



콩줄기에 찔린 후 *Kosakonia cowanii*에 의해 발생한 손 건초염

Tenosynovitis of the Hand Caused by *Kosakonia cowanii* After Being Pricked by a Beanstalk

윤신영 · 김정림 · 홍기호 · 용동은 · 이혁민

Shin Young Yun, M.D., Jung Rim Kim, Bachelor, Ki Ho Hong, M.D., Dongeun Yong, M.D., Hyukmin Lee, M.D.

연세대학교 의과대학 진단검사의학교실 및 세균내성연구소

Department of Laboratory Medicine and Research Institute of Bacterial Resistance, Yonsei University College of Medicine, Seoul, Korea

A 77-year-old man with pain in his left hand after being pricked by a beanstalk visited an emergency center. Symptoms and laboratory findings suggested infectious tenosynovitis. Using the Bruker Biotyper (Bruker Daltonics, Germany), *Kosakonia cowanii* was identified in the swab culture and was confirmed by full-length 16S ribosomal RNA (rRNA) and *gyrB* sequencing. To the best of our knowledge, this is the first reported case of *K. cowanii* infection in Korea. Performing only 16S rRNA sequencing was insufficient for accurate identification, and *gyrB* sequencing or using the Bruker Biotyper (Bruker Daltonics, Germany) confirmed the identification. A positive sorbitol fermentation test can indicate the microorganism to be *K. cowanii* even if the VITEK 2 system (bioMérieux, France) identifies it as *Pantoea* spp. Additionally, *K. cowanii* may be resistant to beta-lactam antibiotics, including cefazolin; therefore, it may be appropriate to use carbapenems, third- or fourth-generation cephalosporins, aminoglycosides, fluoroquinolones, and trimethoprim/sulfamethoxazole. Finally, antibiotic susceptibility testing (AST) should be conducted using a reference method, and Etest AST is unreliable for *K. cowanii*, especially with beta-lactam/beta-lactamase inhibitors.

Key Words: *Kosakonia cowanii*, Tenosynovitis, 16S rRNA, DNA gyrase B subunit

CASE REPORT

A 77-year-old male, with underlying coronary artery obstructive disease and diseases of idiopathic pulmonary fibrosis, hypertension, diabetes, and dyslipidemia, visited the emergency center presenting with left hand pain. He was administered aspirin, trim-

etazidine, sacubitril/valsartan, torasemide, neustatin, metformin, dapagliflozin, lansoprazole, theobromine, bromhexine, and lafutidine at our hospital. Three days prior to visiting the emergency center, he fell and his left palm was pricked by a beanstalk. A day later, he visited a local hospital for treatment and was administered cefazolin intravenously. Computed tomography (CT) was performed at a local hospital and infectious tenosynovitis of the left hand was suspected. He was then referred to our hospital for emergency surgery. Approximately 2 cm of linear abrasion with pus and sanguineous discharge were observed in the middle of the left palm (Fig. 1). The left hand was diffusely swollen, and the patient complained of heating sensation, tenderness, and pain. No neurological deficits or blood circulation problems were found. Vital signs were 103/69 mmHg for systolic/diastolic blood pressure, 118/min for pulse rate, 18/min for respiratory rate, and 36.8°C for body temperature. The routine laboratory test detected leukocyte count of 10,820/mm³ with 72.7% of neutrophils, erythrocyte sedimentation rate of 26 mm/h (reference interval [RI], 0.0–22.0 mm/h), C-reac-

Corresponding author: Hyukmin Lee, M.D., Ph.D.

<https://orcid.org/0000-0002-8523-4126>

Department of Laboratory Medicine and Research Institute of Bacterial Resistance, Yonsei University College of Medicine, 50-1 Yonsei-ro, Seodaemun-gu, Seoul 03722, Korea
Tel: +82-2-2228-2446, Fax: +82-2-2228-8353, E-mail: hmlee.labmed@gmail.com

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Fig. 1. Clinical manifestations of the patient. Approximately 2 cm of linear abrasion with pus and sanguineous discharge were observed in the middle of the left palm. The patient's left hand was diffusely swollen.

tive protein of 119.4 mg/L (RI, 0–8 mg/L), and procalcitonin of 0.14 ng/mL (RI, 0.00–0.50 ng/mL). Typical symptoms and laboratory findings strongly suggested infectious tenosynovitis. The patient was referred to another hospital because of emergency surgery difficulties after collecting the wound discharge for microbiological investigations.

