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CASE REPORT

The First Case of *Arthrobacter woluwensis* Bacteremia Diagnosed using MALDI-TOF MS

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SUMMARY

Background: Thus far, seven cases of *Arthrobacter woluwensis* (*A. woluwensis*) infections have been reported globally. Its rarity and overlapping characteristics with other bacterial species make identification challenging, necessitating 16S ribosomal RNA sequencing and whole-genome sequencing.
Methods: **For the first time, *A. woluwensis* bacteremia was diagnosed using matrix-assisted laser desorption/ionization-time of flight mass spectrometry (MALDI-TOF MS).**
Results: **The isolated *A. woluwensis* was susceptible to vancomycin. *A. woluwensis* bacteremia was resolved with the administration of intravenous vancomycin and the removal of the chemoport.**
Conclusions: **MALDI-TOF MS can be useful in diagnosing *A. woluwensis* infections.**
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KEYWORDS

Arthrobacter woluwensis, bacteremia, MALDI-TOF MS

CASE PRESENTATION

In September 2023, a 68-year-old man presented to the Korea University Anam Hospital in Seoul, Korea, with hematuria and weight loss. Computed tomography revealed stomach, colon, and bladder wall thickenings, as well as enlarged lymph nodes in both iliac chains. Following trans-urethral removal of the bladder lesion, a high-grade invasive urothelial carcinoma involving subepithelial connective tissue and lymphovascular structures was diagnosed. Chemotherapy was initiated with 5-fluorouracil and cisplatin on the 18th day of hospitalization and lasted until the 20th day.

On the 38th day, the patient developed a fever peaking at 38.1°C with elevated leukocyte counts and inflammatory markers: leukocyte 14.77×10^9/L; C-reactive protein 0.154 g/L; procalcitonin 3.44×10^{-7} g/L. Blood culture samples from the peripheral veins of both arms and chemoport were collected in aerobic BACT/ALERT FA Plus bottles (bioMérieux, Marcy-l'Etoile, France) and anaerobic BACT/ALERT FN Plus bottles. The samples

Table 1. Reported cases of *Arthrobacter woluwensis* infections.

Year/ country	Gender/ age	Underlying diseases, catheterization	Infection type	Identification methods	Antimicrobial susceptibility test	Treatments	Outcome
1996 Belgium/ Switzerland * [1]	F/33	HIV, CMV retinitis, Port-A catheter	bacteremia	biochemical tests, chemotaxonomic investigations, 16S rRNA sequencing	agar dilution method	2 weeks IV AMP	survived
2004 Switzerland [2]	M/39	IV drug user	infective endocarditis	biochemical tests, chemotaxonomic investigations, 16S rRNA sequencing	disk diffusion assay	6 weeks IV TEC	survived
2006 Korea [3]	M/56	Metastatic colon cancer, subclavian catheter	bacteremia	biochemical tests, 16S rRNA sequencing	E-test	IV VAN, catheter removal	discharged †
2007 Korea [4]	F/91	Stroke	bacteremia	biochemical tests, 16S rRNA sequencing	disk diffusion assay	10 days IV LZD	survived
2012 Korea [5]	F/76	Multiple myeloma, central venous port	bacteremia	biochemical tests, 16S rRNA sequencing	broth microdilution test	19 days IV TEC, catheter removal	survived
2021 France [6]	Unknown/ 52	IV drug user	infective endocarditis	biochemical tests, 16S rRNA sequencing	not tested	TEC, LZD, TMP/SMX, vegetectomy, valve replacement	survived
2021 Taiwan [7]	M/93	Gastrectomy for gastric cancer, perioperative wound infection, central venous catheter	bacteremia	whole-genome sequencing	E-test, VITEK 2 (bioMérieux)	AMP	survived
2024 Korea (present case)	M/68	Metastatic bladder cancer, chemoport	bacteremia	MALDI-TOF MS	E-test	VAN	expired ††

* **Colonies isolated in Belgium and identified in Switzerland.**
† **Discharged against medical advice with aggravation of the underlying diseases.**
†† **Expired due to the aggravation of the underlying diseases.**
F - female, M - male, IV - intravenous, AMP - ampicillin, TEC - teicoplanin, VAN - vancomycin, LZD - linezolid, TMP-SMX - trimethoprim/ sulfamethoxazole.

were incubated on BACT/ALERT VIRTUO systems (bioMérieux), and positive signals were detected in all aerobic bottles after 15 - 21 hours of incubation. In overnight subculture on sheep blood agar plates (BANDIO, Pocheon, Korea), circular, whitish, slightly glistening, fluffy, convex, and non-hemolytic colonies with diameters of 2 - 3 mm formed (Figure 1A). Gram staining revealed gram-positive, club-shaped bacteria arranged in V-, L-, or palisade formations (Figure 1B). The colonies were analyzed with MALDI Biotyper sirius (Bruker, Bremen, Germany) and MBT Compass 4.1 version (Bruker) (Figure 1C). The matched log score was 2.56 when compared to DSM 10495^T strain and 2.50 when compared to DSM 10495^T strain 2, which were from Funke's specimen (Figure 1D) [1]. The high matched log scores of 2.56 and 2.50 indicated *A. woluwensis* with a high probability, allowing for diagnosing *A. woluwensis* bacteremia. The chemoport was removed, and echocardiography was performed to rule out complicated endocarditis.

