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Case Report

Severe Sepsis Secondary to Persistent *Lysinibacillus sphaericus*, *Lysinibacillus fusiformis* and *Paenibacillus amylolyticus* Bacteremia

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ABSTRACT

Lysinibacillus and *Paenibacillus* are pervasive bacteria rarely associated with human disease. Less sophisticated microbiology techniques may frequently incorrectly identify these genera as *Bacillus spp.*, often regarded as environmental contamination. This report describes a case of severe sepsis due to persistent *Lysinibacillus* and *Paenibacillus* bacteremia, identified by matrix-assisted laser desorption and ionization time-of-flight mass spectroscopy and 16S rRNA gene sequencing.

1. Introduction

Lysinibacillus and Paenibacillus are ubiquitous Gram-positive bacteria initially designated as *Bacillus spp.* and often regarded as environmental contaminants when isolated in the clinical microbiology laboratory, although their potential to cause human disease has been documented.[1,2] The introduction of reliable identification techniques such as matrix-assisted laser desorption and ionization time-of-flight mass spectroscopy and 16S rRNA gene sequencing allows for accurate delineation of potentially pathogenic bacteria. We report a case of severe sepsis caused by persistent *Lysinibacillus* and *Paenibacillus* bacteremia in a patient with a history of intravenous drug abuse and splenectomy. Our report highlights the critical importance of accurate laboratory techniques in the diagnosis of infectious syndromes due to uncommonly isolated microorganisms whose pathogenicity can be ambiguous.

2. Case report

A 36 year old woman with a history of splenectomy secondary to a motor vehicle accident and intravenous heroin abuse presented to the emergency department at The Ohio State University Wexner Medical Center (OSUWMC) with a 10 day history of fever, chills, emesis, myalgia, rhinorrhea and coughing. One week prior, she was seen at an outside hospital for worsening general malaise and was prescribed a course of oral ciprofloxacin for a presumed urinary tract infection and discharged. Subsequently, her condition continued to deteriorate and she was unable to eat, drink, urinate or have bowel movements and was experiencing new left lower quadrant pain. The patient reported that, after her discharge, the outside hospital called to inform her that her blood cultures were positive and instructed her to begin a new antibiotic regimen, although it is unclear whether she took this antibiotic. On admission to OSUWMC, she was profoundly dehydrated, hypotensive, hyperkalemic, metabolically acidotic, and was in acute renal failure. Pertinent laboratory results on admission included a carbon dioxide level of 16 mmol/L, blood urea nitrogen (BUN) 132 mg/dL, serum creatnine 8.73 mg/dL, anion gap 32, total bilirubin 1.5 mg/dL, alkaline phosphatase 162 U/L, alanine aminotransferase 121 U/L, aspartate aminotransferase 46 U/L, white blood cell count of 62,700 cells/μL with 91% segmented neutrophils, and a hemoglobin of 11.3 g/dL. A chest x-ray revealed no acute cardiopulmonary process and a renal

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ultrasound was unremarkable. A computed tomography scan of the abdomen and ultrasound of the right upper quadrant revealed significant thickening of the gallbladder wall consistent with acute cholecystitis although no surgical intervention was required. A hepatobiliary iminodiacetic acid scan was recommended but the patient refused due to lack of insurance coverage. On hospital day three, two peripheral blood cultures collected during admission signaled positive for Gram variable bacilli (Figure 1) using the BD BACTEC 9240 system (Becton-Dickinson, Sparks, MD) with charcoal-free culture bottles and the patient was started on intravenous vancomycin and ertapenem. The Infectious Diseases (ID) service was consulted and she was transferred to the intensive care unit (ICU) for hypotension, but responded to a fluid bolus and never required vasopressor support. On day four of hospitalization, her antibiotics were switched to ampicillin/sulbactam by the ID physician due to concern for listeriosis as she remained hypotensive and the Gram stain from her blood cultures was now being reported as Gram positive bacilli. On day five, peripheral blood cultures grew two colony types, the larger and grayer colony was identified as *Lysinibacillus fusiformis* by matrix-assisted laser desorption and ionization time-of-flight mass spectroscopy (MALDI-TOF MS) using the Bruker Microflex instrument, Biotyper software version 3.0, and database version 3.1.0 (Bruker Daltonik, Bremen, Germany) and the second smaller and whiter as *Lysinibacillus sphaericus* by 16S rDNA sequencing as MALDI-TOF MS did not generate a reliable identification. Antibiotic susceptibilities were performed via Etest (bioMérieux, Durham, NC) and minimum inhibitory concentrations and interpretations are listed in Table 1. The identification of this polymicrobial *Bacillus*-like bacterium prompted a more targeted physical exam which revealed that the patient had very poor dentition. A Panorex exposed a focal lucency around tooth 15 that was consistent with abscess formation. On day six, the patient was transferred out of the ICU as her hypotension and acute renal failure had resolved. Her peripheral blood cultures were again positive for *Lysinibacillus spp.* along with *Paenibacillus amylolyticus* identified using MALDI-TOF MS and 16s rRNA sequencing. The patient then decided to leave against medical advice with no antibiotic therapy.

