

Nosocomial outbreak of *Aeromonas hydrophila* surgical site infections after spinal surgery: Identification and control

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Abstract

Background: *Aeromonas hydrophila* surgical site infections (SSIs) were diagnosed in April 2017 in four patients who had received spinal surgery. We launched an outbreak investigation to identify the source, and accordingly, preventive and control measures were implemented.

Methods: Environmental samples and samples from the healthcare providers were collected for microbiological analysis. The clonal relatedness of *A. hydrophila* strains was determined by pulsed-field gel electrophoresis (PFGE) and multilocus sequence typing (MLST). Whole genome sequencing (WGS) of one clinical *A. hydrophila* isolate (AE456) was performed using an Illumina NovaSeq PE150.

Results: We identified eight case patients with SSIs due to *A. hydrophila* in orthopaedic ward 2 (three males; median age, 58 years). Strict infection control measures were adopted, particularly contact precautions and unit disinfection. We also identified *A. hydrophila* from water in a fish tank. PFGE and MLST revealed identical patterns and STs among the 10 clinical *A. hydrophila* strains (clone A, ST 517), which were different from those of the strains from the fish tank (clone B, ST518). WGS of isolate AE456 revealed the presence of *cepS*, *cphA* and *bla*_{OXA-12} genes encoding resistance to β -lactams. All patients recovered after antimicrobial therapy and/or surgical debridement. After removal of the fish tank, no new case occurred, and the outbreak was stopped.

Conclusions: *Aeromonas hydrophila* is rare, but severe, pathogen in surgical infections and caused long hospital stay and physical suffering. Strict measures, including environmental disinfection and contact precautions, are needed to prevent infection outbreak after surgery.

Introduction

Aeromonas hydrophila, which is mesophilic, gram negative, rod shaped, and fermentative, is a common fresh or brackish water-borne pathogen that can cause a variety of human diseases, including gastroenteritis, hepatobiliary infection, skin and soft-tissue infections, septicemia, meningitis and endocarditis [1–4]. *A. hydrophila* can produce rapidly progressing wound infections in healthy individuals after trauma and burns and is recognized as an agent of skin and soft-tissue infections associated with water exposure [5–7]. Surgical site infection (SSI) is a significant complication and is associated with increased duration of hospitalization, high healthcare costs, and poor patient outcomes [8].

SSIs due to *A. hydrophila* have rarely been reported. Daniel Tena and his colleagues reviewed 24 cases of SSIs due to *Aeromonas* species and found that SSIs occurred rapidly after abdominal or pelvic surgeries (91.3%), possibly due to endogenous sources [9]. Exogenous sources such as exposure of wounds to contaminated water or leech therapy have also been reported for SSIs due to *A. hydrophila* [10]. *A. hydrophila* has often been reported to cause infection outbreaks in fish stocks,[11] but outbreaks in humans are relatively rare. An outbreak of wound infections with *A. hydrophila* occurred after a “mud football” competition [12].

In our hospital, only sporadic and nonrelated cases of *A. hydrophila* infection were identified during 2008–2015. In April 2017, four cases of postoperative *A. hydrophila* SSIs were diagnosed in orthopaedic ward 2. Two additional cases were identified after review of the clinical and microbiological records of patients in the same ward in 2016. We describe here an outbreak investigation to identify the source and transmission route, as well as the clinical and molecular epidemiology of this outbreak.

Methods

Setting

Peking University Third Hospital is a 1400-bed tertiary-care teaching hospital. There are six wards in the orthopaedic department with a total of 200 hospital beds. Approximately 10,000 patients received orthopaedic surgeries annually, including spinal, articular and traumatic surgeries.

Background of the outbreak

In April 2017, four patients in orthopaedic ward 2 experienced postoperative SSIs, showing fever, pain, and purulent drainage from the surgical site. Magnetic Resonance Imaging (MRI) showed a large amount of effusion deep in the wound. Wound drainage and/or peripheral blood samples were collected and sent for microbiological analysis. The following day, *A. hydrophila* was recovered from the pus or drainage samples. One patient also tested positive for *A. hydrophila* in blood samples.

