Identification of *Brucella* (*Ochrobactrum*) *anthropi* by MALDI-TOF MS from blood: a case report

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Summary

This case report describes the identification of *Ochrobactrum anthropi* (*O.a.*), from a positive blood culture using MALDI-TOF spectrometry system. According to the information available this is the first case in AORN Sant'Anna and San Sebastiano, Caserta, Campania, Italy. This microbe is moderately pathogenic and hazardous, with the potential to cause hospital-acquired infections. It is recommended that it be included in hospital "germ alerts".

Introduction

Brucellaceae family consists of six genera in addition to *Brucella (Ochrobactrum, Crabtreella, Daeguia, Mycoplana, Paenochrobactrum, and Pseudochrobactrum*). *Ochrobactrum* is an alphaproteobacterium belonging to the *Rhizobiales* order divided in six species based on 16S rDNA sequencing (*O. anthropi, O. intermedium, O. tritici, O. grignonense, O. gallinifaecis, and O. lupine*) [9]. *Ochrobactrum anthropi* is a Gram-negative bacillus, non-fermenting bacillus, obligate aerobe bacillus, flagellate bacillus, oxidase-positive and indole-negative bacillus that thrives in the rhizosphere and does not multiply in host cells. This micro-organism exhibits low virulence and seldom causes human infections [9]. It is found in soil and water but can also be isolated from contaminated biological products, such as human wastes, fluids and medical devices [7]. *Ochrobactrum anthropi* has been described as an opportunistic pathogen that causes infections in severely ill or immunocompromised patients with indwelling catheterization, which can commonly lead to clinical manifestations such as catheter-related bloodstream infections [1,8]. In the last decade, *O. anthropi* has been associated to bacteremia, brain empyema, endophthalmitis, septic shock, septic arthritis, endocarditis [4]. In recent years, there have been reports *O. anthropi* clinical isolates resistant to multiple drugs, which have been linked to several hospital outbreaks [5]. Pathogen microbiological characterization is difficult due to its phenotypic similarities with other microorganisms, leading to potential mistakes in its diagnosis [6]. Based on molecular markers and genome comparisons, *Brucella* genus is closest to *Ochrobactrum* genus [3]. This case report describes the identification of *O. anthropi* using MALDI-TOF method and the importance of this emergent *bacterium* in hospital acquired infections. The patient gave her written consent for the publication of this case report.

Case Report

An 84-year-old woman presented to our Emergency Department at AORN Sant'Anna and San Sebastiano with abdominal pain and generalized body aches. The woman had past history of cystitis, hyperthyroidism, hypertension and diabetes mellitus. Physical exam revealed globose abdomen, sore from palpation, hapless. Three days before hospital admission, she displayed rectorargy with blood in the stool and abdominal pain. Laboratory testing was significant for white blood cells (12.94x10^3/uL), Lactate Dehydrogenase (LDH) (337 UI/L), elevated transaminase levels (52 UI/L). A chest X-ray was performed to check for the presence of a pneumonic focus or injuries; however, no sign of infection could be detected. CT and ultrasound abdomen showed no perceivable alterations. Aerobic and anaerobic blood cultures were positive at 47 hours. The patient is currently febrile (38.2°C) and shivering with chills. Microscopic examination on positive blood cultures showed Gram-negative rods observed after Gram staining (Figure 1a) and microbiological culture showed growth on blood, chocolate and also MacConkey agar plates (Figure 1 b, Figure 1 c).

The organism was identified by matrix-assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF MS, Biomèrieux, Marcy l'Étoile, France) as *Ochrobactrum anthropi* with 99.9% accuracy as a "claimed" organism (Figure 2). The identification was repeated with MALDI-TOF MS and confirmed with Vitek2 (Biomèrieux, Marcy l'Étoile, France).