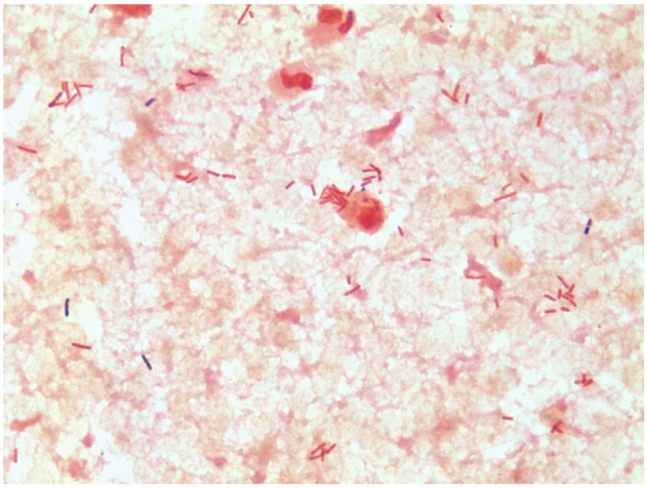

Figure 1. Gram variable rod (1000x).

Table 1
L. fusiformis and *L. sphaericus* antibiotic susceptibilities

Antibiotic	*L. fusiformis*		*L. sphaericus*	
	MIC	Interpretation	MIC	Interpretation
Ceftriaxone	4	S	-	-
Linezolid	0.75	U	1	U
Moxifloxacin	0.38	U	0.125	U
Penicillin	0.12	S	0.125	S
Vancomycin	2	S	1	S

The next day, the ID team was notified by the clinical microbiology laboratory that fungal elements were seen on Gram stain for both sets of blood culture taken on hospital day three. Consequently, the ID team contacted the patient and persuaded her to return to the hospital for additional antimicrobial therapy. Upon readmission, she had no subjective complaints and was treated with ampicillin/sulbactam and caspofungin. Repeat blood cultures were drawn and were again positive for *Paenibacillus amylolyticus* and *Bacillus spp.* The ID team recommended drainage of the dental abscess along with a dilated fundus exam and transesophageal echocardiogram due to presumed candidemia, which were both negative for endopthalmitis and endocarditis, respectively. Fungal elements seen on Gram stain were non-viable on culture and determined only to resemble zygomycetes. Before further diagnostic or therapeutic modalities could be employed, the patient again decided to leave against medical advice. She was given a prescription for amoxicillin/clavulanate with no further follow up available. Her last set of blood cultures from day three of the second hospital admission finalized as no growth after five days of incubation.

3. Discussion

Lysinibacillus spp., are Gram positive or Gram variable, aerobic or facultative anaerobic rod-shaped bacilli that form endospores, tolerate extremes of environment and are ubiquitous.[1] Initially designated *B. sphaericus*, the name was subsequently changed based on the distinctive peptidoglycan composition of its cell wall, along with further phylogenetic and physiological analyses.[3] Organisms within the *Bacillus* genus often show only subtle distinctions from one another on chromosomal DNA sequencing, likely due to a high prevalence of lateral gene transfer. Despite this commonality, there are often pronounced clinical differences between disease states caused by specific species.[1] *L. sphaericus* is fairly large in size and grows easily on culture media at environmental temperatures, but is rarely isolated from human specimens.[1]

Bacillus spp. and related bacteria such as *Lysinibacillus spp.* are commonly regarded as contaminants if isolated in the laboratory primarily due to their ubiquitous nature and perceived lack of pathogenicity. Despite this perception, they have been known to cause serious infections in humans. Bacteremia has been the most common presentation of systemic infections due to *Bacillus* species, particularly associated with the presence of an intravascular or surgically implanted catheter. Disseminated *Bacillus spp.* infections have been reported in neonates, neutropenic patients and injection drug users. The largest case report of *L. sphaericus* bacteremia to our knowledge represented 12 cases over a 10 year period at a children's cancer hospital in Italy.[2] *L. sphaericus* was demonstrated to cause 12 out of 469 (2%) episodes of bacteremia in children with cancer or those undergoing bone marrow transplant. All isolates were susceptible to fluoroquinolones and all 12 patients were treated with ciprofloxacin and survived.

