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First Case of Janibacter Indicus Bacteremia in a Pediatric Immunocompetent Patient

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1. ABSTRACT

1.1. Background: The Janibacter genus is a Gram-positive, coryneform bacteria that belong to the Actinobacteria phylum. Bacteria from this group have been associated with bacteremia in immuno-compromised children. In the literature, there are few reported cases of infection, and data is lacking about this species, especially for J. indicus.

Here, we present the first documented case of Janibacter indicus bacteremia in an immunocompetent pediatric patient. This case report aims to provide support for the management of this rare infection and to share our experience for future studies and management. Also, it highlights the importance of advanced microbiological methods for timely diagnosis and management. Further research is needed to define therapeutic approaches and susceptibility patterns better.

Keywords: Janibacter indicus; Bacteremia; Pediatric infection; Antimicrobial susceptibility; Case report

2. INTRODUCTION

The genus Janibacter belongs to the family Intrasporangiaceae and was first described [1]. Since then, a total of ten species have been identified with validly published names: J. limosus, J. terrae, J. melonis, J. anophelis, J. corallicola, J. hoylei, J. alkaliphilus, J. cremeus, J. indicus and J. endophyticus. The natural reservoir is not fully known. Recent studies and reports show that members of the genus Janibacter have been found in various environments, including polluted samples, melons, mosquito midgut, corals, marine sediments, and air samples [2-8]. Other Janibacter species have been identified in vaginal secretions and heart valves [9,10].

Janibacter spp. are rarely implicated in human pathology, typically in cases of bacteremia associated with neoplasms and related immunosuppression [7,11-13].

Another Janibacter infection occurred in an 8-week-old infant without underlying medical conditions who presented with fever without an obvious source of infection, and it was caused [12], The only other reported case of this rare infection was described in an adult patient in a recent study of 2021 [14]. To our knowledge, this is the first case report of J. indicus bacteriemia in an immunocompetent child.

3. CASE PRESENTATION

A 12-year-old girl presented to the Emergency Pediatric Room of the Hospital of Chioggia for fever and swelling in her left arm.

History collection revealed a previously left arm fracture from road trauma treated with a cast appliance for three weeks in February 2023. On cast removal, the arm presented with tumefaction and with a reactive fibrous cord in the anteromedial location associated with an area of hypoesthesia in the anterolateral upper extremity. An orthopedic examination raised a suspicion of erysipelas. Antibiotic therapy with amoxicillin/clavulanic acid 875/125 mg bid and topical gentamicin tid was started. The swelling didn't seem to improve, and fever (39.5°C) with shivering appeared too.

Cardiac activity was rhythmic and the CXR showed no alterations. At the examination, two excoriations in the middle area of the same location were highlighted. The arm's active and passive mobility and extension were preserved entirely and the muscular tone was normal.

On suspicion of deep venous thrombosis, an echocolordoppler of the upper extremity was performed. The echography excluded thrombosis but showed a corpuscular fluid stratum above and below the fascial thickness of about 5 mm, deepening toward the muscle structures and humerus near the humeral joint on the external side. She was then admitted to broad the investigations. Laboratory exams were performed and showed a White Blood Count (WBC) of 19.090/mm^3 with 14.620/mm^3 neutrophils and an elevation of inflammation parameters with C-Reactive Protein (CRP) of 163.5 mg/L, while Procalcitonin (PCT) was negative. Blood cultures were performed and infusion antibiotic therapy with ceftriaxone at a dosage of 1 g a day was started. On the first day of hospitalization, the antimicrobial treatment was then shifted to ampicillin/sulbactam at a dosage of 2gr/1gr four times per day plus clindamycin 600 mg three times per day after consultation with the Infectious Diseases department. After 72 hours of hospitalization, blood cultures tested positive for Janibacter indicus. The antibiotic therapy was confirmed as agreed with the Infectious Diseases specialist due to the clinical improvement of the patient.

A cardiac echocardiography was performed and it was negative for endocarditis.

On orthopedic indication, clinical and ultrasound monitoring were undertaken. At diagnostic completion, a radiograph of the left arm was performed, which excluded fractures in place. On day five of hospitalization, because of the slight increase in size and thickness of the flap found, a surgical cleaning of the lesion was performed, and a drainage for aspiration and purulent fluid collection was positioned. Control blood cultures were performed six days after initiation of antibiotic therapy and were negative, as was the surgical drainage fluid culture. After that, the patient's conditions remained consistently stable, with progressive improvement of the edema and local inflammation in her left arm. The surgical drainage was removed on day 10 of hospital care, after seriate dressings.