The white blood cells (WBCs) were 3+ in gram staining of wound discharge, but no organism was seen by microscopy. However, few medium-sized, colorless, smooth, convex, and glistening colonies were observed on blood agar and few medium-sized, pink, smooth, convex, punctate, umbilicated, and glistening colonies were observed on MacConkey agar plates incubated at 35°C under 5% CO₂ for 24 hours (Fig. 2). The microorganisms were medium-to-long, plump, gram-negative rods (Fig. 3) and were oxidase-negative and catalase-positive. In the triple sugar iron medium, the slant and the butt were yellow, and gas generation was confirmed, but hydrogen sulfide was absent. The Bruker Biotyper (Bruker Daltonics, Bremen, Germany) identified the microorganism as *Kosakonia cowanii* with a score of 2.12, which is reliable for species level identification according to the manufacturer's instruction. As *K. cowanii* is a rarely identified bacteria in clinical



Fig. 2. Morphology of *Kosakonia cowanii* colony on blood agar plate after incubation for 24 hr.

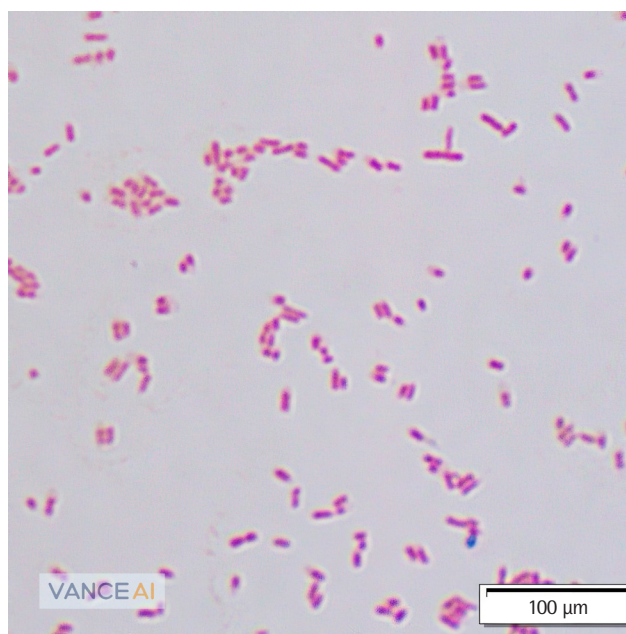


Fig. 3. Gram-stain of *Kosakonia cowanii* (×1,000). Medium-to-long, plump, gram-negative rods were observed by Gram staining.

laboratories, the VITEK 2 system (bioMérieux, Marcy l'Étoile, France) and VITEK MS (bioMérieux) were additionally used for confirming the results of bacterial identification. However, the former was unable to identify this isolate and the latter misidentified the isolate as *Pantoea* spp. with 98% probability (Table 1). To

Table 1. Biochemical characteristics of *Kosakonia cowanii*

	Type strain 888-76 [5]	Current case
Acid production		
Adonitol	-	-
L-Arabitol	-	-
Cellobiose	+	+
Glucose	+	+
Maltose	+	+
Mannitol	+	+
Mannose	+	+
Sucrose	+	+
Sorbitol	+	+
Tagatose	-	-
Trehalose	+	+
Citrate	-	+
β-Galactosidase	+	+
β-Glucuronidase	+	-
β-Xylosidase	-	+
Indole	+	-
Lysine decarboxylase	-	-
Malonate	-	-
Ornithine decarboxylase	-	-
Urease	-	-

solve the discrepancy, full-length 16S ribosomal RNA (rRNA) gene sequencing was performed for species-level identification, according to the guidelines of the Clinical and Laboratory Standards Institute (CLSI) [1]. The amplicon was sequenced by Macrogen (Korea) to yield 16S rRNA gene sequence. The length of the sequence was 1455 bp. BLAST (<http://www.ncbi.nlm.nih.gov/>) search was conducted and the EzBioCloud (<http://eztaxon-e.ezbiocloud.net/>) [2] was employed using the sequence. For species level, we considered over 99% homology as acceptable criteria. However, we could not identify the sequence accurately because some of the sequences with over 99% homology included the 16S rRNA gene sequence of *K. cowanii*, *Salmonella bongori*, and *Atlantibacter bermannii*. Based on a previous report, we additionally performed *gyrB* sequencing using the primers UP1 and 181r [3]. Finally, the microorganism was identified as *K. cowanii* with 99.04% identity, based on similarity with 521 base pairs of *K. cowanii* strain Pa82 (GenBank accession number CP069319.1).