Table 1 summarizes the Minimum Inhibitory Concentrations (MIC) values and interpretations tested with Vitek2. The inter-

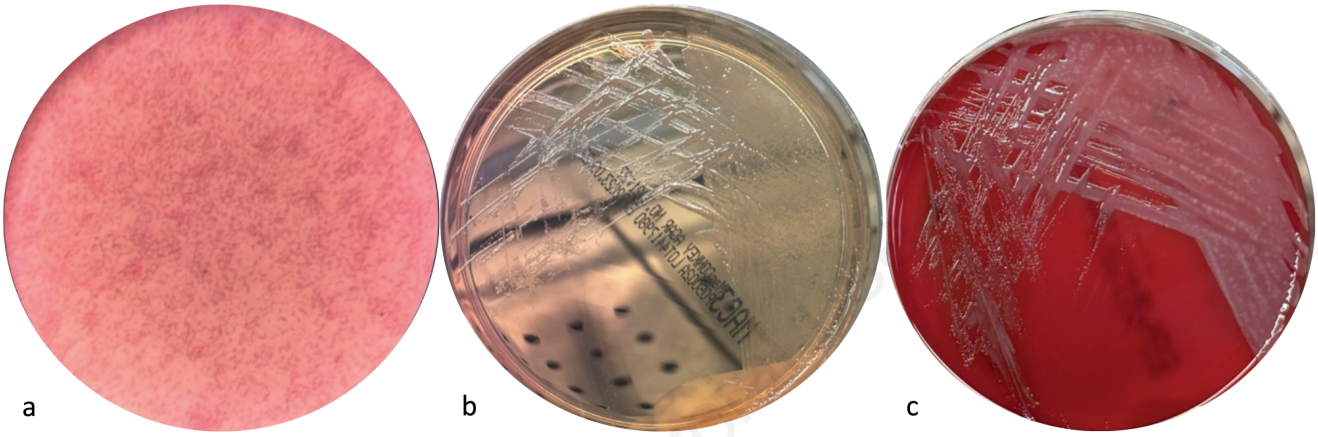

Figure 1. Gram and culture characteristics of *O. anthropi*.