An outbreak of nosocomial infection due to *A. hydrophila* was suspected. All patients in the same ward were checked carefully for possible SSIs. To obtain baseline prevalence data, hospital microbiological laboratory databases were reviewed for *A. hydrophila* isolates identified from 2008 to 2017, which was the period for which complete records were available. Two similar cases were identified in June and July 2016 in orthopaedic ward 2. Physicians reviewed the 6 cases for clinical manifestations and risk factors for the acquisition of *A. hydrophila* SSIs in detail. The phenotypes and drug resistance patterns of the *A. hydrophila* isolates were compared by microbiologists. After discussion and combination of the data, infection control measures were taken, and the first environmental investigation was initiated but failed to identify the *A. hydrophila* isolate. Unexpectedly, in May 2017, a seventh patient developed a postoperative SSI of *A. hydrophila* in the same ward, and in July 2017, an eighth patient was identified as having a postoperative *A. hydrophila* SSI and bloodstream infection in orthopaedic ward 6. He was previously hospitalized in orthopaedic ward 2 for two days after operation.

Case definition

A case was defined as any patient with symptoms and signs consistent with SSIs and drainage or blood culture positive for *A. hydrophila* since 2008. SSI was defined as erythema, induration, pain, and septic drainage from the surgical site. Postoperative *A. hydrophila* infection was defined as being nosocomial if the infection was acquired within 30 days after a surgical procedure [13].

Infection control strategies implemented

An infection control team consisting of surgeons, microbiologists, nosocomial infection experts and head nurses in the hospital was quickly set up to stop the occurrence of similar infections. All healthcare workers in the ward were informed of the possibility of an *A. hydrophila* SSI outbreak. Heightened infection control measures were enforced, especially contact precautions for all infected patients. The team initiated the first environmental and healthcare worker screening cultures in April 2017, the time of occurrence of the fourth case. The environmental cultures included samples from taps, sinks, drains, toilets, showers, door handles in wards, doctors' offices, treatment rooms and changing rooms. Work countertops, keyboards, computer mice, stethoscopes, and telephones were also included. The surfaces of pillows, sheets, callers, tables, drainage tubes, drainage bottles, and measuring cylinders in the rooms of patients were also sampled for culture. Blood agar plates (5% Sheep Blood) and China blue agar plates were used for environmental surveillance. However, there were no positive results in the first screening culture. A second environmental screening culture was performed in July 2017. A fish tank in the nurses station caught our attention. The second culture included samples from the decorative stones, filter gauze, inner walls and water in the fish tank.

Identification of isolates and antimicrobial susceptibility testing

The organisms in all the patient samples (n = 10) and tank samples (n = 2) were identified as *A. hydrophila/caviae* using a biochemical phenotypic identification system (Vitek 2 Compact, bioMérieux, Marcy l'Etoile, France). The organisms were identified as *A. hydrophila* using matrix-assisted laser desorption ionization mass spectrometry-time of flight (MALDI-TOF MS). DNA sequencing analysis was performed using three pairs of primers, including 16S rRNA, *gyrB* and *rpoB*, as described previously [14, 15] to further confirm the identity of the isolates. Antibiotic susceptibility tests were conducted for 12 isolates using the Vitek 2 Compact system and the disk diffusion method. The Clinical and Laboratory Standards Institute (CLSI) M45-A3 criteria were used to define susceptibility and resistance to the antibiotics tested [16].

Pulsed-field gel electrophoresis (PFGE)

Pulsed-field gel electrophoresis (PFGE) was conducted according to the standardized PulseNet PFGE protocol for pathogenic gram-negative enteric bacteria [17] with a slight modification. Briefly, whole-cell genomic DNA from lysed cell cultures of each isolate embedded in 1% agarose plugs (Bio-Rad, Richmond, CA, USA) were digested with the restriction enzyme XbaI (New England Biolabs, Ipswich, MA) and separated by electrophoresis through 1% pulsed-field-certified agarose (Bio-Rad) using a CHEF-Mapper instrument. Electrophoretic switch times of 4–40 s were used with a 6 V/cm current and a switch angle of 120° at a constant temperature of 14 °C. PFGE patterns were interpreted using the criteria proposed by Tenover et al [18]. An isolate is designated genetically indistinguishable if its restriction pattern has the same numbers of bands and the corresponding bands are the same apparent size; An isolate is considered unrelated to the outbreak strain if there are seven or more band differences between the outbreak pattern and that of the test isolate.