The Epsilometer test (E-test, bioMérieux) revealed a susceptible breakpoint to vancomycin (MIC (minimum inhibitory concentration) 1.5 µg/mL) and an intermediate breakpoint to benzylpenicillin (MIC 3.0 µg/mL) according to Clinical and Laboratory Standards Institute (CLSI) document M45-ED3 for *Corynebacterium* spp. and related coryneform genera. Intravenous vancomycin was added to the patient who had previously received oral vancomycin. *A. woluwensis* was not detected in subsequent blood cultures after the 40th day. However, on the 55th day, the patient died as a result of complications related to terminal cancer.

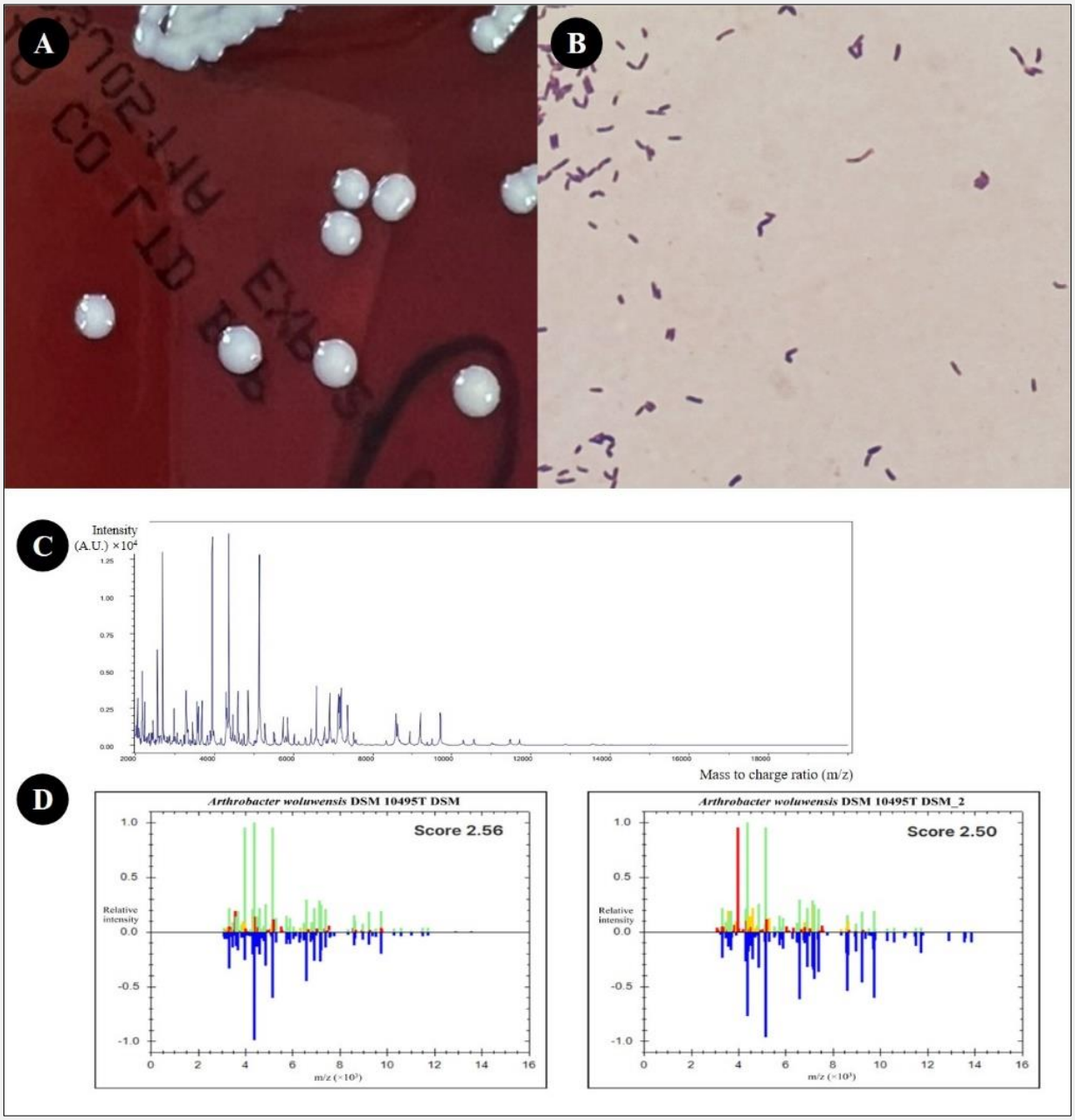

Figure 1. Characteristics of *A. woluwensis.*

A) *A. woluwensis* colonies on blood agar plate showing round, whitish, slightly glistening, fluffy, convex clusters of colonies with diameters of 2 - 3 mm. B) *A. woluwensis* on Gram stain (light microscopy, × 1,000) showing gram-positive, club-shaped bacteria arranged in angular (V- or L-shaped) or palisade formations. C) Mass spectrum of the colony sample on the MALDI Biotyper (Bruker). D) Matched log scores on spectrum matching. Upper half of the graph: colony mass spectrum; lower half of the graph: reference mass spectra for *A. woluwensis* DSM 10495^T strain (left) and *A. woluwensis* DSM 10495^T strain 2 (right).

DISCUSSION AND CONCLUSION

Among various *Arthrobacter* spp., *Arthrobacter woluwensis* is the most well-known species causing human infections [1-7]. *A. woluwensis* is an obligately aerobic, non-spore-forming, non-motile, catalase-positive, and nitrate reductase-negative bacteria [1-7], whose identification is complex because of the low incidence of infection, overlapping characteristics with other bacterial species, especially *Corynebacterium*, and frequent misidentifications on commercial kits [2-6]. Misidentifications for *Corynebacterium aquaticum* (synonym, now reclassified as *Leifsonia aquatica*) [2,3,5], *Microbacterium* spp./*Leifsonia aquatica* [4] on the API Coryne kit (bioMérieux), and *Corynebacterium jeikeium* [6] on phenotyping method have been reported. Thus, the identifications of *A. woluwensis* have primarily relied on molecular approaches such as 16S rRNA sequencing and whole-genome sequencing [1-7]. However, in our case, *A. woluwensis* bacteremia can be effectively diagnosed using MALDI-TOF MS.

