of Infectious Diseases, the puncture specimen culture revealed *S. aureus* susceptible to oxacillin, vancomycin, clindamycin, and lincomycin. Due to respiratory insufficiency, mechanical ventilation and intravenous noradrenaline to support blood pressure were needed, and cefotaxime, clindamycin, gentamicin, fluconazole, and corticosteroids were given. After a temporary 70-h clinical and laboratory improvement, the patient got worse, developing an extensive left-sided empyema with fluid reaccumulation after repeated thoracocentesis. After 6 days, the patient with bilateral pneumonia, left-sided empyema, suspected mediastinitis, and respiratory insufficiency was transferred to the Department of Anaesthesiology and Resuscitation for possible surgical intervention.

During his 4-day stay at the Department of Anaesthesiology and Resuscitation, mechanical ventilation and noradrenaline to support blood pressure were continued. Based on susceptibility testing results, the patient was switched to oxacillin and clindamycin in combination with gentamicin. During the drainage of the left hemithorax, 50 mL of thick creamy pus was discharged. On the second day, effusion appeared in the neck and upper chest soft tissues.

Neck ultrasonography revealed a left-sided fluid collection starting from the mediastinum, along the sternocleidomastoid muscle to the cranial base. The puncture yielded 20 mL of pus. *S. aureus* was detected by culture again. Despite a decrease in inflammatory parameters, ventilation parameters declined after 4 days of stay at the third consecutive clinical department, with progression of barotraumas, bilateral pneumothorax, pneumoperitoneum (Fig. 1c), and circulatory failure leading to death.

Causative agent

The causative agent was a methicillin-susceptible *S. aureus* strain with pronounced production of the PVL toxin and hyperproduction of enterotoxin A, classified into *spa* type t443 and ST-30 and phage-typeable into phage group I. The strain was susceptible to 16 antibiotics tested.

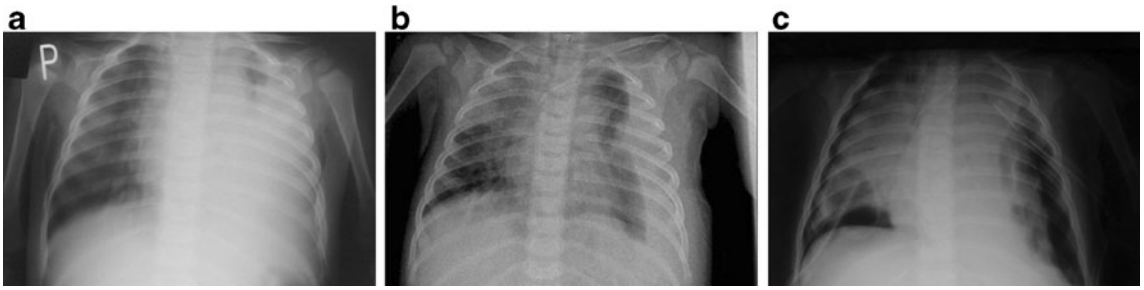

Fig. 1 X-ray investigation of the lungs: **a** Bulovka Hospital, Prague; **b** Thomayer Hospital, Prague; **c** Motol University Hospital, Prague

Discussion

Here, we report the first and, so far, only known case of severe necrotizing pneumonia complicated with mediastinitis in a child in the Czech Republic. The onset of the disease was clearly atypical: the child was free of any respiratory signs but had gastrointestinal symptoms. Their presence can be explained by the hyperproduction of enterotoxin A induced by the PVL-positive *S. aureus* strains. Based on high inflammatory parameters and findings in the urine, infection of the urinary tract was first suspected. After the diagnosis was made, treatment with antibiotics was started and soon adjusted according to culture and susceptibility testing results. Although the strain was susceptible to the antistaphylococcal antibiotics used, only a temporary improvement was achieved followed by the development of uncontrollable empyema and mediastinitis. In severe infections caused by the PVL-producing strains of *S. aureus*, there is an urgent need for an early diagnosis and a targeted, uncompromising antibiotic therapy combined, if necessary, with a surgical intervention.

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