Detection of virulence genes and multilocus sequence typing (MLST)

Polymerase chain reactions (PCRs) using previously described primers and conditions were conducted to detect the virulence genes encoding heat-stable enterotoxin (*ast*), haemolysin (*ahh1*), hemolysin (*asa1*), cytotoxic enterotoxin (*act*), enolase (*eno*) and components of the type III secretion system (*ascV* and *aexT*) [19]. Six housekeeping genes (*gyrB*, *groL*, *gltA*, *metG*, *ppsA* and *recA*) were chosen for the MLST analysis according to the method previously reported by Martino. Amplified PCR products corresponding to the expected sizes were sequenced as previously described [19]. Each unique allelic profile, as defined by the allele numbers of the 6 loci, was assigned a sequence type (ST) number. New nucleotide sequences were deposited in the *Aeromonas* MLST database (<http://pubmlst.org/aeromonas>).

Whole genome sequencing (WGS) and genome assembly

Resistance to aztreonam and ceftazidime of one *A. hydrophila* isolate collected from patient 4 developed after one week of empirical therapy with ceftriaxone. WGS of the latter resistant *A. hydrophila* isolate 5 (AE456) was performed using an Illumina NovaSeq PE150. Illumina PCR adapter reads and low quality reads from the paired-end were filtered by the step of quality control using Readfq (vision 10). All good quality paired reads were assembled using the SOAP denovo (<http://soap.genomics.org.cn/soapdenovo.html>) and ABySS into a number of scaffolds. We used the ARDB (Antibiotic Resistance Genes Database) to perform the antibiotic resistant genes analyses.

Results

Epidemiological findings

Among the 34 patients with positive *A. hydrophila* cultures isolated in the microbiological laboratory since 2008, eight were current outbreak cases with postoperative SSIs, five had skin and soft-tissue infections, two had diarrhea, nine had a hepatobiliary infection, five had a urinary infection, and five were colonized. Eight case patients were all hospitalized in orthopaedic ward 2.

The *A. hydrophila* infection appeared to be intermittent, as shown in Fig. 1. From June 2016 to July 2017, i.e., from the first case to the last case recorded eight patients tested positive for *A. hydrophila* in fluids from the surgical sites or blood cultures, and the clinical characteristics of these patients are shown in Table 1. Three patients were male (37.5%), and five were female; the median age of the patients was 60 years (range, 42 to 68 years). The case patients had been admitted for various underlying medical conditions, including thoracic spinal stenosis (n = 2), scoliosis (n = 1), lumbar spinal stenosis (n = 4), and a giant cell tumor of the thoracic vertebra (n = 1). The types of surgery included laminectomy or tumor excision, decompression, bone graft fusion and internal fixation. Drainage tubes were placed in the surgical site after the operations, and intravenous cefuroxime was used to prevent infection. SSIs occurred 3–6 days after operation, 0.5–13 hours after removal of drainage tubes. The clinical manifestations included fever (n = 6), purulent drainage fluid (n = 3), pain in the wounds (n = 2), and hypotension and oliguria (n = 1). Six patients underwent wound debridement and irrigation. Two or three drainage tubes were placed in

the wounds for continuous irrigation-suction with saline and hydrogen peroxide. All the patients received imipenem or ertapenem after obtaining the antimicrobial susceptibility test results of *A. hydrophila*. All patients recovered and were subsequently discharged from the hospital after 11–59 days (median, 27 days).

Table 1
Characteristics of patients with *Aeromonas hydrophila* infection after spinal surgery