Thus far, there have only been seven reported cases of *A. woluwensis* human infections, with five cases of bacteremia and two cases of infective endocarditis (Table 1). Including the patient in our case, the median age was 62.0 years (range: 33 - 91 years), with four males, three females, and one unknown. Of the patients, 62.5% (n = 5) had central venous catheters, 37.5% (n = 3) had solid tumors (colon, stomach, or bladder cancer), while 12.5% (n = 1) had a hematologic malignancy (multiple myeloma). 25.0% (n = 2) were intravenous drug users, and 12.5% (n = 1) had HIV. Immunocompromised status and central catheterization have been suggested as the major risk factors for *A. woluwensis* bacteremia [7]. Although 87.5% (n = 7) were sufficiently treated and recovered with antibiotic treatment and catheter removal, 12.5% (n = 1) with infective endocarditis required vegetectomy and valve replacement. Although *A. woluwensis* infections were successfully treated in all instances, clinical outcomes varied, with 75.0% (n = 6) surviving and 25.0% (n = 2) succumbing to the deterioration of their underlying diseases.

MALDI-TOF MS utilizes molecular fingerprints that are distinct and unique to each species [8]. The MALDI Biotyper (Bruker) compares the mass spectra of colonies to those of existing reference libraries, and the log score corresponding to each specimen is calculated and ranked, a process known as pattern matching [9,10]. The reference library's spectrum is represented in the lower half of the graph as a blue column. The colony's spectrum is represented in the upper half of the graph as a green column when a peak completely matches the reference spectrum, yellow when a peak partially matches, and red when a peak does not match at all (Figure 1D). The Bruker's scoring criteria are represented as log scores ranging from 0 to 3. A log score of ≥ 2.0 indicates high confidence identification (species-level identification), ≥ 1.7 indicates low confidence identification (genus-level identification), and < 1.7 indicates unreliable identification [10,11]. The log score of 2.56 for *A. woluwensis* DSM 10495^T strain and 2.50 for *A. woluwensis* DSM 10495^T strain 2 significantly exceeded the minimum threshold in species-level identification, strongly indicating the identification of *A. woluwensis* (Figure 1D).