Although specific antibiotic therapy is not indicated for all infections due to *Bacillus* species, removal of prosthetic material and implanted catheters seems to be vital to achieve cure.[3] The majority of *Bacillus* species are susceptible to vancomycin, clindamycin, fluoroquinolones, aminoglycosides and carbapenems while penicillin and cephalosporin susceptibility is variable. Overall, the reported susceptibilities in our case were similar to previous literature primarily focused on the *Bacillus* species[4,5] although both of this patient's *Lysinibacillus* isolates demonstrated susceptibility to penicillin.

Paenibacillus is a spore-forming Gram positive, mesophilic, heterotrophic rod, which was first isolated and identified as a

novel species by the Jet Propulsion Laboratory Spacecraft Assembly Facility in December 2000.[6] *Paenibacillus* spp. are also ubiquitous and found in the environment. Some members of the *Paenibacilli* were originally classified as bacilli but then reclassified as *Paenibacillus* genus. Because of the genetic similarity to *Bacillus* spp., treatment is chosen according to the standard treatment of *Bacillus* spp. (i.e. *B cereus*) in the absence of sensitivity testing. There are more than 100 species identified in the genus *Paenibacillus* although exceedingly few have been found to cause human disease, with *P. alvei* being the most common. In the literature, multiple infectious syndromes associated with *P. alvei* have been described, including pneumonia and endophalmitis.[7–10]

This report represents a rare case of bacteremia due to *Lysinibacillus and Paenibacillus spp.* in an immunocompetent patient without implanted prosthesis or intravascular catheters. To our knowledge, bacteremia due to *Bacillus*-like species has not been associated with poor dentition or odontological disease. Although the undrained dental abscess could have been a potential source of her persistent bacteremia, her history of intravenous drug abuse and splenectomy likely also contributed, despite her claims of abstinence from heroin. Clinically relevant infections with *Lysinibacillus* and *Paenibacillus* species are uncommon but proper management with aggressive source control and timely, susceptibility-guided antibiotic therapy are essential for optimal clinical outcomes.

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References

1. Fekete T. Bacillus Species and Related Genera Other than Bacillus anthracis. In Mandell GL, Bennett JE, Dolin R. Mandell, Douglas and Bennett's Principles and Practice of Infectious Diseases. 7th Edition. 2010; 209: 2727–2731.
2. Castagnola E, Fioredda F, Barretta MA, Pescetto L, Garaventa A, Lanino E, et al. Bacillus sphaericus bacteraemia in children with cancer: case reports and literature review. *J Hosp Infect* 2001;**48**:142–5.
3. Ahmed I, Yokota A, Yamazoe A, Fujiwara T. Proposal of Lysinibacillus boronitolerans gen. nov. sp. nov., and transfer of Bacillus fusiformis to Lysinibacillus fusiformis comb. nov. and Bacillus sphaericus to Lysinibacillus sphaericus comb. nov. *Int J Syst Evol Microbiol* 2007;**57**:1117–25.
4. Andrews JM, Wise R. Susceptibility testing of Bacillus species. *J Antimicrob Chemother* 2002;**49**:1040–2.
5. Bottone EJ. Bacillus cereus, a volatile human pathogen. *Clin Microbiol Rev* 2010;**23**:382–98.
6. Osman S, Satomi M, Venkateswaran K. Paenibacillus pasadenensis sp. nov. and Paenibacillus barengoltzii sp. nov., isolated from a spacecraft assembly facility. *Int J Syst Evol Microbiol* 2006;**56**:1509–14.
7. Antonello A, Weinstein GW. Successful treatment of Bacillus alvei endophthalmitis. *Am J Ophthalmol* 1989;**108**:454–5.
8. Coudron PE, Payne JM, Markowitz SM. Pneumonia and empyema infection associated with a Bacillus species that resembles B. alvei. *J Clin Microbiol* 1991;**29**:1777–9.
9. Reboli AC, Bryan CS, Farrar WE. Bacteremia and infection of a hip prosthesis caused by Bacillus alvei. *J Clin Microbiol* 1989;**27**:1395–6.
10. Wiedermann BL. Non-anthrax Bacillus infections in children. *Pediatr Infect Dis J* 1987;**6**:218–20.