The intravenous antibiotic therapy was discontinued on the same day and switched to oral amoxicillin/clavulanic acid treatment

The girl was discharged in apyretic and in excellent overall conditions.

The antimicrobial therapy was continued at home for 21 days and the dressing was changed every two days. Regular wound follow-ups were performed every 7 days with progressive improvement of edema until complete resolution. The last follow-up visit showed minimal and nonsignificant residual edema with mild difficulties in flexion of the limb.

3.1. Microbiology findings

Janibacter indicus was isolated from the blood culture incubated in a pediatric blood culture bottle in a BACTEC FX during the third day of incubation. From a microscopy point of view, Gram-stained smears from the blood culture showed pleomorphic, Gram-variable cocci.

The blood was processed according to this protocol for Direct Identification via MALDI-TOF MS: an aliquot of 5 ml was drawn and transferred to a BD Vacutainer SST II Advance, which was centrifuged at 3500 rpm for 10 minutes.

The pellet was subsequently drawn with a cotton swab and resuspended in 2 ml of sterile physiologic solution (0,9% NaCl). 1,5 mL of this suspension was transferred to an Eppendorf tube and centrifuged at 12000 rpm for 2 minutes.

The supernatant was discarded, and the resulting pellet was resuspended in 30 microliters of formic acid for protein extraction.

Two spots of a polished plate for MALDI-TOF MS were spotted with 1 microliter of the resuspension each, left to air dry, and then covered with the appropriate matrix.

The plate was then inserted into the instrument for identification.

Janibacter indicus was then presumably identified with a score of 1,82, allowing immediate communication with the clinicians.

The specie identification was confirmed the following day from colonies grown on Columbia blood agar.

3.2. Cultural characteristics

The isolate was subcultured on different solid media and incubated in different conditions to assess its cultural characteristics.

The used media were as follows: Columbia 5% Blood Agar (COL-S), Schaedler Agar (SCH), Chocolate Agar (PVX), Brucella Fastidious Agar (BRU), Mannitol Salt Agar (MSA), Colistin-Nalidixic Acid Agar (CNA)

The Incubation conditions were as follows: anaerobiosis 37°C (COL-S, SCH, CNA), aerobiosis 37°C (COL-S, PVX, BRUCELLA, MSA, CNA), 5% CO_2 37°C (COL-S, PVX, BRU, MSA, CNA), aerobiosis 42°C (COL-S, PVX, BRU, MSA, CNA).

The culture plates were read at 24h, 48h, and 72h.

The best growth was recorded on COL-S incubated in aerobiosis at 37°C for 72 hours. The colonies appeared cream-colored, opaque, circular, convex with linear margins, showed no hemolysis, and were characterized by a fecaloid smell.

There was no growth in anaerobic conditions; however, anaerobiosis didn't seem to affect the isolate's survival, given that growth of colonies was recorded on the anaerobic plates (SCH, CNA, and COL-S), which, after the 72h read, were left on the bench at room temperature for another day.

Interestingly, the isolate grew better on COL-S than on PVX, suggesting an impact on its growth from the factors released by the platelets destroyed during its preparation.

A light growth was also recorded on MSA, suggesting halotolerance and a weak capability to metabolize mannitol.

Growth was also present in the culture media incubated at 42°C and in CO_2 atmosphere; however, the best conditions appeared to be on COL-S.

3.3. Antimicrobial susceptibility testing

Antimicrobial susceptibility testing was performed via broth microdilution using two commercial panels, one for Gram-positive bacteria and one for fastidious bacteria. Minimal Inhibitory Concentrations (MICs) were determined as follows.

For the Gram-positive panel, 100 ul of a 0,5 McF suspension of J. indicus was mixed with a Cation-Adjusted Muller Hinton Broth (CAHMB) vial. The mixture was then distributed in the 96-wells plate containing the lyophilized antibiotics. The plate was covered with an adhesive unperforated foil and incubated in aerobic conditions at 37°C for 72 h.

For the fastidious bacteria panel, 200 ul of a 0,5 McF suspension of J. indicus was mixed with a vial of MICRONAUT-H broth. The mixture was then aliquoted in the 96-wells plate containing the lyophilized antibiotics. The plate was covered with a plastic lid and incubated in aerobic conditions at 37°C for 72 h.