Patient	Age (y), gender	Diagnosis at admission	Date of Surgery	Date of removal of drainage tubes	Clinical manifestation	Time from Surgery to infection (days)	Time from removal of drainage tubes to infection (hours)	Positive cultures for <i>A. hydrophila</i>	Empirical antibiotic therapy	Antibiotic therapy after culture	Surgical debrider
Patient 1	61, M	Thoracic spinal stenosis	2016/6/2	NA	Wound pain, fever, low blood pressure	3	NA	Drainage, tissue	SCF, IPM, MZ	IPM	Yes
Patient 2	61, F	Scoliosis	2016/7/7	2016/7/12	Purulent drainage	5	5	Drainage	SCF, VA	IPM	No
Patient 3	56, M	Lumbar spinal stenosis	2017/3/31	2016/4/5	Purulent drainage, fever	5	0.5	Drainage	No	ETP	No
Patient 4	68, F	Thoracic spinal stenosis	2017/4/6	2016/4/10	Fever, chill	5	13	Drainage, blood	SCF, CRO	IPM	Yes
Patient 5	50, F	Giant cell tumor of the thoracic spine	2017/4/13	NA	Purulent drainage	4	NA	Drainage	No	IPM	Yes
Patient 6	42, F	Lumbar spinal stenosis	2017/4/6	2017/4/10	Fever, wound pain	4	5	Drainage	IPM	IPM	Yes
Patient 7	68, F	Lumbar spinal stenosis	2017/5/12	2017/5/17	Fever	5	6	Drainage	SCF, VA	IPM	Yes
Patient 8	59, M	Lumbar spinal stenosis	2017/7/20	2017/7/24	Fever	5	10	Drainage, Blood	SCF	IPM	Yes

M, male; F, female; NA, not applicable; SCF, cefoperazone sodium and sulbactam; IPM, imipenem; MZ, metronidazole; VA, vancomycin; CRO, ceftriaxone; ETP, ertapenem.

Laboratory findings

Aeromonas species were cultured from the water ($> 10^5$ CFU/ml) and stone surfaces in the fish tank, including *A. hydrophila*, *A. veronii* and *Aeromonas sobria*. No other bacteria were detected. Colonies of *A. hydrophila* isolated from the fish tank were light green, but those from patients were not. DNA sequencing showed that the 10 isolates from patients and 2 isolates from the fish tank were most closely related to *A. hydrophila* ($> 99\%$ homology for three fragments, namely, the 16S rRNA, *gyrB* and *rpoB* gene sequences).

Ten strains of *A. hydrophila* collected from the patients were resistant to cefuroxime, cefoxitin, ceftriaxone, and trimethoprim-sulfamethoxazole and susceptible to cefepime, imipenem, meropenem, and amikacin, as shown in Table 2. Two out of the 10 strains were resistant to ceftazidime, ciprofloxacin and levofloxacin. The antibiotic susceptibility patterns of the 2 isolates collected from the fish tank were similar to those from patients, except the susceptibility to cefoxitin, cefuroxime and ceftriaxone.

Table 2
In vitro susceptibilities of 12 strains of *A. hydrophila* determined by a Vitek 2 Compact system

Isolate	Source	Samples	MIC ($\mu\text{g/ml}$)													
			CN	AK	CIP	LEV	ATM	FOX	CRO	CXM	CAZ	FEP	TZP	IPM	MEM	SXT
Isolate 1	Patient 1	Drainage	4	≤ 2	1	1	≤ 1	≥ 64	32	≥ 64	4	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 2	Patient 2	Drainage	≤ 2	≤ 2	1	1	≤ 1	≥ 64	32	≥ 64	2	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 3	Patient 3	Drainage	8	≤ 2	2	1	≤ 1	≥ 64	32	≥ 64	4	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 4 ^a	Patient 4	Blood	8	≤ 2	2	1	≤ 1	≥ 64	32	≥ 64	4	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 5 ^a	Patient 4	Drainage	4	≤ 2	≥ 4	≥ 8	≥ 64	≥ 64	≥ 64	≥ 64	≥ 64	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 6	Patient 5	Drainage	8	≤ 2	2	1	≤ 1	≥ 64	32	≥ 64	4	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 7	Patient 6	Drainage	4	≤ 2	2	1	≤ 1	≥ 64	32	≥ 64	4	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 8	Patient 7	Drainage	4	≤ 2	≥ 4	≥ 8	4	≥ 64	≥ 64	≥ 64	≥ 64	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 9	Patient 8	Drainage	8	≤ 2	1	1	≤ 1	≥ 64	32	≥ 64	4	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 10	Patient 8	Blood	4	≤ 2	1	1	≤ 1	≥ 64	16	≥ 64	2	≤ 1	≤ 4	≤ 1	≤ 0.25	$\geq 16/304$
Isolate 11	Fish tank	Water	≥ 32	≤ 2	0.5	0.5	≤ 1	4	≤ 0.25	4	0.5	≤ 1	≤ 4	0.5	≤ 0.25	$\geq 16/304$
Isolate 12	Fish tank	Stone	≥ 32	≤ 2	0.5	0.5	≤ 1	4	≤ 0.25	4	0.5	≤ 0.12	≤ 4	0.5	≤ 0.25	$\geq 16/304$

^aIsolates 4 and 5 were identified from blood and drainage samples from one patient, respectively, at intervals of one week.