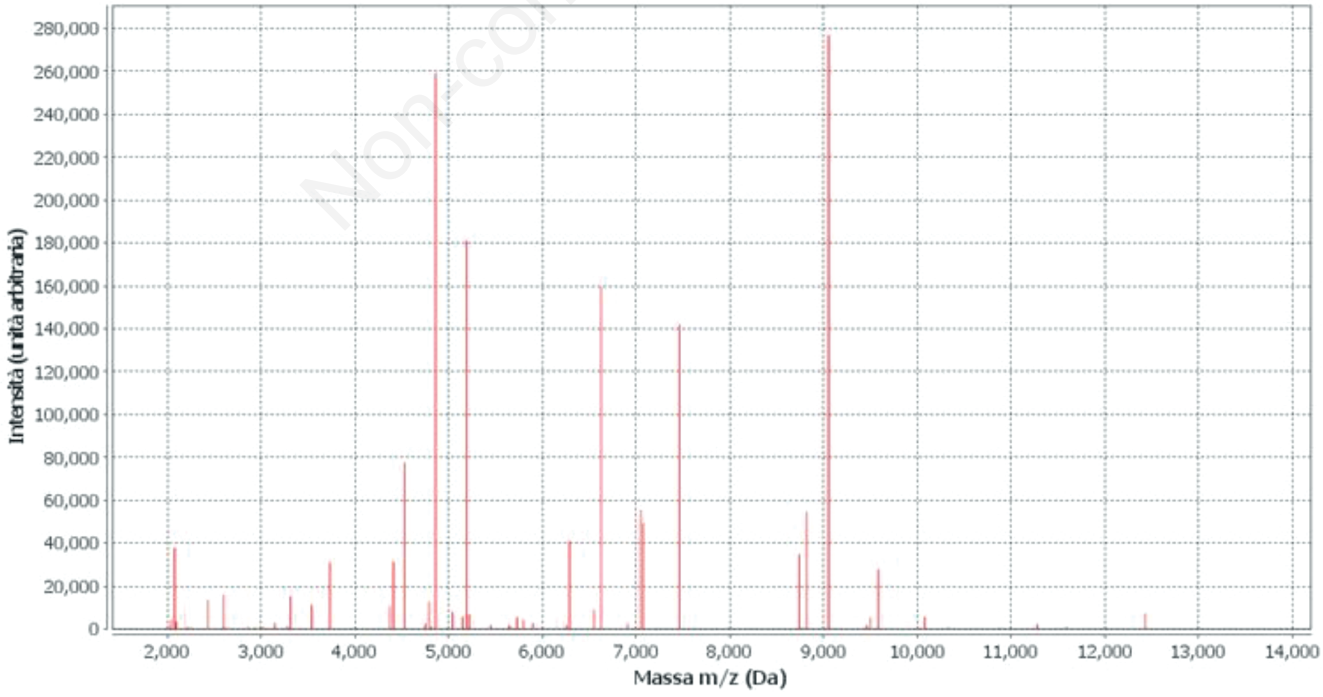

Figure 2. *O. anthropi spectrum*.

Table 1. Antibiotic susceptibility pattern of *O. anthropi*.

Antibiotics	MIC	Interpretation
Amikacin	8	R
Amoxicillin/Clavulanic acid	16	R
Aztreonam	32	R
Cefotaxime	8	R
Ceftazidime	16	R
Ceftazidime/Avibactam	16	R
Ceftolozane/Tazobactam	32	R
Ciprofloxacin	0.25	S
Levofloxacin	0,0094	S
Colistin	2	IE
Ertapenem	0.5	S
Gentamicin	1	R
Imipenem	≤0.5	S
Meropenem	0.5	S
Piperacillin/Tazobactam	32	R
Tigecycline	0.5	S
Tobramycin	≤1	NA
Trimethopim/Sulfamethoxazole	<1	IE

MIC, Minimum Inhibitory Concentration; R, resistant; S, sensible; IE, insufficient evidence; NA, not applicable.

pretation was reported following the European Committee on Antimicrobial Susceptibility Testing (EUCAST) breakpoint tables [2].

The patient received levofloxacin (500mg IV/24 h) for 16 days. A new blood culture sample collected at the follow-up visit showed no bacterial growth after 5 days of incubation. At last follow-up, the patient had no symptoms consistent with relapsed disease.

Discussion

This case report highlights the need for laboratories to adopt modern approaches for the identification and management of potential hospital-acquired infection micro-organisms. In the last years, the *Ochrobactrum* species has been considered an emerging hospital-acquired pathogen, but it has not yet been included among the "common" germ alert and it is likely to be underdiagnosed. Rapid microbiology is considered to be essential for identifying and differentiating the *Brucella species*, particularly due to the phenotypic similarities that makes species characterization by routine biochemical analysis difficult. In the current era of antimicrobial resistance, it is important to consider personalized antibiotic therapies as a potential solution to the therapeutic challenge. *O. anthropi* susceptibility patterns were in accordance with other cases of human infection in contrast to some reported cases that report this bacterium as multidrug-resistant. Treatment options were carefully considered and a combination therapy of ciprofloxacin and amikacin and/or trimethoprim-sulfamethoxazole was chosen. This particular therapy may be considered as a viable option for managing *O. anthropi* infections. *Ochrobactrum* spp. are not currently recognized as serious pathogens, so the idea that they may be harmless should be reconsidered.

Conclusions

Despite the fact that *O.a.* is generally considered to be of low virulence and low risk compared to other non-fermenting Gram-negative *bacteria*, it should not be ignored as a potential cause of hospital-acquired infections. Consequently, it should be included in hospital germ alert notification. In microbiological and epidemiological order, *O.a.* could be a future difficult to threat pathogen, particularly in immunocompromised patients who might report Multi Drug Resistance (MDR) bacterial co-infections.

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