Given the long incubation, purity check plates were performed for both inoculations and showed no growth of contaminants. The results are shown in Table 1-3.

Table 1: Minimal inhibitory concentrations - Gram Positive Panel (ITGP).

Drug	MIC (mg/l)	Drug	MIC (mg/l)
DPT	≤ 0,25	TEZ	>1
TGC	≤ 0,125	T/S	≤ 0,25/4,75
DOX*	<0,5	ERY*	1
MOX	≤ 0,125	OXA	>2
AMP*	2	LIZ	≤ 1
LEV*	≤ 1	CLI	1
COX	≤ 4	GEN	≤ 0,25
TPL	≤ 0,25	VAN	≤ 0,5
FUS	≤ 0,25	MUP	>256
AMS	≤ 2/4	DVA	≤ 0,0625
RAM	=0,5	GNH	S
CBP	≤ 0,25	NFT	32
CFL	=0,5	ERC	NEG
SNH	S		

AMP: Ampicillin; AMS: Ampicillin/Sulbactam; COX: Cefoxitin; CFL: Ceftaroline; CBP: Ceftobiprole; CLI: Clindamycin; DVA: Dalbavancyn (RUO); DPT: Daptomycin; DOX: Doxycicline; ERY: Erythromycin; ERC: Erythromycin/Clyndamycin; FUS: Fusidic Acid; GEN: Gentamyicin; GNH: Gentamycin High-Level; LEV: Levofloxacine; LIZ: Linezolid; MOX: Moxifloxacine; MUP: Mupirocine; NFT: Nitrofurantoine; OXA: Oxacillin;

RAM: Rifampicin; SNH: Streptomycin High Level; TEZ: Tedizolid; TPL: Teicoplanin; TGC: Tigecyclin; T/S: Trimethoprim/Sulfamethoxazole; VAN: Vancomycin

Table 2: Minimal inhibitory concentrations - Fastidious bacteria panel (ITHMN).

Drug	MIC (mg/l)	Drug	MIC (mg/l)
CFI	>1	MER	0.125
AZM	>4	T/S	≤ 0,125/2,375
CIP	=0,5	CLR	4
AMP*	2	DOX*	=0,25
CRO	>2	LEV *	=0,5
AMC	1	TET	1
ERY*	1	PEN	1
CTX	>2		

AMC: Amoxicillin/Clavulanate; AMP: Ampicillin; AZM: Azithromycin; CFI: Cefixime; CTX: Cefotaxime; CRO: Ceftriaxone; CIP: Ciprofloxacine; CLR: Clarithromycin; DOX: Doxycycline; ERY: Erythromycin; LEV: Levofloxacine; MER: Meropenem; PEN: Penicillin G; TET: Tetracycline; T/S: Trimethoprim/Sulfamethoxazole

Table 3: A tentative interpretation was performed using the EUCAST method involving PK/PD breakpoints.

Drug	SIR
Penicillin	I
Ampicillin	S
Ampicillin/Sulbactam	S
Amoxicillin/Clavulanate	S
Cefotaxime	R
Ceftriaxone	R
Ceftobiprole	S
Meropenem	S
Ciprofloxacin	S
Levofloxacin	S
Moxifloxacin	S
Gentamycin	S
Dalbavancin	S
Linezolid	S

4. CONCLUSION

To our knowledge, this is the first documented case of an infection due to Janibacter indicus occurring in an immunocompetent pediatric patient. In our case report, we tested all the main antibiotics used to treat skin and soft tissue infections, showing an evident susceptibility to these drugs.

Our case has some limitations. First, we could not recover clear microbiological isolation from the purulent specimen of the abscess due to the drainage performed days after the start of the antimicrobial therapy, which indeed was effective against Janibacter indicus.

Another limitation is that it is impossible to state what was the source of infection clearly. The main hypothesis remains the environmental source when the patient had broken her arm, but this will remain a mere speculation.

In addition, a lack of microbiological information such as the presence of virulence factors, the ability to produce toxins, the unknown mechanisms of resistance and the lack of international MIC breakpoints is a matter of concern.

5. DECLARATIONS

5.1. Consent for publication

Written informed consent was obtained from the patient's legal guardians for publication of this case report and any accompanying images.

5.2. Authors' contributions

FG drafted the manuscript. NG contributed to clinical data collection and patient management. LF performed microbiological analyses. All authors reviewed and approved the final version of the manuscript..

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