The antimicrobial susceptibility test (AST) was performed using MicroScan (Beckman Coulter, CA, USA) (Table 2). Antimicrobial susceptibility was interpreted according to the CLSI M100 guidelines [4]. This isolate was resistant to ampicillin, ampicillin/sulbactam, amoxicillin/clavulanic acid, cefoxitin, and cefuroxime. It was

susceptible to penicillin in a dose dependent manner, intermediately susceptible to colistin, and highly susceptible to the other tested antibiotics. Due to the concentration range of antibiotics, the minimal inhibitory concentrations (MIC) of ciprofloxacin and levofloxacin could not be interpreted precisely by MicroScan. We also performed Etest (bioMérieux) (Table 2). The Etest results were concordant with the MicroScan results, except in the case of levofloxacin and amoxicillin/clavulanic acid. The levofloxacin MIC in the Etest was interpreted as susceptible. Interestingly, the MIC of amoxicillin/clavulanic acid was significantly different, resulting in a “resistant” result according to MicroScan and a “susceptible” result according to Etest.

DISCUSSION

The name *Enterobacter cowanii* was first proposed in 2000 and changed to *K. cowanii* in 2013 [5, 6]. The *Kosakonia* spp. was differentiated from the *Enterobacter* spp. using the multilocus sequence analysis method, which analyzed the genes of *gyrB*, *rpoB*, *infB*, and *atpD* [6]. There are nine species of *Kosakonia*, including *K. cowanii*. *K. cowanii* belongs to the family Enterobacteriaceae, order Enterobacterales, and class Gammaproteobacteria. It is a gram-negative rod, facultatively anaerobic, oxidase-negative, catalase-positive, and non-spore-forming bacterium, which ferments glucose and reduces nitrates to nitrites [7]. Most *K. cowanii* individuals are motile and have peritrichous flagella; they grow well on MacConkey agar and have pink, smooth, convex, punctate, umbilicated, glistening colonies [7].

K. cowanii is reported to cause bacterial wilt in some plants [8, 9]; however, its human pathogenicity is not well known. Few case reports have described *K. cowanii* infection, including bacteremia in neonates [3], rhabdomyolysis as a result of bacteremia preceded by a rose thorn prick [10], and acute cholecystitis [11]. *K. cowanii* infection seems to occur from exogenous sources [10] and endogenous sources, such as the patient’s microbiota [3, 11]. The possibility of *K. cowanii* infection must be considered in patients with plant-related injuries. In this case, the infectious synovitis was caused by the prick of a beanstalk.

K. cowanii grows well in routine laboratory cultures; however, its identification method is limited. The VITEK MS (bioMérieux) and VITEK 2 systems (bioMérieux) provide no information on *K. cowanii* and the VITEK 2 system (bioMérieux) misidentified it as

Table 2. Comparison between the results of *Kosakonia cowanii*'s antibiotic susceptibility test in the current case and the results in previous reports