CN, gentamicin; AK, amikacin; CIP, ciprofloxacin; LEV, levofloxacin; ATM, aztreonam; FOX, ceftioxin; CRO, ceftriaxone; CXM, cefuroxime; CAZ, ceftazidime; FEP, cefepime; TZP, piperacillin-tazobactam; IPM, imipenem; MEM, meropenem; SXT, trimethoprim-sulfamethoxazole.

PFGE analysis of the 12 strains revealed two banding patterns (designated as PFGE A and B), as shown in Fig. 2. Ten isolates from eight patients (2 from blood samples and, 8 from drainage samples) shared the same PFGE pattern, suggesting a clonally related origin. An exact match of PFGE patterns between *A. hydrophila* strains from the fish tank (PFGE type B) and *A. hydrophila* strains isolated from infected patients (PFGE type A) could not be established.

Two STs and 10 alleles were newly identified in the PubMLST database. Analysis of MLST profiles revealed that the 10 isolates from 8 patients belonged to ST517, and the two isolates from fish tank belonged to ST518. The clinical isolates harbored four virulence genes: *ahh1*, *eno*, *ast*, and *ascV*. *A. hydrophila* isolates from the fish tank carried less virulence genes, only *ahh1* and *eno* genes.

Assembly and annotation of the draft genome of isolate AE456 resulted in 5,062,684-bp and revealed resistance genes encoding AmpC-like β -lactamases (*cepS*), metallo-beta-lactamase (*cphA*) and oxacillinase (*bla_{OXA-12}*). Resistance genes to sulphonamides (*sul1*), macrolide (*mphA*) and tetracycline [*tet(E)*] have been identified. Class 1 integron related integrase (*intI1*) gene with class 1 integron gene cassette (*dfrA12-aadA2*, 2000 bp) have also been identified. No plasmid was found.

Discussion

Aeromonas species are common inhabitants of fresh and brackish water. These species have also been recovered from chlorinated tap water, including hospital water supplies. Esteban et al. reported a pseudo-outbreak of *A. hydrophila* isolates from endoscopies without related symptoms [20]. Nosocomial outbreaks of *A. hydrophila* have rarely been reported. The first outbreak *A. hydrophila* infection in a hospital was reported in 1984, in which 3 patients with SSI and 3 patients with pneumonia. *A. hydrophila* was isolated from a tank supplying the sluices in theatres, but the isolate was present in low numbers, so it was difficult to identify this tank as the source [21].

SSI after spinal surgery is one of the most serious complications and is usually caused by gram-positive cocci, such as *Streptococcus* and *Staphylococcus* species [22, 23]. To the best of our knowledge, this is the first published study of *A. hydrophila* SSI outbreak after spinal surgery. In the present study, we suspect that the current outbreak occurred due to the presence of *A. hydrophila* in the fish, which in turn contaminated the water in the tank. Such a scenario can be reasoned as follows. First, the water samples obtained from the fish tank grew a high number of colony-forming units of *Aeromonas* species including *A. hydrophila*, and no other bacteria were identified. Second, the surgeons, nurses in charge, operation rooms and implants differed among the case-patients.

Finally, the outbreak seemed to be under control from July 2017, after removal of the fish tank. Up to the end of December 2019, no new *A. hydrophila* isolates had been detected in our hospital.

It is very difficult to determine the source and transmission route of an outbreak, even if the isolates were identified from patients or the environment [24, 25]. There were 6 nursing workers who were the only people who had direct contact with the patients and the fish tank. These workers participated in the postoperative care of the patients and usually helped empty the drainage bags. These nursing workers also cleaned the fish tank and changed water. A large plastic box was used to hold the aseptic wound dressing packages in the ward. To throw the dead fishes away and change water, the nursing workers sometimes removed the dressing change packages and poured the water and dead fishes into the box temporarily. The packages were wrapped in cloth with a bending plate, gauze, tweezer and scissor. On the second day after the operation, the doctors disinfected the patients' wounds and replaced the dressing gauze with new gauze from the packages. Therefore, if the box had not been dried and sterilized after holding water and dead fishes, the dressing packages toward the bottom of the box would absorb the residual water and be contaminated.