As technology progresses and reference libraries expand, the use of MALDI-TOF MS for pathogen identification becomes increasingly effective [10,12]. Aside from MALDI Biotyper, Vitek MS (bioMérieux) RUO version 4.16 and microIDsys (ASTA, South Korea) coreDB version 1.28.01 include databases for *A. woluwensis*. However, MALDI-TOF MS is not a one-size-fits-all solution. Given the risk of false negativity in blood cultures and impurity of the colonies, repeated testing, pretreatment measures, and 16S rRNA sequencing can be supplemented [8,9]. Furthermore, it is vital to update reference libraries continuously to reflect new taxonomy and encompass a broader spectrum of pathogens [9,12].

In our present case, the isolated *A. woluwensis* is susceptible to vancomycin and intermediate to benzylpenicillin, which is in line with previous studies [1-7]. Previous studies reported 100.0% susceptibility to vancomycin, with 7 susceptible results on 7 tests (7/7), followed by teicoplanin (3/3), tetracycline (2/2), trimethoprim/sulfamethoxazole (2/2), and linezolid (1/1). Resistances to penicillin, ampicillin, amoxicillin-clavulanic acid, oxacillin, cephalosporin, imipenem, daptomycin, gentamicin, erythromycin, clindamycin, ciprofloxacin, and rifampin were observed in subsets of cases [1-7].

In conclusion, microbiologists and clinicians should be aware of *A. woluwensis* infections, particularly in immunocompromised patients undergoing catheterization. MALDI-TOF MS as well as molecular tests may be effective for identification. Vancomycin can be considered primarily. When endocarditis is complicated, surgical interventions should be undertaken. Competent clinical decision-making for diagnosis and treatment is the key.

Declaration of Interest:
None declared.

References:

1. Funke G, Hutson RA, Bernard KA, Pfyffer GE, Wauters G, Collins MD. Isolation of *Arthrobacter* spp. from clinical specimens and description of *Arthrobacter cumminsii* sp. nov. and *Arthrobacter woluwensis* sp. nov. J Clin Microbiol 1996;34(10): 2356-63. (PMID: 8880479)

2. Bernasconi E, Valsangiacomo C, Peduzzi R, Carota A, Moccetti T, Funke G. *Arthrobacter woluwensis* subacute infective endocarditis: case report and review of the literature. Clin Infect Dis 2004;38(4):e27-31. (PMID: 14765360)

3. Shin KS, Hong SB, Son BR. [A Case of Catheter-Related Bacteremia by *Arthrobacter woluwensis*]. Korean J Lab Med 2006; 26(2):103-6. (PMID: 18156709)

4. Kim G-Y, Suh J-T, Choi S-K, Lee H-J. A case of bacteremia caused by *Arthrobacter woluwensis*. Korean Journal of Clinical Microbiology 2007:160-3. https://www.acm.or.kr/wp-content/uploads/2024/05/ACM-1002-15.pdf

5. Park CH, Han MS, Kim JK, et al. Development of *Arthrobacter woluwensis* bacteremia in a patient with multiple myeloma: a case report and comprehensive literature review. Infection & Chemotherapy 2012;44(3):205-9. https://icjournal.org/DOIx.php?id=10.3947/ic.2012.44.3.205

6. Durand C, Kouchit Y, Prots L, et al. A case of infective endocarditis caused by Arthrobacter woluwensis. Eur J Clin Microbiol Infect Dis 2021;40(6):1329-31. (PMID: 33432493)

7. Li SY, Kao CC, Hu YC, et al. Arthrobacter woluwensis Bacteremia: A Clinical and Genomic Report. Pathogens 2021;10(4): 443. (PMID: 33917709)

8. Li D, Yi J, Han G, Qiao L. MALDI-TOF Mass Spectrometry in Clinical Analysis and Research. ACS Meas Sci Au 2022;2(5): 385-404. (PMID: 36785658)

9. Patel R. MALDI-TOF MS for the diagnosis of infectious diseases. Clin Chem 2015;61(1):100-11. (PMID: 25278500)

10. Kostrzewa M, Nagy E, Schröttner P, Pranada AB. How MALDI-TOF mass spectrometry can aid the diagnosis of hard-to-identify pathogenic bacteria - the rare and the unknown. Expert Rev Mol Diagn 2019;19(8):667-82. (PMID: 31303071)

11. Kim KJ, Yun SG, Cho Y, Nam MH, Ko YJ, Lee CK. Evaluation of a sterile, filter-based, in-house method for rapid direct bacterial identification and antimicrobial susceptibility testing using positive blood culture. Eur J Clin Microbiol Infect Dis 2023;42(6): 691-700. (PMID: 37012540)

12. Welker M, Van Belkum A, Girard V, Charrier JP, Pincus D. An update on the routine application of MALDI-TOF MS in clinical microbiology. Expert Rev Proteomics 2019;16(8):695-710. (PMID: 31315000)