Antibiotics	Berinson B [11]		Washio K [10]		Duployez C [3]		Current case			
	Etest with BMD		N/A		VITEK 2		MicroScan		Etest	
	MIC	Susceptibility	MIC	Susceptibility	MIC	Susceptibility	MIC	Susceptibility	MIC	Susceptibility
Ampicillin	≥ 256	R	> 16	R	-	-	> 16	R	-	-
Piperacillin	8	S	> 64	R	64	R	16	SDD	-	-
Amoxicillin/clavulanic acid	-	-	-	-	≤ 2	S	> 16	R	1.5	S
Ampicillin/sulbactam	≤ 2	S	< 8	S	-	-	> 16	R	-	-
Piperacillin/tazobactam	≤ 0.5	S	< 16	S	-	-	≤ 8	S	0.5	S
Cefepime	-	-	< 2	S	≤ 1	S	≤ 1	S	-	-
Cefotaxime	-	-	< 1	S	≤ 1	S	≤ 1	S	< 1	S
Ceftriaxone	-	-	< 1	S	≤ 1	S	-	-	< 0.5	S
Cefoxitin	4	S	-	-	-	-	> 16	R	-	-
Cefuroxime	-	-	-	-	-	-	> 16	R	-	-
Ceftazidime	-	-	< 4	S	-	-	≤ 1	S	< 1	S
Aztreonam	-	-	< 4	S	-	-	≤ 1	S	-	-
Doripenem	-	-	-	-	-	-	≤ 1	S	-	-
Ertapenem	-	-	-	-	-	-	≤ 0.5	S	-	-
Imipenem	-	-	-	-	-	-	≤ 1	S	< 0.5	S
Meropenem	≤ 0.0032	S	< 1	S	-	-	≤ 1	S	< 0.25	S
Colistin	≤ 1	I	-	-	-	-	≤ 2	I	-	-
Gentamicin	-	-	< 2	S	-	-	≤ 2	S	< 0.5	S
Tobramycin	-	-	-	-	-	-	≤ 2	S	-	-
Amikacin	-	-	< 4	S	-	-	≤ 8	S	-	-
Tetracycline	-	-	-	-	-	-	≤ 4	S	-	-
Minocycline	-	-	< 2	S	-	-	≤ 4	S	-	-
Ciprofloxacin	≤ 0.04	S	-	-	-	-	≤ 0.5	S or I	-	-
Levofloxacin	-	-	< 0.5	S	-	-	≤ 1	S or I	< 0.5	S
Trimethoprim/ Sulfamethoxazole	-	-	< 2	S	-	-	≤ 2	S	0.125	S
Chloramphenicol	-	-	-	-	-	-	≤ 8	S	-	-

Abbreviations: BMD, broth microdilution; MIC, minimal inhibitory concentration; S, susceptible; I, intermediate; SDD, susceptible-dose-dependent; R, resistant.

Pantoea spp., which was the same as that obtained in a previous report [3, 11]. Additionally, the colony morphology is very similar between the *Pantoea* spp., and *K. cowanii*, making it difficult to differentiate the two organisms [7, 12]. The biochemical characteristics of *K. cowanii* are similar to those of *Pantoea* spp., but if the organisms produce gases during glucose, dulcitol or sorbitol fermentation, they are more likely to be *K. cowanii* [5]. In this case, *K. cowanii* showed the same biochemical characteristics as previously reported (Table 1) [5, 7], and importantly, with positive sorbitol fermentation. For *Pantoea agglomerans* ATCC 27155 and *Pantoea dispersa* ATCC 14589, sorbitol fermentation is negative [13], although there are reports of some *Pantoea* spp. isolates showing sorbitol fermentation [14, 15]. In addition, full-length 16S rRNA sequencing is insufficient to differentiate among *K. cowanii*, *S. bongori*, and *A. bermannii*; therefore, 16S rRNA with additional

gyrB sequencing is the most accurate method to identify *K. cowanii*. The Bruker Biotyper (Bruker Daltonics) with reliable scores is also conveniently available in clinical laboratories. Additionally, a positive sorbitol fermentation test can indicate the microorganism to be *K. cowanii* even if the VITEK 2 system (bioMérieux) identifies it as *Pantoea* spp. in patients who are pricked by plants or have a plant-related job.