Alternatively, direct transmission may also be caused by nursing workers lacking hand hygiene; when these workers poured the drainage fluid every day, the bacteria, via retrograde motion, may have entered the wound through the drainage tube. The reason for this speculation is that the skin of the patients' surgical incisions healed well, and a large amount of fluid was found under the muscular layer or subcutaneous tissue layer during debridement.

However, there remain many questions regarding the transmission hypothesis. First, a large number of *Aeromonas* species, including *A. hydrophila*, were isolated from the fish tank, but only *A. hydrophila* ST517 could be identified in patients. A plausible reason for this finding may be the different antimicrobial susceptibility patterns between the strains. The clinical *A. hydrophila* strains were resistant to cefuroxime used for infection prophylaxis after surgery, but *Aeromonas* species collected from the fish tank were sensitive to cefuroxime and may be uncultured strains. Second, an exact match of PFGE patterns and STs between *A. hydrophila* strains from the fish tank and *A. hydrophila* strains isolated from infected patients could not be established. This finding may be due to the wide diversity and large number of strains detected in the fish tank. It is difficult to identify pathogenic bacteria by culture and based on colony morphology.

The pathogenicity of *Aeromonas* species appears to be due to the ability of the bacteria to produce several virulence factors that are highly heterogeneously present among clinical isolates [26]. In this study, all the *A. hydrophila* isolates from patients harbored multiple virulence genes. *A. hydrophila* isolates from the fish tank carried less virulence genes tested than the clinical isolates. Antimicrobial resistance was more commonly observed among clinical isolates than among environmental isolates, as previously described [27]. Saavedra et al has demonstrated that extensive use of antimicrobial drugs for prophylaxis and treatment unquestionably plays a role in the increased number of resistant *A. hydrophila* strains [28]. Resistance to cefotaxime can develop during therapy [29, 30]. The clinical isolates in our study were all resistant to ceftriaxone and cefuroxime, used for infection prophylaxis. Resistance to aztreonam and ceftazidime of one *A. hydrophila* isolate (AE456) collected from patient 4 developed after one week of empirical therapy with ceftriaxone. WGS obtained from the latter resistant isolate (AE456) revealed that the isolate harbored Ambler molecular class B (cphA), class C (cepS) and class D (bla_{OXA-12}) β -lactamase genes. *A. hydrophila* can produce chromosomally mediated AmpC-like β -lactamases under the selective pressure of antibiotics, which can lead to resistance to cephalosporins [31]. It also intrinsically harbors a chromosome-mediated cphA gene which encodes a metallo-beta-lactamase (MBL) highly active against carbapenem antibiotics. The expression of CphA MBL is induced by β -lactams such as imipenem and emerging carbapenem resistance have also been reported in infections due to *Aeromonas* species carrying cphA [32–34]. Therefore, the use of β -lactams (except 4th-generation cephalosporins) should be cautious for clinical failure; therapy should be modified according to the in vitro susceptibility for individual causative isolates.

There are some limitations associated with this study. First, we could not determine the specific link between these outbreak cases and the fish tank. As a possible source of the outbreak, the fish tank remained a concern because a large amount of *A. hydrophila* was recovered from the tank. Another limitation of the investigation was that we did not use a selective culture medium with cefuroxime or ceftriaxone to identify the pathogen. In addition, the wound dressing bags were not sampled, which may have been vital sources associated with the outbreak. Several lessons can be learned from this outbreak. First, it is necessary to increase staff awareness of nosocomial outbreaks in patients undergoing surgery, especially of infections caused by uncommon organisms. Early identification and control could help stop outbreaks from spreading. Furthermore, nursing workers should be educated to follow infection prevention guidelines during wound care practices. It is also important that sterilized or aseptic instruments or devices be protected against bacterial contamination. Finally, there is a need to strengthen environmental surveillance and disinfection to eliminate potentially harmful microorganisms, especially in surgical areas and wards.

Conclusions

Aeromonas hydrophila is rare, but severe, pathogen in surgical infections. Strict measures, including environmental disinfection and contact precautions, are needed to prevent post-operative infection outbreak.

Abbreviations

SSIs: Surgical site infections; PFGE: Pulsed-field gel electrophoresis; MLST: Multilocus sequence typing; WGS: Whole genome sequencing; MRI: Magnetic Resonance Imaging; MALDI-TOF MS: Matrix-assisted laser desorption ionization mass spectrometry-time of flight; CLSI: Clinical and Laboratory Standards Institute; PCR: Polymerase chain reactions; ST: sequence type; MBL: metallo-beta-lactamase

Declarations

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Authors' contributions

XX, YJ, LC and WL conceived the project, XX, XY and JZ did laboratory work and data analysis, YJ, ZG, QQ, YZ, CS and ZC collect the clinical data. XX and YJ are the major contributors in writing the manuscript. All authors read and approved the final manuscript.

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Availability of data and materials

The datasets are available from the corresponding author under reasonable request.

Ethics approval and consent to participate

Permission to use the information in the medical records of the patients and the *A. hydrophila* isolates for research purposes was given by the Ethics Committee of Peking University Third Hospital. Based on

the approval of Ethics Committee, all informations were collected only by patient's code and their identity was not disclosed. Patient's information was in private.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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Figures

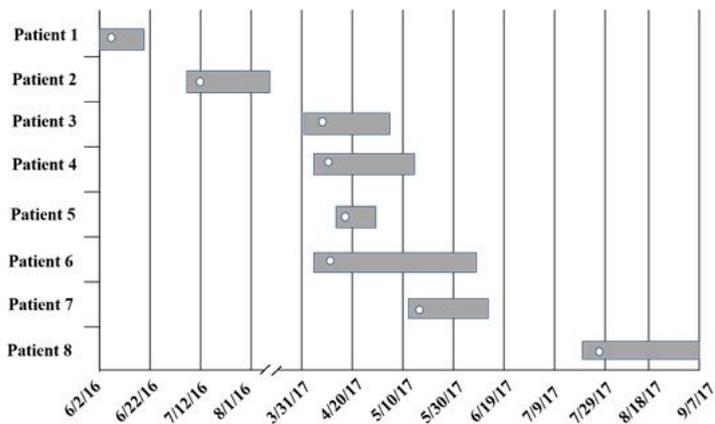


Figure 2
 Timeline for admission of outbreak patients with surgical site infections of *Aeromonas hydrophila* (N=8) to orthopaedic ward 2. Circles indicate onset of infection. Gray boxes indicate the period of hospitalization in orthopaedic ward.

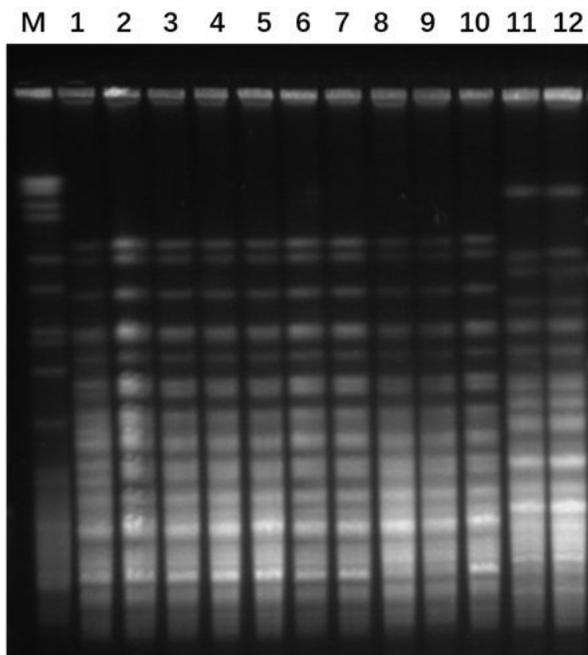


Figure 4
 PFGE separation of *Xba*I-digested chromosomal DNA from *Aeromonas hydrophila* obtained from June 2016 to July 2017 in Beijing, China. Lane M, molecular weight marker (*Salmonella* H9812); lanes 1 to 10, 10 clinical strains of *A. hydrophila* involved in this outbreak (belonging to clone A); lanes 11 and 12, 2 environmental strains of *A. hydrophila*, identified from the water and stones in the fish tank, respectively (designated clone B).