The antibiograms of the isolates in this study and previous reports are shown in Table 2 [3, 10, 11]. The *Kosakonia* genomes were known to lack AmpC β -lactamase [16]; however, in this study, the isolate seemed to have a mechanism for acquired resistance. The isolate was resistant to first- and second-generation cephalosporin antibiotics; therefore, it was likely to be resistant to cefazolin. In general, cefazolin is widely used for skin and soft tissue infections; however, it may not be appropriate in this case. It may

be appropriate to use carbapenems, third- or fourth-generation cephalosporins, aminoglycosides, fluoroquinolones, or trimethoprim/sulfamethoxazole. We also performed some antibiotic susceptibility tests using Etest, most of which showed concordant results with MicroScan. However, the AST results for amoxicillin/clavulanic acid showed discrepancies. Some reports about the discrepancy between the broth microdilution method and other beta-lactams/beta-lactamase inhibitors for Enterobacteriaceae exist [17-19], which may be because beta-lactamase is better expressed in liquid media. Therefore, Etest for *K. cowanii* should be avoided, especially with beta-lactam/beta-lactamase inhibitors. Meanwhile, *Pantoea* spp. is also usually susceptible to many antibiotics, but resistance to amoxicillin/clavulanic acid or ampicillin appears to be common [20].

The limitation of this study was that we could not follow-up on the patient's prognosis. In addition, we did not perform cefazolin AST. Therefore, we could not predict or evaluate the response of this isolate to cefazolin treatment. Finally, whole-genome sequencing was not performed. This might be helpful in determining the antimicrobial resistance mechanisms of *K. cowanii*.

In conclusion, *K. cowanii* can cause infectious tenosynovitis, and this is the first case of human infection in Korea. 16S rRNA sequencing is not enough to identify *K. cowanii*, and *gyrB* sequencing should be performed as well. Bruker Biotyper (Bruker Daltonics) also seems to be good for identify it. A positive sorbitol fermentation test can indicate the microorganism to be *K. cowanii* even if the VITEK 2 system (bioMérieux) detects the presence of *Pantoea* spp. Additionally, *K. cowanii* may be resistant to beta-lactam antibiotics, including cefazolin; therefore, it may be appropriate to use carbapenems, third- or fourth-generation cephalosporins, aminoglycosides, fluoroquinolones, and trimethoprim/sulfamethoxazole. Furthermore, AST should be conducted using a reference method, and AST using the Etest is unreliable for *K. cowanii*, especially with beta-lactam/beta-lactamase inhibitors.

요 약

77세 남성이 콩줄기에 찔려 생긴 왼손 통증으로 응급센터를 방문하였다. 전형적인 증상과 실험실 소견은 감염성 건초염을 강하게 시사했다. 먼봉 배양에서 *Kosakonia cowanii*가 Bruker Biotyper (Bruker Daltonics, Germany)로 확인되었으며, 전체 16S 리보솜 RNA와 *gyrB* 염기서열분석으로 확인되었다. 이는 국내 최초로 보

고된 *K. cowanii* 인체 감염 사례이다. 16S rRNA 염기서열분석만 시행하는 것은 정확한 동정을 하기에 충분하지 않았으며, *gyrB* 염기서열분석을 추가하거나 Bruker Biotyper (Bruker Daltonics)를 사용하면 동정이 가능하다. 혹은 만약 *Pantoea* spp.가 Vitek 2 system (bioMérieux, France)에서 의심되지만 소르비톨 발효가 양성인 경우 *K. cowanii*를 의심할 수 있다. 또한 *K. cowanii*는 세파졸린을 포함한 베타락탐 항생제에 내성이 있을 수 있으므로 그러한 경우에는 카바페넴, 3세대 또는 4세대 세팔로스포린, 아미노글리코사이드, 플루오로퀴놀론 또는 트리메토프림/슈팔메톡사졸을 사용하는 것이 적절할 수 있다. 마지막으로 항생제감수성검사는 표준검사 방법을 사용해야 하며 Etest를 이용한 항생제감수성검사는 아직 신뢰하기 어렵다.

Conflicts of Interest

None declared.

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- cowanii*, *E. radicincitans*, *E. oryzae* and *E. arachidis* into *Kosakonia* gen. nov. as *Kosakonia cowanii* comb. nov., *Kosakonia radicincitans* comb. nov., *Kosakonia oryzae* comb. nov. and *Kosakonia arachidis* comb. nov., respectively, and *E. turicensis*, *E. belveticus* and *E. pulveris* into *Cronobacter* as *Cronobacter zurichensis* nom. nov., *Cronobacter belveticus* comb. nov. and *Cronobacter pulveris* comb. nov., respectively, and emended description of the genera *Enterobacter* and *Cronobacter*. Syst Appl Microbiol 2013;36:309-19.
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